HEATHER ABNEY MERRIOTT O'NEAL

Effects of Chronic Activity Wheel Running or Antidepressant Pharmacotherapy on
     Appetitive Behavior and Gene Expression in Rat Locus Coeruleus after Olfactory
     Bulbectomy

(Under the Direction of ROD K. DISHMAN)

Evidence suggests that exercise can be effective for the prevention and treatment of depression. A meta-analysis was conducted to quantify effects from studies examining the effect of an exercise intervention on depression scores. The overall effect was moderate, $d = -0.57$ (95% CI, -0.79 to -0.34), suggesting that exercise groups exhibit a 1/2 SD reduction in depression relative to comparison groups.

However, biological mechanisms to explain such effects have not been established. Common antidepressant medications target the noradrenergic system, and exercise influences noradrenergic transmission. Thus, adaptations in central noradrenergic function represent a plausible mechanism to explain antidepressant effects of exercise. The first study examined the effect of 6 weeks of treadmill training on gene expression for prepro-galanin (GAL) and tyrosine hydroxylase (TH) within the locus coeruleus using *in situ* hybridization. Levels of GAL messenger RNA (mRNA) were higher in treadmill-trained compared to sedentary animals, but there was no effect on TH mRNA. These results support a potential role of GAL in modulating noradrenergic activity after chronic exercise.

The second study examined the effect of chronic activity wheel running and imipramine administration on appetitive behavior and gene expression in the locus coeruleus using the olfactory bulbectomy model of depression. Male rats were randomly assigned to the following conditions: (1) bilateral olfactory bulbectomy or sham surgery, (2) activity wheel running or sedentary home cage, and (3) daily imipramine or saline injections. After 21 days, animals underwent behavioral testing for sucrose preference and copulatory activity. Levels of TH, GAL, and prepro-neuropeptide Y (NPY) mRNA were measured using *in situ* hybridization. Bulbectomized animals exhibited reductions in sucrose intake and decrements in copulatory performance compared to sham animals. Imipramine administration was also associated with decreased copulatory rates and

ejaculation frequency. However, activity wheel running after bulbectomy improved copulation rates, reduced ejaculation latency, and increased ejaculation frequency. Level of GAL mRNA was increased after imipramine treatment whereas activity wheel running after bulbectomy increased gene expression for NPY. No differences were found in TH mRNA. Adaptations in noradrenergic function after 3 weeks of imipramine treatment and activity wheel running do not appear to occur via similar mechanisms of gene expression within the locus coeruleus.

INDEX WORDS: *In situ* hybridization, Exercise, Depression, Norepinephrine, Tyrosine hydroxylase, Galanin, Neuropeptide Y, Sexual behavior, Sucrose preference

EFFECTS OF CHRONIC ACTIVITY WHEEL RUNNING OR ANTIDEPRESSANT

PHARMACOTHERAPY ON APPETITIVE BEHAVIOR AND GENE EXPRESSION

IN RAT LOCUS COERULEUS AFTER OLFACTORY BULBECTOMY

by

HEATHER ABNEY MERRIOTT O'NEAL

B.S., Louisiana Tech University, 1993

M.A., Louisiana Tech University, 1995

A Dissertation Submitted to the Graduate Faculty of the University of Georgia in Partial

Fulfillment of the Requirements for the Degree

DOCTOR OF PHILOSOPHY

ATHENS, GEORGIA

2000

EFFECTS OF CHRONIC ACTIVITY WHEEL RUNNING OR ANTIDEPRESSANT

PHARMACOTHERAPY ON APPETITIVE BEHAVIOR AND GENE EXPRESSION

IN RAT LOCUS COERULEUS AFTER OLFACTORY BULBECTOMY

by

HEATHER ABNEY MERRIOTT O'NEAL

Approved:

Major Professor: Rod K. Dishman

Committee: Kirk J. Cureton
                     Philip V. Holmes
                     Patrick J. O'Connor
                     Amos Zeichner

Electronic Version Approved:

Gordhan L. Patel
Dean of the Graduate School
The University of Georgia
August 2000

ACKNOWLEDGMENTS

Trust in the Lord with all your heart
and lean not on your own understanding;
in all your ways acknowledge Him,
and He will make your paths straight.
Proverbs 3:5-6

First and foremost, I would like to express my deepest gratitude to my family for their encouragement, love, and support; but most of all, for providing my most valuable education. By example, they have taught me what is truly important – faith in God, love of family, and fellowship with friends.

I also extend my sincere appreciation to Dr. Rod Dishman for his direction and support during these last four years. His guidance has enabled me to develop my talents and pursue my interests while challenging me to excel both as a student and scientist. As a mentor, he has taught me the importance of scholarship in all endeavors, and his sincere concern for my welfare and success will always be remembered.

I would also like to thank the members of my committee, Dr. Kirk Cureton, Dr. Phil Holmes, Dr. Pat O'Connor, and Dr. Amos Zeichner, for imparting knowledge and guidance through teaching and research. In particular, I would like to recognize Dr. O'Connor for his continual encouragement. A special thanks also goes to Dr. Holmes for his generosity in providing his laboratory and equipment as well as the technical expertise that made my research possible.

I wish to express my gratitude and best wishes to fellow graduate students from the Exercise Psychology Laboratory who have been an invaluable source of advice, assistance, and amusement: Dane Cook, J.B. Crabbe, Erica Jackson, Suzi Hong, Rob Motl, Pat O'Brien, and Carson Smith. In particular, I extend a heartfelt thank you to Jacquie Van Hoomissen for her research assistance, intellectual contributions, and

iv

steadfast friendship. I'm also appreciative of the many individuals who facilitated various research projects that I have been involved with at the University of Georgia: Allison Barton, Carrie Brady, Dr. Bradford Bunnell, Dr. Linda Campbell, Camillia Clark, Jessica Dishman, Alicia Duncan, Dr. Andrea Dunn, Josh Garrett, Sandy Hively, Dr. Andrea Hohmann, Robin Kavanaugh, Philip MacArthur, Femke O'Brien, Stefany Primeaux, Dr. Jesus Soares, and Marlee Stewart.

I also extend my gratitude to the numerous individuals who have contributed to my education through the years. I would like to acknowledge the Departments of Biological Sciences and Behavioral Sciences at Louisiana Tech University. In particular, I'd like to recognize Dr. James Liberatos, Dr. Jim Spaulding and Dr. Mark Miller who were excellent mentors during my study toward undergraduate and master's degrees as well as two special science teachers, Donna White and Mary Jo Newell, who inspired my early pursuit of science. Finally, I extend my earnest appreciation to the University of Georgia Department of Exercise Science for establishing a distinguished standard of excellence in both academics and research.

On a personal note, I'd like to thank Kent Reynolds, St. James UMC, Ray Crenshaw, and the O'Neil family for helping to make Georgia feel like home. I also extend my love and appreciation to friends who have supported me throughout this endeavor and reminded me that the journey is more important than the destination… Marnie, Julie, Jacquie, Mark, Darby, John, Elise, Keith, Ward… your friendship has truly been a blessing!

This work is dedicated to my family whose unfailing love and generous support have enabled me to pursue my goals. Thank you for always believing in me.
You truly are my inspiration.
My prayers and love are with you always!

TABLE OF CONTENTS

<div align="right">Page</div>

INTRODUCTION AND LITERATURE REVIEW

As too much and violent exercise offends on the one side, so doth an idle life on the other…
Opposite to Exercise is Idleness or want of exercise, the bane of body and mind… the chiefe
author of all mischiefe, one of the seven deadly sinnes, and a sole cause of Melancholy.
~Burton, 1632

Evidence from anecdotal reports, epidemiological research, and intervention studies suggests that exercise may be an effective intervention for the prevention and alleviation of depressive symptoms for many individuals with mild to moderate depression (O'Neal, Dunn, & Martinsen, 2000). In addition, exercise training in animals influences neurobiological systems involved in the etiology of depressive disorders (Dishman, 1997). However, few studies have used an animal model of depression to investigate the effect of physical activity on biological indices of depression (Dishman, Renner, Youngstedt, Reigle, Bunnell, Burke, Yoo, Mougey, & Meyerhoff, 1997; Yoo, Tackett, Crabbe, Bunnell, & Dishman, 2000). To my knowledge, chronic exercise has not been implemented as an antidepressant treatment in the olfactory bulbectomy animal model of depression, which has predictive validity for pharmacological interventions and induces alterations in neurobiological systems that are influenced by physical activity (Kelly, Wrynn, & Leonard, 1997).

Depression: A Public Health Concern

Unipolar depressive disorders, including major depression and dysthymia, are prevalent psychiatric disorders in modern society. Depression is a debilitating psychological condition characterized by disturbances in cognitive, physiological, and emotional functioning. In research and clinical settings, criteria for depressive disorders are commonly defined using *Research Diagnostic Criteria* (RDC), *International*

Classification of Diseases (ICD), or *Diagnostic and Statistical Manual of Mental Disorders* (DSM-IV). For example, DSM-IV criteria for major depression include the presence of at least one of the following symptoms almost daily for at least 2 weeks: 1) depressed mood most of the day, nearly every day or 2) reduced or loss of interest or pleasure in almost all activities. Five of the following symptoms must be also be exhibited: significant weight loss or gain without dieting, insomnia or hypersomnia, feelings of lethargy or restlessness, fatigue or loss of energy, psychomotor retardation or agitation, feelings of worthlessness or excessive guilt, reduced ability to think or concentrate, and recurrent thoughts of death or suicide. Symptoms of dysthymia resemble those of major depression but symptoms are less severe and the condition is more chronic with depressed mood persisting for at least 2 years (APA, 1994).

Depression is a widespread public health problem that poses a significant burden on society in terms of cost, loss of productivity, and mortality in addition to the detrimental effects on quality of life (Greenberg, Stiglin, Finkelstein, & Berndt, 1993). Major depression has been identified as the most common psychiatric disorder in the United States. A national survey determined that 17% of respondents experienced a lifetime history of major depressive episode, and 10% reported a depressive episode within the past year (Kessler, McGonagle, Zhao, Nelson, Hughes, Eshleman, Wittchen, & Kendler, 1994). The financial burden of depression in the United States has been estimated to be $44 billion annually including $12 billion in direct health care costs and $31 billion in indirect costs due to reduced productivity, employee absenteeism, and premature death. An estimated 15,000 men and 3,400 women committed suicide as a result of depression in 1990 (Greenberg et al., 1993), and depression increases risk of cardiac mortality after myocardial infarction (Frasure-Smith, Lesperance, Juneau, Talajic, & Bourassa, 1999). In addition, depression is associated with increased morbidity including heightened risk for coronary heart disease (Ferketich, Schwartzbaum, Frid, & Moeschberger, 2000) and exacerbation of existing medical conditions and disabilities. Accordingly, medical patients with depression have

longer average hospital stays and illness duration (Greenberg et al., 1993), and individuals with depression are at increased risk for developing a physical disability of daily living and mobility (Penninx, Leveille, Ferrucci, van Eijk, & Guralnik, 1999). In terms of life years lost to disability and premature mortality, major depression was ranked as the fourth leading international disease burden in 1990 according to the Global Burden of Disease Study and is predicted to rise to second by the year 2020 (Murray & Lopez, 1996).

Despite the prevalence of depression, it is estimated that less than 20% of individuals with a recent depressive disorder seeks professional treatment within the year of initial onset (Kessler et al., 1994). In addition, approximately 25% of individuals diagnosed with major depression remain depressed after one year (Sargeant, Bruce, Florio, & Weissman, 1990). The prevalence of depression in the population, the financial burden to society, the adverse effects on quality of life, and the ineffectiveness and/or lack of utilization of traditional antidepressant therapies underscore the need to examine other treatments such as exercise that may be effective in alleviating depression.

The most common forms of treatment for depression include psychotherapy and antidepressant medication. Although many individuals find relief from depressive symptoms in traditional treatment programs, neither psychotherapy nor medication is effective in alleviating depression in all individuals. The National Depressive and Manic-Depressive Association reports that depressive disorders are widely undertreated with estimates suggesting that only 10% of depressed individuals receive adequate treatment (Hirschfeld et al., 1997). Additionally, many people fail to adhere to traditional treatment programs for various reasons. Psychotherapy is often costly and time-intensive, and persons with depression may be reluctant to seek counseling due to social stigma or other concerns (Greenberg et al., 1993). Antidepressant medications, if prescribed in the appropriate dose and taken for the prescribed length of time, can be expensive and may produce undesirable side effects (Hirschfeld, 1998). Thus, the study

of additional treatment modalities such as exercise may present an affordable, accessible, and healthy option to complement traditional antidepressant therapies.

Evidence for the Antidepressant Effect of Exercise

Evidence for the antidepressant effect of exercise has consisted of epidemiological research, including cross-sectional and longitudinal studies, as well as experimental and clinical studies of exercise interventions for both depressed and mentally-healthy individuals. Population-based studies provide the first line of evidence that physical activity is associated with a reduction in depression (e.g., Camacho, Roberts, Lazarus, Kaplan, & Cohen, 1991; Krause, Goldenhar, Liang, Jay, & Maeda, 1993; Farmer, Locke, Moscicki, Dannenberg, Larson, & Radloff, 1988; Paffenbarger, Lee, & Leunge, 1994; Rajala, Uusimaki, Keinanen-Kiukaanniemi, & Kivela, 1994; Weyerer, 1992). Results from quantitative reviews of intervention studies also support an antidepressant effect of exercise. North, McCullagh, & Tran (1990) conducted a meta-analysis of 80 studies published through June 1, 1989 that examined the effect of exercise on depression. The overall effect size (d) was –0.53 indicating that, on average, individuals in exercise conditions exhibited a 1/2 SD greater reduction in depression scores than individuals in comparison groups. Similar results were obtained from a recent meta-analysis by Craft & Landers (1998) that examined the effect of exercise interventions in persons with clinically diagnosed depressive disorders. A larger overall effect size (d) of -0.72 was obtained, indicating that exercise participation resulted in a 3/4 SD greater reduction in depression scores relative to individuals in control groups.

Evidence from experimental research suggests that chronic physical activity including aerobic and resistance exercise may produce meaningful reductions in depressive symptoms in individuals with mild to moderate depression, and continued exercise may help prevent recurrence of depressive symptoms. Over twenty years have passed since experimental studies first demonstrated antidepressant effects of exercise in individuals with a clinical diagnosis of depression. Greist, Klein, Eischens, Faris,

Gurman, & Morgan, (1978) examined the effect of ten weeks of walking/running treatment on symptoms of depression in 24 men and women diagnosed with minor depression. Exercisers exhibited significant reductions in depressive symptoms, and the treatment response to exercise was similar to that of psychotherapy. Moreover, exercisers maintained improvements during the 1-month follow-up period. Martinsen, Medhus, & Sandvik (1985) also examined the effect of aerobic exercise training on depression in 43 hospitalized psychiatric patients diagnosed with clinically defined depressive disorders. Patients were randomly assigned to either exercise or an occupational therapy control group in addition to their standard treatment that included psychotherapy and pharmacotherapy. After 9 weeks of training, patients in the exercise group exhibited significantly larger reductions in self-reported symptoms of depression relative to the control group. Patients were also followed one to two years after discharge from the hospital. Most subjects reported continued exercise participation, and exercisers tended to exhibit lower depression scores compared to nonexercisers at the follow-up assessment (Martinsen & Medhus, 1989).

Resistance exercise training interventions have also been effective in alleviating symptoms of depression. For example, Doyne, Ossip-Klein, Bowman, Osborn, McDougall-Wilson, & Neimeyer (1987) compared the effectiveness of aerobic exercise (running) and weight lifting as an antidepressant intervention in 40 women diagnosed with depression according to RDC criteria. Both exercise groups exhibited significant reductions in depression scores after 8 weeks of training compared to the wait-list control condition. Similar results were obtained in a study conducted by Martinsen, Hoffart, & Solberg (1989) in which psychiatric inpatients meeting DSM-III-R criteria for unipolar depressive disorders (major depression, dysthymia, and atypical depression) were randomly assigned to either an aerobic or nonaerobic exercise group. After eight weeks of training, both groups exhibited significant reductions in depression scores. However,

the change in depression scores did not differ between the two conditions although only the aerobic group exhibited significant increases in maximal oxygen uptake.

Despite abundant evidence that supports an antidepressant effect of physical activity, prominent mental health organizations have yet to recognize exercise as a viable alternative or adjunctive treatment for depression. Thus, randomized controlled trials comparing exercise interventions to standard antidepressant therapies are needed. Perhaps the most convincing evidence for the effectiveness of exercise in the treatment of depression emerged from a recent clinical trial that compared exercise and antidepressant medication (Blumenthal et al., 1999). Approximately 150 older patients diagnosed with major depression were randomly assigned to one of 3 groups: aerobic exercise (walking/jogging, 3 times/week, 30 minutes), antidepressant medication (sertraline hydrochloride, i.e., Zoloft), or exercise plus medication. Though individuals in the medication group exhibited the fastest initial response to treatment, all groups exhibited significant reductions in depression scores. Moreover, there were no significant differences between groups after the 16-week intervention period.

Although many studies in human populations support an antidepressant effect of exercise, the evidence for a plausible mechanism to explain observed effects is lacking. Numerous hypotheses have been proposed to explain improvements in depression after exercise training, including enhanced social support, improved self-efficacy, elevated self-esteem, and distraction from cares and worries. However, the potential for exercise to make a unique contribution to mental health beyond the benefits that can be achieved with traditional psychotherapy lies in biological alterations in brain function resulting from physical activity (Dishman, 1998).

Neurobiology of Depression

Although numerous theories have been proposed to explain the etiology and maintenance of depressive disorders including social, cognitive, and psychodynamic theories, abundant scientific evidence suggests a substantial biological component in the

development, symptomatology, and treatment of depression (Charney, 1998). Of particular importance are central monoaminergic neurotransmitter systems (i.e., norepinephrine, serotonin, and dopamine) that regulate various emotional and stress responses. Although continued research is needed to gain a better understanding of the complex interaction among these neural systems, prominent hypotheses for the development and maintenance of depression involve dysregulation of central monoaminergic neurotransmission.

Norepinephrine

Much of the research examining the biological basis of depression has focused on the central noradrenergic system (Charney, 1998). Noradrenergic neurons project extensively throughout the brain and arise from brain stem cell bodies. Of particular importance in the study of affective disorders is the locus coeruleus complex, a small region in the brain stem that projects to numerous brain regions including areas associated with the regulation of emotion (Dahlstrom & Fuxe, 1964; Foote, Bloom, & Aston-Jones, 1983). The LC-NE system is a diffuse modulatory network that regulates attention, vigilance, and arousal in response to stressful stimuli (Aston-Jones, Rajkowski, & Cohen, 1999).

Norepinephrine (NE) is synthesized in noradrenergic neurons from tyrosine removed from blood. Within the neuron, tyrosine is converted to dihydroxy-phenylalanine (DOPA) by tyrosine hydroxylase, the rate-limiting enzyme for norepinephrine synthesis. DOPA is then converted to dopamine by amino acid decarboxylase, and dopamine β-hyrdoxylase converts dopamine to norepinephrine. Postsynaptic receptors for NE include α_1, α_2, β_1, and β_2 subtypes; however, the presence of receptors may vary by brain region. Binding of NE to α_2 receptors decreases noradrenergic transmission, and α_2 autoreceptors are also found presynaptically for feedback inhibition of NE synthesis and release. Alpha- and β-adrenergic receptors are coupled to G-proteins, and the effects of NE are mediated by second messenger systems.

NE is metabolized by monoamine oxidase (MAO) to produce 3,4 dihydroxy-phenylglycol (DHPG) within the neuron and 3-methoxy-4-hydroxy-phenylglycol (MHPG) in the synapse. Thus, the noradrenergic system can be studied by measuring neurotransmitters, enzymes, receptors, and/or metabolites. Proposed theories for involvement of the noradrenergic system in depression include decreased synthesis and/or release from presynaptic neurons, increased activity of presynaptic α_2 receptors, and reduced postsynaptic receptor or 2nd messenger system activity (Schatzberg, 1998).

Evidence for the role of norepinephrine in the etiology of depression arose from observations in the 1950's that patients with hypertension developed depression when they were given reserpine, a drug that depletes monamines. In addition, studies conducted in the 1960's and 1970's found that depressed individuals often exhibit decreased levels of NE metabolites in cerebrospinal fluid and urine compared to normal individuals (Schildkraut, 1965), and post-mortem examination has revealed upregulation of cortical β-receptors in some suicide victims (Cheetham, Katona, & Horton, 1991). Additional support of norepinephrine involvement in depression is obtained from pharmacological studies that demonstrate alterations in noradrenergic transmission upon treatment with antidepressant agents. The primary action of many commonly prescribed antidepressant medications is to increase the level of NE at the synapse by influencing reuptake, metabolism, or release of NE. For example, MAO inhibitors (MAOIs) exert their effect by preventing the breakdown of NE whereas tricyclic antidepressants (TCAs) prevent reuptake of NE by blocking presynaptic receptors (Charney, 1998).

Norepinephrine and Peptide Neurotransmitters

Neuropeptides such as galanin and neuropeptide Y that modulate noradrenergic activity may also be involved in depressive disorders. Galanin is a 29-amino acid peptide neurotransmitter that coexists with norepinephrine in approximately 80% of LC neurons (Holets, Hokfelt, Rokaeus, Terenius, & Goldstein, 1988). Galanin-positive neurons within the LC tend to be dendrodendritically arranged, and galanin hyperpolarizes noradrenergic

neurons to inhibit locus coeruleus firing *in vitro* (Seutin, Verbanck, Massote, & Dresse, 1989; Pieribone, Xu, Zhang, Grillner, Bartfai, & Hokfelt, 1995). Thus, it is plausible that upregulation of galanin within the LC may function as a negative feedback mechanism for inhibition of NE release (Holmes & Crawley, 1995). Though it has been proposed that increased galanin activity could result in deleterious changes in brain function with chronic stress (Weiss, Bonsall, Demetrikopoulos, Emery, & West, 1998), it is equally plausible that alterations in galanin within the LC play an adaptive role by regulating responsiveness of the noradrenergic system to stress.

Neuropeptide Y is a 36-amino acid neurotransmitter that is colocalized with NE in approximately 40% of LC neurons (Holets et al., 1988) and inhibits LC firing in vitro (Finta, Regenold, & Illes, 1992). NPY-immunoreactive neurons project to various brain structures involved in the regulation of mood and emotion including the hypothalamus, nucleus accumbens, amygdala, hippocampus, and cortex (Heilig & Widerlov, 1990). Furthermore, decreased levels of NPY have been observed in patients with major depression (Hashimoto, Onishi, Koide, Kai, & Yamagami, 1996), and alterations in cortical and hypothalamic NPY activity have been found after antidepressant administration in animals (Baker, Herkenham, & Brady, 1996; Smialowska & Legutko, 1991). In addition, gene expression for NPY in limbic regions including the nucleus accumbens and hippocampus is reduced in a genetic animal model of depression but increased after treatment with the antidepressant, fluoxetine (Caberlotto, Fuxe, Overstreet, Gerrard, & Hurd, 1998). Thus, it has been hypothesized that the pathology associated with depression might involve a deficiency of central NPY, and pharmacological interventions for depression could act by modulating NPY activity (Heilig & Widerlov, 1990).

Serotonin

A second monoamine neurotransmitter thought to be involved in the etiology of depression is serotonin (5-HT). The majority of 5-HT neurons originate from the raphe

nuclei located on the midline of the brain stem and project extensively to various structures throughout the brain including the olfactory bulb, hypothalamus, thalamus, amygdala, hippocampus, cortex, and locus coeruleus (Steinbusch, 1981). Thus, the central 5-HT system modulates activity of diverse brain systems that regulate autonomic functions, motor control, cognition, nociception, arousal, and emotion (Halliday, Harding, & Paxinos, 1995).

Serotonin (5-hydroxytryptamine or 5-HT) is synthesized from tryptophan, a dietary amino acid that enters serotonergic neurons from circulating blood. Tryptophan is converted to 5-hydroxytryptophan (5-HTP) by the enzymatic action of tryptophan hydroxylase and then to 5-HT by amino acid decarboxylase. Synthesis of brain 5-HT is limited by the availability of tryptophan in the blood. Upon release, serotonergic activity is terminated by presynaptic reuptake by 5-HT transporters, activation of 5-HT$_{1A}$ autoreceptors, or metabolism by MAO to 5-hydroxy indoleacetic acid (5-HIAA). Serotonergic activity is mediated by pre- and post-synaptic receptors, and at least 15 types of 5-HT receptors with various molecular and pharmacological properties have been identified (Murphy, Andrews, Wichems, Li, Tohda, & Greenberg, 1998).

Convincing evidence for the role of 5-HT in depression is obtained from the pharmacological research demonstrating the efficacy of serotonergic drugs in alleviating depressive symptoms. The most widely prescribed antidepressant medications are selective serotonin reuptake inhibitors (SSRIs) that specifically target the 5-HT system by blocking presynaptic 5-HT reuptake and allowing the neurotransmitter to remain in the synapse (DeVane, 1998). In addition, symptoms of depression including disturbances in sleep, appetite, locomotion, sexual activity, cognition, and mood are regulated by structures receiving serotonergic projections (Maes & Meltzer, 1995), and there is evidence suggesting that individuals exhibiting certain alterations in the serotonin transporter gene are at increased risk for developing major depression (Ogilvie, Battersby, Bubb, Fink, Harmar, Goodwin, & Smith, 1996).

Serotonergic systems appear to be dysregulated in depressed patients; however, the mechanisms for 5-HT modulation of depression have not been established. Although reductions in gene expression for 5-HT transporters have been observed in raphe nuclei after antidepressant administration (Kuroda, Watanabe, and McEwen, 1994; Lesch, Aulakh, Wolozin, Tolliver, Hill, & Murphy, 1993), other studies have found no effect (Spurlock, Buckland, O'Donovan, & McGuffin, 1994). Pharmacological agents that act on 5-HT$_{1A}$ receptors also have antidepressant properties, and a reduction in cortical 5-HT$_{1A}$ receptor densities has been observed after chronic ipsapirone antidepressant treatment (Shiro, Fujiwara, Hikiji, Hamamura, Shomori, & Kuroda, 1996). Still other antidepressants act on 5-HT$_{2A}$, 5-HT$_{2C}$, and/or 5-HT$_3$ receptors (Murphy, 1998). In addition, various tricyclic antidepressants act on both NE and 5-HT transporters that have been shown to exhibit numerous structural similarities (Barker & Blakely, 1995). Moreover, serotonin-positive neurons have been identified in the LC, and norepinephrine-positive neurons have been found in the dorsal raphe nucleus (Steinbusch, 1981). Because 5-HT interacts with other neurotransmitter systems, particularly NE and DA, the role of serotonin in the etiology of depression requires further study.

Dopamine

Although research has focused primarily on the mechanistic role of NE and 5-HT in depression, there is evidence that the dopaminergic system may also be involved in the etiology of depression. Dopamine shares a common synthetic pathway with norepinephrine (Willner, 1995b). As previously described, tyrosine is transported into the neuron from circulation and is converted to dihydroxyphenylalanine (DOPA) by tyrosine hydroxylase, the rate-limiting enzyme for both dopamine and norepinephrine synthesis. Amino acid decarboxylase then converts DOPA to dopamine. Upon release into the synapse, DA activity is regulated by presynaptic D$_2$ autoreceptors and DA transporters as well as postsynaptic receptors (Bannon, Granneman, & Kapatos, 1995).

The mesocorticolimbic DA system has been implicated in the symptomatology of depression (Willner, 1995b). Dopaminergic projections to the forebrain and limbic structures, including the nucleus accumbens, amygdala, hippocampus, and prefrontal cortex, arise from the ventral tegmental area (VTA) in the mesencephalon. Because DA plays a critical role in reward, motivation, and locomotion, dysregulation of this system may contribute to the anhedonia and psychomotor disturbances that are commonly observed in depression (Willner, 1995b). For example, disturbances in dopaminergic projections to limbic structures have been shown in response to prolonged elevations in CRH and cortisol levels with chronic stress (Chrousos, 1998), and certain antidepressant drugs influence dopaminergic activity by targeting DA receptors or altering DA metabolism (Willner, 1995b). Papp, Klimek, & Willner (1994) observed a reduction in D_2-receptor binding in limbic forebrain structures of rats after chronic mild stress, but effects were reversed by imipramine antidepressant treatment. Antidepressant administration has also been shown to increase gene expression for D_2 autoreceptors in the nucleus accumbens (Ainsworth, Smith, Zetterstrom, Pei, Franklin, & Sharp, 1998) and ventral tegmental area (Dziedzicka-Wasylewska, 1997). Thus, adaptations in dopaminergic function may mediate antidepressant effects, and further research is needed to determine the role of DA in the etiology and treatment of depression.

Neurobiological Adaptations to Exercise

One hypothesis that has been proposed to explain beneficial changes in brain function after exercise is the cross-stressor adaptation hypothesis. The cross-stressor adaptation hypothesis states "a stressor of sufficient intensity and/or duration will induce an adaptation of the stress response systems, which becomes apparent under other similarly taxing states" (Sothmann, Buckworth, Claytor, Cox, White-Welkley, & Dishman, 1996). Thus, chronic physical activity may result in cross-stressor adaptations, and these adaptations could protect the animal against the physiological effects of a nonexercise stressor. As previously discussed, much of the physiological research on

depression has focused on the brain monoaminergic systems, and animal research has provided evidence for a mediating effect of exercise on monoaminergic activity. Thus, these systems may represent a plausible mechanism to explain the antidepressant effects of exercise.

Norepinephrine and neuropeptides. The most consistent evidence for adaptations in brain neurotransmission after physical activity involves the central noradrenergic system. Acute exposure to a physical stressor increases NE release, and prolonged exposure to a severe stressor may result in depletion of NE. However, repeated exposure to a stressor attenuates the depletion of NE, and NE levels may increase, suggesting the acquisition of a resistance to stress. Expression of noradrenergic receptors is also responsive to stress, and reductions in brain cortical β-adrenoreceptor binding density and affinity have been observed after repeated exposure to a stressor (Stanford, 1995).

Both acute and chronic exercise have been shown to influence noradrenergic activity. For example, Kurosawa, Okada, Sato, and Uchida (1993) examined cortical release of NE with acute exercise and found that rats exhibited a 131% increase in NE release after 5 minutes of treadmill walking. Acute treadmill running in rats increases extracellular NE concentrations in brain frontal cortex (Pagliari & Peyrin, 1995a), results in decreased post-mortem levels of brain NE (Barchas & Friedman, 1963), and depletes NE levels in the LC (Stone, 1973). Furthermore, increased levels of NE are lower after a familiar (i.e., repeated) compared to novel duration of treadmill exercise (Pagliari & Peyrin, 1995b).

Though an acute bout of exercise may reduce NE availability, adaptations in noradrenergic systems after chronic exercise appear to preserve brain NE concentrations. Dunn, Reigle, Youngstedt, Armstrong, & Dishman (1996) examined the effects of treadmill training and wheel running on NE levels and metabolites in rats and found increased NE in the pons region of exercise-trained compared to control animals.

Moreover, exercise has been shown to reduce the depletion of NE in response to laboratory stressors. Dishman et al. (1997) examined NE levels after uncontrollable footshock in sedentary and activity wheel animals. Activity wheel runners exhibited 61% greater NE in the LC and 44% higher NE in the dorsal raphe nucleus after footshock compared to sedentary animals. This elevation in NE levels in activity wheel animals is consistent with a blunted release of the neurotransmitter in response to a stressor. These results were supported in a microdialysis study that measured *in vivo* release of NE in response to footshock stress. Soares, Holmes, Renner, Edwards, Bunnell, & Dishman (1999) observed that activity wheel animals exhibited an attenuation of extracellular NE levels in cortex after footshock relative to sedentary animals, suggesting a protective effect of exercise against novel stress. In addition, exercise resulted in favorable adaptations in noradrenergic transmission in a study that used the neonatal clomipramine model of depression. Yoo et al. (2000) observed increased NE concentration and downregulation of β-adrenoreceptors in frontal cortex after chronic wheel running, and effects were similar to those obtained with imipramine antidepressant treatment.

Though chronic exercise appears to induce favorable adaptations in noradrenergic function that have implications for the mental health benefits of physical activity, the mechanisms by which exercise influences noradrenergic transmission is yet unknown. However, it is possible that exercise-induced adaptations in modulatory neuropeptides such as galanin could regulate noradrenergic activity. Soares et al. (1999) reported that activity wheel animals exhibited a 1 SD higher level of GAL mRNA in the LC in conjunction with reduced extracellular NE levels after footshock compared to sedentary rats. Because galanin exerts an inhibitory influence on NE neurons within the LC, this difference in gene expression for galanin might be involved in the attenuation of cortical NE release in response to acute footshock stress in activity wheel animals (Soares et al., 1999).

Basal gene expression for galanin may also be responsive to chronic exercise stress. O'Neal, Van Hoomissen, Holmes, & Dishman (2000) examined GAL mRNA levels in the LC after 6 weeks of treadmill training and found that gene expression for galanin was higher in treadmill-trained animals compared to sedentary animals. Although prior research examining GAL mRNA levels after chronic voluntary exercise found no difference in basal concentration between sedentary and activity wheel animals (Soares et al., 1999), adaptations to exercise may be dependent upon the nature or intensity of the exercise stimulus. If increases in GAL mRNA result in enhanced post-translational activity within LC neurons, adaptations in galanin represent a plausible mechanism to explain the blunted noradrenergic response observed in trained animals upon exposure to both novel (e.g., footshock) and familiar (e.g., acute exercise) stress (Soares et al., 1999).

Serotonin. It is plausible that affective changes after exercise may also be mediated by adaptations in serotonergic transmission, and evidence from animal studies suggests that physical activity influences serotonergic function. Because serotonin synthesis is limited by the availability of tryptophan in the blood, conditions that increase free circulating tryptophan will influence 5-HT function. Tryptophan is carried in the blood bound to protein and competes with free fatty acids (FFA) for albumin. During exercise, lipolysis causes an increase in plasma FFAs that displace tryptophan from albumin and increase the level of free tryptophan in the circulation (Chaouloff, Elghozi, Guezennec, & Laude, 1985). Thus, enhanced availability of tryptophan during exercise could contribute to increased 5-HT synthesis and metabolism in central serotonergic neurons (Chaouloff, 1997). Although Kurosawa et al. (1993) found that cortical 5-HT release was elevated by 133% after 5 minutes of exercise, most studies have examined 5-HT after exhaustive exercise. For example, Romanowski & Grabiec (1974) observed that treadmill exercise to exhaustion was associated with increased brain 5-HT concentration and proposed a mechanistic role of 5-HT in fatigue. In addition, acute exhaustive exercise has produced midbrain elevations in 5-HT and 5-HIAA levels (Bailey, Davis, &

Ahlborn, 1992), increased concentration of brain tryptophan, elevations in 5-HT levels in the brain stem and hypothalamus, and increased 5-HIAA in the brain stem, hypothalamus, hippocampus, and striatum (Blomstrand, Perrett, Parry-Billings, & Newsholme, 1989).

Chronic exercise may also produce long-term adaptations in serotonergic function. Yoo et al. (2000) observed that serotonergic activity as indicated by the cortical 5-HIAA/5-HT ratio tended to be greater for exercise-trained compared to sedentary animals. Exercise may also influence 5-HT response to stressors. For example, Dishman et al. (1997) examined serotonergic function in sedentary and activity wheel animals after uncontrollable footshock. Sedentary animals exhibited 28% higher 5-HT in the amygdala following footshock compared to activity wheel animals. Sedentary animals also exhibited 33% and 31% higher concentrations of 5-HIAA in the amygdala and CA1 of the hippocampus respectively after footshock compared to activity wheel runners, and sedentary animals had 17% higher 5-HIAA/5-HT in the paraventricular nucleus after footshock stress.

However, exercise-induced changes in serotonergic transmission are often smaller in magnitude and less consistent than other neurochemical indices, and there is currently little evidence that serotonergic mechanisms play a primary role in the mental health benefits of exercise. Although the most common antidepressant medications act to block 5-HT transporters, exercise training does not appear to influence gene expression for the 5-HT transporter in the dorsal raphe. However, post-transcriptional adaptations in transporter function are possible and require further study (Van Hoomissen, O'Neal, Dishman, Holmes, & Dishman, 2000). In addition, adaptations in serotonergic function with exercise training vary among animals and might be dependent on the stress component of the exercise stimulus (Wilson & Marsden, 1996). Adaptations in 5-HT transmission could also be attributable to changes in other systems that are modulated by exercise. Therefore, although animal research has found that brain 5-HT synthesis and

metabolism is influenced by physical activity, it is not currently known whether exercise-induced alterations in serotonergic function directly mediate mood responses to exercise.

Dopamine. Most studies examining the effect of exercise on dopaminergic activity have focused on the nigrostriatal pathway which is involved in the regulation of motor activity, and both acute and chronic exercise in animals have been shown to influence dopaminergic activity. For example, Bailey et al. (1992) observed increases DA and DOPAC levels in midbrain, hippocampus, and striatum after exhaustive exercise. Liste, Guerra, Caruncho, & Labandeira-Garcia (1997) found that 20 minutes of treadmill running produced elevations in Fos expression in the striatum, but this effect was prevented by pretreatment with a D_1 receptor antagonist. In addition, acute bouts of treadmill exercise have been shown to increase turnover of dopamine in the striatum (Hattori, Naoi, & Nishino, 1994). Changes in density and affinity of DA receptors have also been observed after chronic training in animals (Mazzeo, 1991). For example, chronic treadmill training increases density of dopamine (D_2) receptors in the striatum (Gilliam et al., 1984; MacRae, Spirduso, Cartee, Farrar, & Wilcox, 1987).

The evidence for a mediating effect of DA on the antidepressant effects of exercise is limited, as most studies have limited examination of DA activity to striatal regions. However, exercise has been shown to induce changes in brain structures that have relevance for depression. For example, Liste et al. (1997) observed increases in Fos expression in the nucleus accumbens after 20 minutes of treadmill running. In addition, Speciale, Miller, McMillen, & German (1986) examined the effects of acute bouts of high and low speed forced wheel running on DA levels and metabolism in the nucleus accumbens and prefrontal cortex. High intensity exercise resulted in increased DA metabolism in the nucleus accumbens. However, there was no effect of low speed exercise, and no differences were observed in the prefrontal cortex. Dopamine activity in hypothalamic nuclei may also be influenced by exercise. Dishman et al. (1997) observed that sedentary animals had 52% higher DA concentrations in the arcuate nucleus and

50% greater DOPAC/DA in the paraventricular nucleus of the hypothalamus compared to activity wheel animals after uncontrollable footshock. Thus, exercise appears to induce adaptations in the central dopaminergic systems; however, it is not known whether DA responses have relevance for mood and emotion or whether effects are indices of locomotion. In order to support the biological plausibility of exercise as an antidepressant intervention, additional studies that focus on mesocorticolimbic structures are needed.

Olfactory Bulbectomy Animal Model of Depression

Various neurotransmitter systems have been implicated in the pathology of depression; however, the etiology of depressive disorders is not fully understood. Because brain systems cannot be adequately studied in humans, animal models of depression are used to further investigate the neurobiological mechanisms underlying depression. Although the use of animal models has limitations in applicability to mental illness in humans, the study of animals permits examination of neural functioning at a level that is not possible using human subjects. Over the last several decades, several animal models of depression have been developed to investigate the etiology of depressive disorders and to test the antidepressant activity of various drugs. When selecting an animal model, considerations must be given to the evidence for the validity of the model and its applicability to human populations as well as its feasibility and reproducibility in a laboratory setting (Willner, 1995a).

One such model is bilateral olfactory bulbectomy (OBX) in rats, a procedure that produces a syndrome of behavioral and physiological abnormalities. This model has been widely used to test the efficacy of antidepressant medications and to examine neurobiological mechanisms of depressive disorders. Furthermore, OBX appears to have good evidence to support its validity as an animal model of depression. (Kelly et al., 1997).

Validity Evidence. The first line of validity evidence for an animal model of depression is predictive validity. Antidepressant drugs should be effective in alleviating features of the depressive syndrome and follow a clinically consistent time course. In addition, the syndrome should be selectively responsive to agents demonstrated to have antidepressant properties and not responsive to other types of drugs (Willner, 1995a). Though several animal models such as the Porsolt forced swim test induce signs that are reversed upon antidepressant administration (Porsolt, Le Pichon, & Jalfre, 1977), symptoms are often responsive to acute treatment with antidepressant agents. A stronger argument for validity of a model is presented for manipulations that induce symptoms that are reversed only with chronic pharmacological administration, simulating the treatment response observed in depressed patients. Various classes of antidepressant drugs have been tested using the OBX model including tricyclic antidepressants (TCAs), atypical antidepressants, monoamine oxidase inhibitors (MAOs), and selective serotonin reuptake inhibitors (SSRIs). Chronic but not acute administration of these agents has been shown to reduce or reverse behavioral and neurobiological features of the OBX syndrome (Kelly et al., 1997).

The animal model must also demonstrate evidence of validity in which animals exhibit signs that are isomorphic with features of human depression. Though depressed mood cannot be determined in animals, anhedonia, or a loss of interest in previously enjoyed activities, may be simulated in animal models. Other features of depression that can be measured in animals include psychomotor disturbances, changes in sleeping and/or eating patterns, and sexual dysfunction. After olfactory bulbectomy, animals display behaviors that are consistent with features of human depression. Though the expression of anhedonia in bulbectomized rats is more difficult to interpret because of the anosmic component of the model, evidence suggests that anhedonic-like behaviors are associated with the OBX syndrome. For example, Calcagnetti, Quatrella, and Schecter (1996) examined conditioned place preference for cocaine in OBX and anosmic rats and

observed a disruption in learned place preference in bulbectomized animals. Additional features of the olfactory bulbectomy syndrome which are indicative of a depressive-like condition include an augmented acoustic startle response (McNish & Davis, 1997), sexual dysfunction (Edwards, Griffis, & Tardivel, 1990; Lumia, Teicher, Salchli, Ayers, & Possidente, 1992), locomotor disturbances (Lumia et al., 1992; Marcilhac, Maurel, Anglade, Ixart, Mekaouche, Hery, & Siaud, 1997), and altered circadian rhythms (Lumia et al., 1992; Marcilhac et al., 1997). However, consideration should be given as to whether behaviors are due to a depressive-like condition or instead are a direct consequence of the physiological perturbations associated with the model.

Etiological and mechanistic evidence for validity should also be considered to determine whether the model is theoretically consistent with depression. However, this evidence is limited, as an animal model represents an artificial attempt to induce an inherently human condition in a laboratory setting in animal species. Nevertheless, an animal model of depression should be based on a theoretical perspective using neurobiological or behavioral methods. The olfactory bulb is a telencephalic structure that is highly interconnected with limbic areas. Removal of the olfactory bulb produces profound disturbances in monoaminergic transmission and endocrine function, and behavioral abnormalities after olfactory bulbectomy develop 1 to 2 weeks following surgery (Kelly et al., 1997). The olfactory bulb exhibits numerous afferent and efferent projections with brain structures that regulate mood and emotion, and disturbances following OBX are consistent with the neurobiology of depression; thus, there is evidence to support the validity of the OBX model from an etiological and mechanistic perspective.

Olfactory Bulbectomy and Brain Monoamines. Olfactory bulbectomy produces disturbances in brain neurotransmitter systems, including NE, 5-HT, and DA. For example, Iwasaki, Fujiwara, and Ueki (1989) found that norepinephrine release from the lateral hypothalamus was reduced following bulbectomy, and increases in GAL

mRNA levels in the LC have been observed after lesioning of a terminal NE field by OBX (Holmes & Crawley, 1996). Moreover, behavioral and neurobiological abnormalities are reduced after chronic administration with antidepressant agents that target the noradrenergic system. Nesterova, Gurevich, Nesterov, Otmakhova, & Bobkova (1997) observed that OBX produced pathological changes in LC tissue morphology, and these abnormalities were improved with chronic TCA administration.

Olfactory bulbectomy also produces alterations in serotonergic transmission, and researchers have referred to OBX as a model for "hyposerotonergic depression" (Lumia et al., 1992). Nesterova et al (1997) observed a profound degeneration of neurons in the dorsal raphe nucleus following OBX, but these effects were significantly reduced with chronic antidepressant treatment. Gurevich, Aleksandrova, Otmakhova, Katkov, Nesterova, & Bobkova (1993) observed an increase in binding density for 5-HT$_2$ receptors in frontal cortex following bulbectomy that was reduced after treatment with the antidepressant trazodone. Zhou, Grecksch, Becker, Frank, Pilz, & Huether (1998) also observed changes in serotonergic activity after OBX including increased 5-HT transporter density, higher tryptophan hydroxylase levels, elevated 5-HIAA levels, and a higher 5-HIAA/5-HT ratio in the frontal cortex. Although numerous serotonergic changes are observed following OBX, it is not known whether these effects are primary or secondary to other neurochemical disturbances.

Though the effect of olfactory bulbectomy on dopaminergic function has been less frequently studied relative to other monoaminergic systems, alterations in DA transmission have been observed following bulbectomy. For example, Iwasaki et al., (1989) found that DA release from the ventromedial hypothalamus was increased after bulbectomy. However, Lumia et al. (1992) found no difference in DA levels following bulbectomy in striatum or nucleus accumbens. Given the role of DA in reward and motivation as well as the efficacy of atypical antidepressants that target DA, further

research is needed to determine the role of the dopaminergic system in the OBX syndrome.

In Situ Hybridization

A biological framework for the etiology and treatment of depression and other mental disorders can be outlined using the following five principles proposed by psychiatrist Eric Kandel (1998): (1) actions at the brain level are responsible for all mental and psychological processes, (2) brain functioning is controlled by genes, (3) social, developmental, and environmental factors can produce alterations in gene expression, (4) alterations in gene expression induce changes in brain functioning, and (5) treatments for mental illness exert their effect by producing alterations in gene expression resulting in beneficial changes in brain function. Thus, within this framework, depression is proposed to result from disturbances in brain processes, and treatments for depression such as psychotherapy, medication or exercise produce alterations in genes that result in the alleviation of depressive symptoms.

In situ hybridization enables the study of potential antidepressant treatments at the level of gene expression. In this technique, a probe with the complementary sequence of bases is applied to the target tissue and specifically hybridizes to the protein of interest. In addition to specificity of measurement, the use of this method permits anatomical localization of the protein (Tecott, Eberwine, Barchas, & Valentino, 1994). Of particular relevance for the study of depression are genes that encode receptors (e.g., D_2 receptors), synthetic enzymes (e.g., TH), transporters (e.g., SERT), and modulatory neuropeptides (e.g., GAL, NPY) for monoaminergic neurotransmitters that are targeted by pharmacological interventions. Thus, the use of *in situ* hybridization to investigate neurobiological adaptations after chronic exercise may provide additional evidence for the biological plausibility of exercise as an antidepressant intervention.

Though *in situ* hybridization techniques vary across laboratories, the protocols share several primary components as summarized by Tecott et al. (1994). First, the tissue

is prepared to retain nucleic acids, preserve morphology, and promote probe penetration. Various methods are used to preserve the tissue including perfusion, paraffin, and freezing. Common fixatives include ethanol, acetic acid, and/or formaldehyde, and the method is selected based on tissue and probe characteristics.

Second, the type of probe is selected from three main classes of probes including complementary DNA (cDNA), RNA (riboprobe), and oligonucleotide probes. Each class of probe has advantages and disadvantages that must be considered during the selection process. Though cDNA probes are specific and simple to use, they also exhibit greater reannealing due to the double-stranded structure and may produce higher levels of background noise. In contrast, riboprobes are single-stranded and form stable hybrids but also may produce greater nonspecific binding. Oligonucleotide probes are short, single-stranded DNA probes that allow greater penetration and may be precisely tailored for the desired sequence of bases. However, oligonucleotide probes require published sequences and may form less stable hybrids that limit specificity.

Third, the probe is labeled with the desired radioisotope (e.g., ^{32}P, ^{3}H, and ^{35}S). Selection of the labeling method is based on sensitivity criteria, anatomical resolution, and time required for radiographic exposure. A common method for probe labeling involves the enzymatic addition of the radiolabel to the 3'-end of the oligonucleotide.

Fourth, the hybridization solution is prepared and applied to the tissue. In addition to the probe, the solution contains components to reduce nonspecific binding, enhance rate of hybridization, and amplify the signal.

Lastly, after incubation with the hybridization solution, post-hybridization treatments are performed to reduce nonspecific binding and decrease the level of background. Sections are then dipped in emulsion or exposed to autoradiographic film to permit quantitative analysis of the radioactive signal within the tissue.

A primary advantage of *in situ* hybridization for research investigating the biological plausibility of exercise as an antidepressant intervention is the ability to

examine potential mechanisms for neurotransmitter regulation within discrete brain regions that have relevance for the mediation of mood and behavior. Because both exercise and pharmacotherapy alter brain monoaminergic neurotransmission, it is possible that the treatments share a common mechanism of action at the level of gene expression that could explain observed responses in neurotransmitter systems. Based on prior research, it was decided to initially focus on the noradrenergic system, as chronic physical activity and antidepressant administration produce similar alterations in noradrenergic activity (Yoo et al., 2000). Thus, the purpose of the studies reported herein was to examine gene expression for proteins that regulate noradrenergic function in cell bodies of the locus coeruleus after chronic exercise.

References

Ainsworth, K., Smith, S.E., Zetterstrom, T.S.C., Pei, Q., Franklin, M., & Sharp, T. (1998). Effect of antidepressant drugs on dopamine D_1 and D_2 receptor expression and dopamine release in the nucleus accumbens of the rat. Psychopharmacology, 140, 470-477.

American Psychiatric Association. (1994). Diagnostic and statistical manual (4th ed.). Washington, DC: author.

Aston-Jones, G., Rajkowski, J., & Cohen, J. (1999). Role of locus coeruleus in attention and behavioral flexibility. Biological Psychiatry, 46, 1309-1320.

Bailey, S.P., Davis, J.M., & Ahlborn, E.N. (1992). Effect of increased brain serotonergic activity on endurance performance in the rat. Acta Physiologica Scandinavica, 145, 75-76.

Baker, R.A., Herkenham, M., & Brady, L.S. (1996). Effects of long-term treatment with antidepressant drugs on proopiomelanocortin and neuropeptide Y mRNA expression in the hypothalamic arcuate nucleus of rats. Journal of Neuroendocrinology, 8, 337-343.

Bannon, M.J., Granneman, J.G., & Kapatos, G. (1995). The dopamine transporter. In F.E. Bloom & D.J. Kupfer, Psychopharmacology: The Fourth Generation of Progress (pp. 179-187). New York: Raven Press.

Barchas, J.D. & Friedman, D.X. (1963). Brain amines: Response to physiological stress. Biochemistry and Pharmacology, 12, 1232-1235.

Barker, E.L. & Blakely, R.D. (1995). Norepinephrine and serotonin transporters: molecular targets of antidepressant drugs. In F.E. Bloom & D.J. Kupfer, Psychopharmacology: The Fourth Generation of Progress, pp. 321-332. New York: Raven Press.

Blomstrand, E., Perrett, D., Parry-Billings, M. & Newsholme, E.A. (1989). Effect of sustained exercise on plasma amino acid concentrations and on 5-hydroxytryptamine metabolism in six different brain regions in the rat. Acta Physiologica Scandinavica, 136(3), 473-481.

Blumenthal, J.A., Babyak, M.A., Moore, K.A., Craighead, W.E., Herman, S., Khatri, P., Waugh, R., Napolitano, M.A., Forman, L.M., Appelbaum, M., Doraiswamy, P.M., & Krishnan, K.R. (1999). Effects of exercise training on older patients with major depression. Archives of Internal Medicine, 159, 2349-2356.

Burton, R. (1632). The Anatomy of Melancholy. Oxford: Printed by Ion Lichfield for Henry Cripps.

Caberlotto, L., Fuxe, K., Overstreet, D.H., Gerrard, P., & Hurd, Y. (1998). Alterations in neuropeptide Y and Y1 receptor mRNA expression in brains from an animal model of depression: region specific adaptation after fluoxetine treatment. Molecular Brain Research, 59, 58-65.

Calcagnetti, D.J., Quatrella, L.A., & Schechter, M.D. (1996). Olfactory bulbectomy disrupts the expression of cocaine-induced conditioned place preference. Physiology & Behavior 59(4/5), 597-604.

Camacho, T.C., Roberts, R.E., Lazarus, N.B., Kaplan, G.A., & Cohen, R.D. (1991). Physical activity and depression: Evidence from the Alameda County Study. American Journal of Epidemiology, 134, 220-231.

Charney, D.S. (1998). Monoamine dysfunction and the pathophysiology and treatment of depression. Journal of Clinical Psychiatry, 59(Suppl 14), 11-14.

Chaouloff, F. (1997). Effects of acute physical exercise on central serotonergic systems. Medicine and Science in Sports and Exercise, 28, 63-74.

Chaouloff, F., Elghozi, J.L., Guezennee, Y., & Laude, D. (1985). Effects of conditioned running on plasma, liver and brain tryptophan and on brain 5-hydroxytryptamine metabolism of the rat. British Journal of Pharmacology, 86(1), 33-41.

Cheetham, S.C., Katona, C.L.E., & Horton, R.W. (1991). Post-mortem studies of neurotransmitter biochemistry in depression and suicide. In R. Horton & C.L.E. Katona (Eds.) Biological aspects of affective disorders (pp. 192-221). Academic Press: London.

Chrousos, G.P. (1998). Stressors, stress, and neuroendocrine integration of the adaptive response. Annals of the New York Academy of Sciences, 851, 311-335.

Craft, L.L, & Landers, D.M. (1998). The effect of exercise on clinical depression and depression resulting from mental illness: a meta-analysis. Journal of Sport & Exercise Psychology, 20, 339-357.

Dahlstrom, A. & Fuxe, K. (1964). Evidence for the existence of monoamine-containing neurons in the central nervous system. Acta Physiologica Scandinavica, 62(Suppl. 232) 1-55.

DeVane, C.L. (1998). Differential pharmacology of newer antidepressants. Journal of Clinical Psychiatry, 59 (Suppl. 20), 85-93.

Dishman, R.K. (1997). Brain monoamines, exercise, and behavior stress: animal models. Medicine and Science in Sports and Exercise, 29(1), 63-74.

Dishman, R.K. (1998). Physical activity and mental health. In H. Friedman (Ed.), Encyclopedia of Mental Health, Vol. 3 (pp. 171-188).

Dishman, R.K., Renner, K.J., Youngstedt, S.D., Reigle, T.G., Bunnell, B.N., Burke, K.A., Yoo, H.S., Mougey, E.H., & Meyerhoff, J.L. (1997). Activity wheel running reduces escape latency and alters brain monoamine levels after footshock. Brain Research Bulletin, 42(5), 399-406.

Doyne, E.J., Ossip-Klein, K.J., Bowman, E.D., Osborn, K.M., McDougall-Wilson, I.B., & Neimeyer, R.A. (1987). Running versus weight lifting in the treatment of depression. Journal of Consulting and Clinical Psychology, 55(5), 748-754.

Dunn, A.L, Reigle, T.G., Youngstedt, S.D., Armstrong, R.B., & Dishman, R.K. (1996). Brain norepinephrine and metabolites after treadmill training and wheel running in rats. Medicine and Science in Sports and Exercise, 28, 204-209.

Dziedzicka-Wasylewska, M. (1997). The effect of imipramine on the amount of mRNA coding for rat dopamine D_2 autoreceptors. European Journal of Pharmacology, 337, 291-296.

Edwards, D.A., Griffis, K.T., & Tardivel, C. (1990). Olfactory bulb removal: Effects on sexual behavior and partner preference in male rats. Physiology & Behavior, 48(3), 447-450.

Farmer, M.E., Locke, B.Z., Moscicki, E.K., Dannenberg, A.L., Larson, D.B., & Radloff, L.S. (1988). Physical activity and depressive symptoms: the NHANES I epidemiologic follow-up study. American Journal of Epidemiology, 128, 1340-1351.

Ferketich, A.K., Schwartzbaum, J.A., Frid, D.J., & Moeschberger, M.L. (2000). Depression as an antecedent to heart disease among women and men in the NHANES I study. Archives of Internal Medicine, 160, 1261-1268.

Finta, E.P., Regenold, J.T., & Illes, P. (1992). Depression by neuropeptide Y of noradrenergic inhibitory postsynaptic potentials of locus coeruleus neurons. Naunyn Schmiedebergs Arch Pharmacology, 346, 472-474.

Foote, S.L., Bloom, F.E., & Aston-Jones, G. (1983). Nucleus locus ceruleus: New evidence of anatomical and physiological specificity. Physiological Reviews, 63, 844-903.

Frasure-Smith, N., Lesperance, F., Juneau, M., Talajic, M., & Bourassa, M.G. (1999). Gender, depression, and one-year prognosis after myocardial infarction. Psychosomatic Medicine, 61, 26-37.

Gilliam, P.E., Spirduso, W.W., Martin, T.P., Walters, T.J., Wilcox, R.E., & Farrar, R.P. (1984). The effects of exercise training on [^{3}H]-spiperone binding in rat striatum. Pharmacology, Biochemistry & Behavior, 20, 863-867.

Greist, J.H., Klein, M.H., Eischens, R.R., Faris, J., Gurman, A.S., & Morgan, W.P. (1978). Running through your mind. Journal of Psychosomatic Research, 22, 259-294.

Greenberg, P.E., Stiglin, L.E., Finkelstein, S.N., & Berndt, E.R. (1993). The economic burden of depression in 1990. Journal of Clinical Psychiatry, 54(11), 405-418.

Gurevich, E.V, Aleksandrova, I.A., Otmakhova, N.A., Katkov, Y.A., Nesterova, I.V., & Bobkova, N.V. (1993). Effects of bulbectomy and subsequent antidepressant treatment on brain 5-HT$_2$ and 5-HT$_{1A}$ receptors in mice. Pharmacology Biochemistry and Behavior, 45, 65-70.

Halliday, G., Harding, A. & Paxinos, G. (1995). Serotonin and tachykinin systems. In G. Paxinos (Ed.) The rat nervous system, (2nd ed., pp. 929–935). San Diego: Academic Press.

Hashimoto, H., Onishi, H., Koide, S., Kai, T., & Yamagami, S. (1996). Plasma neuropeptide Y in patients with major depressive disorder. Neuroscience Letters, 216, 57-60.

Hattori, S., Naoi, M., & Nishino, H. (1994). Striatal dopamine turnover during treadmill running in the rat: relation to the speed of running. Brain Research Bulletin, 35, 41-49.

Heilig, M. & Widerlov, E. (1990). Neuropeptide Y: an overview of central distribution, functional aspects, and possible involvement in neuropsychiatric illnesses. Acta Psychiatrica Scandinavica, 82, 95-114.

Hirschfeld, R.M.A., Keller, M.B, Panico, S., Arons, B.S., Barlow, D., Davidoff, F., Endicott, J., Froom, J., Goldstein, M., Gorman, J.M., Guthrie, D., Marek, R.G., Maurer, T.A., Meyer, R., Phillips, K., Ross, J., Schwenk, T.L., Sharfstein, S.S., Thase, M.E., & Wyatt, R.J. (1997). The National Depressive and Manic-Depressive Association consensus statement on the undertreatment of depression. Journal of the American Medical Association, 277, 333-340.

Hirschfeld, R.M.A. (1998). American health care systems and depression: the past, present, and future. Journal of Clinical Psychiatry, 59(Suppl. 20), 5-10.

Holets, V.R., Hokfelt, T., Rokaeus, A., Terenius, L., & Goldstein, M. (1988). Locus coeruleus neurons in the rat containing neuropeptide Y, tyrosine hydroxylase or galanin and their efferent projections to the spinal cord, cerebral cortex and hypothalamus. Neuroscience, 24, 893-906.

Holmes, P.V. & Crawley, J.N. (1995). Coexisting neurotransmitters in central noradrenergic neurons. In F.E. Bloom & D.J. Kupfer (Eds.), Psychopharmacology: The Fourth Generation of Progress. New York: Raven Press, Ltd.

Holmes, P.V. & Crawley, J.N. (1996). Olfactory bulbectomy increases prepro-galanin mRNA levels in the rat locus coeruleus. Molecular Brain Research, 36, 184-188.

Iwasaki, K., Fujiwara, M., & Ueki, S. (1989). In vivo changes in brain catecholamine release from rat hypothalamus following olfactory bulbectomy. Pharmacology Biochemistry & Behavior, 34, 879-885.

Kandel, E.R. (1998). A new intellectual framework for psychiatry. American Journal of Psychiatry, 155(4), 457-469.

Kelly, J.P., Wrynn, A.S., & Leonard, B.E. (1997). The olfactory bulbectomized rat as a model of depression: an update. Pharmacology & Therapeutics, 74(3), 299-316.

Kessler, R.C., McGonagle, K.A., Zhao, S., Nelson, C.B., Hughes, M., Eshleman, S., Wittchen, H-U, & Kendler, K.S. (1994). Lifetime and 12-month prevalence of DSM-III-R psychiatric disorders in the United States: Results from the National Comorbidity Survey. Archives of General Psychiatry, 51, 8-18.

Krause, N., Goldenhar, L., Liang, J., Jay, G., & Maeda, D. (1993). Stress and exercise among the Japanese elderly. Social Science Medicine, 36, 1429-1441.

Kuroda,Y., Watanabe, Y., & McEwen, B.S. (1994). Tianeptine decreases both serotonin transporter mRNA and binding sites in rat brain. European Journal of Pharmacology, 268, R3-R5.

Kurosawa, M., Okada, K., Sato, A., & Uchida, S. (1993). Extracellular release of acetylcholine, noradrenaline and serotonin increases in cerebral cortex during walking in conscious rats. Neuroscience Letters, 161, 73-76.

Lesch, K.P., Aulakh, C.S, Wolozin, B.L., Tolliver, T.J., Hill, J.L., & Murphy, D.L. (1993). Regional brain expression of serotonin transporter mRNA and its regulation by reuptake inhibiting antidepressants. Molecular Brain Research, 17, 31-35.

Liste, I., Guerra, M.J., Caruncho, H.J., & Labandeira-Garcia, J.L. (1997). Treadmill running induces striatal Fos expression via NMDA glutamate and dopamine receptors. Experimental Brain Research, 115, 458-469.

Lumia, A.R., Teicher, M.H., Salchli, F., Ayers, E., & Possidente, B. (1992). Olfactory bulbectomy as a model for agitated hyposerotonergic depression. Brain Research, 587, 181-185.

MacRae, P.G., Spirduso, W.W., Cartee, G.D., Farrar, R.P., & Wilcox, R.E. (1987). Endurance training effects on striatal D2 dopamine-receptor binding and striatal dopamine metabolite levels. Neuroscience Letters, 79, 138-144.

Maes, M. & Meltzer, H.Y. (1995). The serotonin hypothesis of major depression. In F.E. Bloom & D.J. Kupfer (Eds.), Psychopharmacology: The Fourth Generation of Progress (pp. 933-943). New York: Raven Press.

Marcilhac, A., Maurel, D., Anglade, G., Ixart, G., Mekaouche, M., Hery, F., & Siaud, P. (1997). Effects of bilateral olfactory bulbectomy on circadian rhythms of ACTH, corticosterone, motor activity and body temperature in male rats. Archives of Physiology and Biochemistry, 105 (6), 552-559.

Martinsen, E.W., Hoffart, A. & Solberg, O. (1989). Comparing aerobic with nonaerobic forms of exercise in the treatment of clinical depression: A randomized controlled trial. Comprehensive Psychiatry, 30, 324-331.

Martinsen, E.W. & Medhus, A. (1989). Adherence to exercise and patients' evaluation of physical exercise in a comprehensive treatment programme for depression. Nordisk Psykiatrisk Tidsskrift, 43, 411-415.

Martinsen, E.W., Medhus, A., & Sandvik, L. (1985). Effects of aerobic exercise on depression: a controlled study. British Medical Journal, 291, 109.

Mazzeo, R.S. (1991). Catecholamine responses to acute and chronic exercise. Medicine and Science in Sports and Exercise, 23(7), 839-845.

McNish, K.A., & Davis, M. (1997). Olfactory bulbectomy enhances sensitization of the acoustic startle reflex produced by acute or repeated stress. Behavioral Neuroscience, 111(1), 80-91.

Murphy, DL., Andrews, A.M., Wichems, C.H., Li, Q., Tohda, M., & Greenberg, B. (1998). Brain serotonin neurotransmission: an overview and update with an emphasis on serotonin subsystem heterogeneity, multiple receptors, interactions with other neurotransmitter systems, and consequent implications for understanding the actions of serotonergic drugs. Journal of Clinical Psychiatry, 59(Suppl. 15), 4-12.

Murray, C.J.L. & Lopez, A.D. (1996). Evidence-based health policy – lessons from the Global Burden of Disease Study. Science, 274, 740-743.

Nesterova, I.V., Gurevich, E.V., Nesterov, V.I., Otmakhova, N.A., & Bobkova, N.V. (1997). Bulbectomy-induced loss of raphe neurons is counteracted by antidepressant treatment. Progress in Neuro-Psychological & Biological Psychiatry, 21, 127-140.

North, T.C., McCullagh, P., & Tran, Z.V. (1990). Effect of exercise on depression. Exercise and Sport Sciences Reviews, 18, 379-415.

O'Neal, H.A., Dunn, A.L., & Martinsen, E.W. (2000). Depression and exercise. International Journal of Sport Psychology, 31, 110-135.

O'Neal, H.A., Van Hoomissen, J.D., Holmes, P.V., & Dishman, R.K. (2000). Prepro-galanin mRNA levels are increased in rat locus coeruleus after treadmill exercise training. Manuscript submitted for publication.

Ogilvie, A.D., Battersby, S., Bubb, V.J., Fink, G., Harmar, A.J., Goodwin, G.M., & Smith, C.A.D. (1996). Polymorphism in serotonin transporter gene associated with susceptibility to major depression. Lancet, 347, 731-733.

Paffenbarger, R.S., Lee, I.M., & Leung, R. (1994). Physical activity and personal characteristics associated with depression and suicide in American college men. Acta Psychiatrica Scandinavica, 377(Suppl.), 16-22.

Pagliari, R. & Peyrin, L. (1995a). Norepinephrine release in the rat frontal cortex under treadmill exercise: a study with microdialysis. Journal of Applied Physiology, 78, 2121-2130.

Pagliari, R. & Peyrin, L. (1995b). Physical conditioning in rats influences the central and peripheral catecholamine responses to sustained exercise. European Journal of Applied Physiology, 71, 41-52.

Papp, M., Klimek, V., & Willner, P. (1994). Parallel changes in dopamine D2 receptor binding in limbic forebrain associated with chronic mild stress-induced anhedonia and its reversal by imipramine. Psychopharmacology, 115, 441-446.

Penninx, B.W.J.H., Leveille, S., Ferrucci, L., van Eijk, J.T.M., & Guralnik, J.M. (1999). Exploring the effect of depression on physical disability: Longitudinal evidence from the established populations for epidemiologic studies of the elderly. American Journal of Public Health, 89(9), 1346-1352.

Pieribone, V.A., Xu, Z.Q., Zhang, X., Grillner, S., Bartfai, T., & Hokfelt, T. (1995). Galanin induces a hyperpolarization of norepinephrine-containing locus coeruleus neurons in the brainstem slice. Neuroscience, 64, 861-874.

Porsolt, R.D., Le Pichon, M., & Jalfre, M. (1977). Depression: a new animal model sensitive to antidepressant treatment. Nature, 266, 730-732.

Rajala, U., Uusimaki, A., Keinanen-Kiukaanniemi, S., & Kivela, S.L. (1994). Prevalence of depression in a 55-year-old Finnish population. Society of Psychiatry and Psychiatric Epidemiology, 29, 126-130.

Romanowski, W. & Grabiec, S. (1974). The role of serotonin in the mechanism of central fatigue. Acta Physiologica Polonica, 25(2), 127-134.

Sargeant, J.K., Bruce, M.L., Florio, L.P., & Weissman, M.M. (1990). Factors associated with 1-year outcome of major depression in the community. Archives of General Psychiatry, 47, 519-526.

Schatzberg, A.F. (1998). Noradrenergic versus serotonergic antidepressants: Predictors of treatment response. Journal of Clinical Psychiatry, 59(Suppl. 14), 15-18.

Schildkraut, J.J. (1965). The catecholamine hypothesis of affective disorders: a review of the supporting evidence. American Journal of Psychiatry, 122, 509-522.

Seutin, V., Verbanck, P., Massote, P., & Dresse, A. (1989). Galanin decreases the activity of locus coeruleus neurons in vitro. European Journal of Pharmacology, 164, 373-376.

Shiro, Y., Fujiwara, Y., Hikiji, M., Hamamura, T., Shomori, T., & Kuroda, S. (1996). Effect of chronic ipsapirone treatment on the density of 5-HT$_{1A}$ receptors and 5-HT$_{1A}$ receptor mRNA in discrete regions of the rat brain. Psychiatry and Clinical Neurosciences, 50, 141-146.

Smialowska, M. & Legutko, B. (1991). Influence of imipramine on neuropeptide Y immunoreactivity in the rat brain. Neuroscience, 41(2/3), 767-771.

Soares, J., Holmes, P.V., Renner, K.J., Edwards, G.L., Bunnell, B.N., & Dishman, R.K. (1999) . Brain noradrenergic responses to footshock after chronic activity-wheel running. Behavioral Neuroscience, 113, 558-566.

Sothmann, M.S., Buckworth, J., Claytor, R.P., Cox, R.H., White-Welkley, J.E., & Dishman, R.K. (1996). Exercise and the cross-stressor adaptation hypothesis. Exercise and Sport Sciences Reviews, 24, 267-287.

Speciale, S.G., Miller, J.D., McMillen, B.A., & German, D.C. (1986). Activation of specific central dopamine pathways: Locomotion and footshock. Brain Research Bulletin, 16(1), 33-38.

Spurlock, G., Buckland, P., O'Donovan, M., & McGuffin, P. (1994). Lack of effect of antidepressant drugs on the level of mRNAs encoding serotonergic receptors, synthetic enzymes and 5HT transporter. Neuropharmacology, 33(3/4), 433-440.

Stanford, S.C. (1995). Central noradrenergic neurones and stress. Pharmacology and Therapeutics, 68(2), 297-342.

Steinbusch, H.W.M. (1981). Distribution of serotonin-immunoreactivity in the central nervous system of the rat—cell bodies and terminals. Neuroscience, 6(4), 557-618.

Stone, E.A. (1973). Accumulation and metabolism of NE in rat hypothalamus after exhaustive stress. Journal of Neurochemistry, 21, 589-601.

Tecott, L.H., Eberwine, J.H., Barchas, J.D., & Valentino, K.L. (1994). Methodological considerations in the utilization of *in situ* hybridization. In J.H. Eberwine, K.L. Valentino, & J.D. Barchas (Eds.), In Situ Hybridization in Neurobiology (pp. 3-23). New York: Oxford University Press.

Van Hoomissen, J.D., O'Neal, H.A., Dishman, J.E., Holmes, P.V., & Dishman, R.K. (2000). Serotonin transporter mRNA in dorsal raphe is unchanged by treadmill running. Medicine and Science in Sports and Exercise, 32(Suppl.5), S42.

Weiss, J.M., Bonsall, R.W., Demetrikopoulos, M.K., Emery, M.S., & West, C.H.K. (1998). Galanin: a significant role in depression? Annals of the New York Academy of Sciences, 863, 364-382.

Willner, P. (1995a). Animal models of depression: Validity and applications. In G. Gessa, W. Fratta, L. Pani, & G. Serra, Depression and Mania: From Neurobiology to Treatment, pp. 19-41. New York: Raven Press.

Willner, P. (1995b). Dopaminergic mechanisms in depression and mania. In F.E. Bloom & D.J. Kupfer, Psychopharmacology: The Fourth Generation of Progress, pp. 921-930. New York: Raven Press.

Wilson, W.M. & Marsden, C.A. (1996). *In vivo* measurement of extracellular serotonin in the ventral hippocampus during treadmill running. Behavioural Pharmacology, 7, 101-104.

Yoo, H.S., Tackett, R.L., Crabbe, J.B., Bunnell, B.N., & Dishman, R.K. (2000). Antidepressant-like effects of physical activity vs. imipramine: Neonatal clomipramine model. Psychobiology, in press.

Zhou, D., Grecksch, G., Becker, A., Frank, C., Pilz, J., & Huether, G. (1988). Journal of Neuroscience Research, 54, 109-116.

ANTIDEPRESSANT EFFECTS OF EXERCISE:

A QUANTITATIVE SYNTHESIS[1]

Abstract

Research conducted during the past three decades suggests that exercise can be an effective intervention for the prevention and alleviation of depressive symptoms. However, this area of research continues to be plagued by methodological flaws that limit inferences that can be drawn from single investigations. The purpose of this paper is to synthesize the results from epidemiological and intervention studies examining the effect of exercise on depression to summarize the existing data and provide direction for future research. A qualitative review of population-based studies was performed, and a summary of the epidemiological literature including cross-sectional and longitudinal research is provided. In addition, a quantitative review of the literature was conducted using meta-analytic methods to quantify the size of effects derived from controlled intervention studies. Moderator analyses were performed to examine features of the studies that could potentially influence the effect of exercise on depression. Fifty-eight studies of approximately 4,100 people yielded 88 effects. The mean effect was moderate, $d = -0.57$ (95% CI, -0.79 to -0.34), suggesting that individuals who participate in exercise programs exhibit a 1/2 SD reduction in depressive symptoms relative to individuals in comparison groups. Contrasts among levels of moderating variables indicated that effects were larger for studies that used clinical ratings of depression, included participants with elevated symptoms of depression or chronic health conditions, and lasted between eight and twenty-four weeks. Results from the qualitative review of literature and quantitative analyses indicate intervention features that may enhance the antidepressant effect of exercise and provide an empirical basis for future directions in research.

KEY WORDS: physical activity, fitness, mood disorder, mental health, meta-analysis

Antidepressant Effects of Exercise: A Quantitative Synthesis

As too much and violent exercise offends on the one side, so doth an idle life on the other…

Opposite to Exercise is Idleness or want of exercise, the bane of body and mind… the chiefe

author of all mischiefe, one of the seven deadly sinnes, and a sole cause of Melancholy.

~Burton, 1632

Depression is a widespread public health problem that poses a significant burden on society in terms of cost, loss of productivity, and mortality in addition to the detrimental effects on quality of life (Greenberg, Stiglin, Finkelstein, & Berndt, 1993). In terms of life years lost to disability and premature mortality, major depression was ranked as the fourth leading international disease burden in 1990 according to the Global Burden of Disease Study and is predicted to rise to second by the year 2020 (Murray & Lopez, 1996). Despite the personal and public burden of depression, depressive disorders are widely undertreated, and it has been estimated that only 10% of depressed individuals receive adequate treatment (Hirschfeld et al.,1997). Thus, study of therapeutic modalities that may provide an accessible, inexpensive, and effective alternative to traditional antidepressant therapies is needed.

Results from epidemiological and intervention research suggest that exercise can produce meaningful reductions in depressive symptoms for many individuals. During the past decade, qualitative reviews supporting the mental health benefits of exercise have been presented by authors representing various scientific disciplines including psychology (e.g., Byrne & Byrne, 1993; Tkachuk & Martin, 1999), psychiatry (e.g., Martinsen, 1990,1994), family medicine (e.g., Nicoloff & Schwenk, 1995; Paluska & Schwenk, 2000), and exercise science (e.g., Morgan, 1994; Dishman, 1998; O'Neal, Dunn, & Martinsen, 2000). In addition, quantitative syntheses of the literature using meta-analytic methods demonstrate an antidepressant effect of exercise. For example, North, McCullagh, & Tran (1990) conducted a meta-analysis of 80 studies published through June 1, 1989 that examined the effect of exercise on depression. The overall effect size (d) was –0.53 indicating that, on

average, individuals in exercise conditions exhibited a 1/2 SD greater reduction in depression scores than individuals in comparison groups. Results were similar from a subsequent meta-analysis by Craft & Landers (1998) that examined the effect of exercise interventions in persons with clinically diagnosed depressive disorders. A larger overall effect size (d) of -0.72 was obtained, indicating that depression scores were reduced by 3/4 SD in individuals who participated in exercise relative to control groups.

A primary objective of this review is to provide a current summary of results from intervention studies using meta-analytic procedures to synthesize data from controlled studies investigating the effects of exercise training on symptoms of depression. A second objective of this paper is to provide a qualitative review of longitudinal and cross-sectional studies that have examined the relationship between physical activity and depression in the population, as most reviews of the literature have given relatively little attention to epidemiological research except for highlighting a handful of population-based studies that have gained widespread attention. Therefore, the current paper provides an update of recent research examining the effect of exercise on symptoms of depression and expands upon conclusions of previous reviews by presenting both epidemiological and experimental perspectives. In addition to providing a summary of the evidence for the antidepressant effects of exercise, limitations in the current literature and recommendations for future research are discussed.

Epidemiological Research: a qualitative synthesis

One line of evidence for the antidepressant effects of exercise is derived from population-based studies that demonstrate an inverse relationship between the level of physical activity participation and the degree of depressive symptomatology. Because causal inferences derived from epidemiological research are limited and studies vary in the application of statistical procedures, a quantitative synthesis of population data could not be performed. However, consideration of these data is important as they provide a basis for intervention research and contribute to the understanding of the practical and

clinical significance of this issue. A summary of population-based studies examining physical activity and depression published through June 1, 2000 that we were able to locate is presented in Table 1.

Although results vary both between and within studies, the totality of the evidence from cross-sectional and longitudinal studies suggests that individuals who participate in regular physical activity exhibit lower levels of depression relative to sedentary individuals (Figure 1). However, scientific evaluation of data obtained from epidemiological research should include considerations of strength of association, temporal sequence, alterability, independence, consistency, and dose-response (Mausner & Kramer, 1985).

Strength of Association. Of the twenty-five population studies we reviewed, twenty-two studies reported a significant negative relationship between physical activity and symptoms of depression for at least one analysis included in the study. For example, Data collected from a 23-27 year longitudinal study of approximately 10,000 male Harvard alumni demonstrated a significant reduction in risk for depression in men who were physically active compared to men who were inactive (Paffenbarger, Lee, & Leunge, 1994). Information regarding physical activity habits such as daily distance walked, daily stair climbing, and sports/recreational activity was obtained in 1962 or 1966 and 1977 using self-report questionnaires. Depression was assessed by self-report of physician-diagnosed depressive disorder through 1988. An approximate 30% reduction in relative risk for depression was observed in men who typically spent 3 or more hours per week in sports play, or who expended 2,500 or more kcal/wk in physical activity (Paffenbarger et al., 1994).

In contrast, the Precursors Study which examined 752 former medical students who attended medical school at Johns Hopkins University between 1948 and 1964, failed to show an association between physical inactivity and depression (Cooper-Patrick, Ford, Mead, Chang, & Klag, 1997). Baseline assessments were conducted in 1978, and subjects

were followed until 1993. Incidence of depression from 1979 through 1993 was determined by self-report of clinical depressive episodes and review of medical records, and level of physical activity in 1978 was assessed by self-report of the frequency of exercise to a sweat during a typical week. No differences in risk for depression were observed between individuals who reported exercising 3 or more times per week and sedentary individuals. However, as with many population-based studies, the generalizability of these results may be limited by the failure to use a standard assessment of physical activity

Two recent reports from cross-sectional studies also support an association between physical activity and reductions in depression. First, data from the Study of Osteoporotic Fractures revealed that elderly women reporting greater weekly energy expenditure also reported fewer symptoms of depression (Yaffe, Blackwell, Gore, Sands, Reus, & Browner, 1999). Similarly, subjects from the Finnish cardiovascular risk factor survey of 3,403 men and women aged 25 to 64 reporting an exercise frequency of 2 to 3 times per week had significantly lower depression scores compared to sedentary individuals (Hassmen, Koivula, & Uutela, 2000). However, as with other cross-sectional data, results must be interpreted cautiously because the inability to establish a temporal sequence prevents inferences of causality (Mausner & Kramer, 1985).

Temporal Sequence. Because a reduction in physical activity may be a symptom of depression, an appropriate temporal sequence is essential for demonstrating physical inactivity to be a risk factor for depression. For example, data from the Alameda County Study permit an evaluation of temporal sequence, as multiple measures of both physical activity and depression were conducted over a time period that permitted natural progression of depressive disorders (Camacho, Roberts, Lazarus, Kaplan, & Cohen, 1991). Data were collected in 1965, 1974, and 1983. Depression was assessed by self-report of current symptoms, and subjects were classified as low, medium, or high active based on frequency and intensity of self-reported leisure activity. After adjusting for age, health, and other psychosocial variables, a significant relationship was found between physical activity in

1965 and self-reported depression in 1974. Men and women reporting low levels of physical activity had a 70% increase in risk of depression compared to subjects classified as highly active. Change in physical activity level between 1965 and 1974 and depression in 1983 was also examined, and results suggested that subjects who altered activity level also incurred a change in risk for depression.

However, other studies that observed a significant relationship between exercise and depression upon cross-sectional analysis, failed to find an association upon examination of longitudinal data. Weyerer (1992) examined data collected from the Upper Bavarian Field Study, an investigation of 1,536 people aged 15 and older randomly selected from the community. Baseline data were collected in 1975-1979, and 87% of subjects were re-assessed 5 years later. Depression was diagnosed by psychiatrist-administered clinical interviews according to International Classification of Diseases (ICD) criteria, and participants were classified by current level of self-reported physical activity. After adjusting for sex, age, social class and physical disorders, cross-sectional analysis of baseline data revealed an odds ratio for depression of 3.15 for individuals reporting no exercise compared to those reporting regular exercise. However, upon longitudinal analysis, physical inactivity did not increase the risk of developing depression 5 years later.

Among studies reporting results from cross-sectional data only, thirteen studies reported a significant inverse relationship between physical activity and depression whereas two studies did not find an association. Of the four studies that reported only prospective data, three studies demonstrated a significant negative association. Six studies reported both cross-sectional and longitudinal data. Three studies demonstrated a significant negative relationship in both analyses, one study exhibited mixed results within both analyses, one study observed a significant effect for only cross-sectional analyses, and one study found a significant effect for only longitudinal data (Table 1).

Alterability. One advantage of a prospective study in addition to establishment of temporal sequence is the ability to determine alterability in which naturally occurring

increases or decreases in physical activity are associated with subsequent change in depression scores. For example, Lampinen, Heikkinen, & Ruoppila (2000) examined the relationship between physical activity and depression in 663 older adults. Assessments were conducted by self-report of current physical activity and depressive symptoms in 1988 and 1996. Subjects were classified into 3 groups according to the level of exercise intensity by the extent to which individuals reported performing necessary chores only, walking, or strenuous exercise. Gender, age, and chronic somatic conditions were controlled in the regression analyses. For subjects who engaged in only chore-related physical activity at baseline, individuals who remained less active were more likely to exhibit elevated symptoms of depression at the follow-up assessment. Individuals who were regular walkers at baseline but reduced physical activity were at greater risk for depression at the follow-up assessment relative to individuals who increased activity. However, individuals who initially participated in strenuous exercise and reduced their physical activity level did not exhibit a subsequent increase in risk for depression. Though the results support the alterability of risk for depression with change in physical activity, small sample size for each level of analysis is a limitation. Of the four studies that examined depression scores after a change in physical activity, all exhibited evidence of alterability to varying degrees, but additional studies are needed to fully evaluate this effect.

Independence. A characteristic limitation of epidemiological research is the potential impact of confounding variables, as there is no random assignment of subjects to groups. Thus, determining the independence of an effect is an important methodological concern, and control for variables that have been identified as common risk factors for depression is critical for determining whether there is an independent effect of physical activity. For example, gender is often associated with depression, and women under age 45 are at highest risk for developing a depressive disorder (Sargeant, Bruce, Florio, & Weissman, 1990). Variables such as social support, level of education, and health status also may influence depressive symptoms (Anonymous, 2000) and should be controlled in

analyses. Analyses of Alameda County Study data yielded an age-adjusted relative risk for depression of 2.48 for men and 2.88 for women who reported low levels of physical activity. After including adjustments for health, socioeconomic status, social support, life events, anomy, alcohol intake, smoking, and relative weight, the association between physical inactivity and depression was attenuated though still significant with relative risks of 1.76 and 1.70 for males and females, respectively (Camacho et al, 1991).

Considerations should also be given to population characteristics that could obscure or complicate interpretation of effects. For example, symptoms of depression include insomnia, fatigue, and loss of concentration (Weissman et al., 1996), all of which may be associated with aging and/or disease. Because approximately one-third of the studies was conducted on older adults and community samples which included participants with chronic diseases, characteristics associated with aging or illness could influence interpretations of results (O'Connor, Aenchbacher, & Dishman, 1993; Palinkas, Wingard, & Barrett-Connor, 1990). Nevertheless, most studies reporting unadjusted and adjusted models have demonstrated a smaller but significant relationship between physical activity and depression after controlling for potential confounding variables (Camacho et al., 1991; Farmer, Locke, Moscicki, Dannenberg, Larson, & Radloff, 1988).

Consistency. An advantage of epidemiological research in the investigation of the antidepressant effect of exercise is the ability to examine large numbers of people representing various demographic groups. Studies have included male and female participants of diverse age groups (10 years and older) representing different countries (United States, Canada, Germany, Japan, and Finland). However, research has yielded inconsistent results both between and within studies. For example, the National Health and Nutrition Examination Survey (NHANES I) examined physical activity and depressive symptoms in 1,497 men and women aged 25-77 years (Farmer et al., 1988). Current level of depressive symptoms and physical activity were assessed by self-report, and assessments were conducted in 1971-1975 and 1982-1984. After adjusting for demographic and

psychosocial variables, cross-sectional analysis of baseline data revealed significant associations between little or no recreational physical activity and depressive symptoms for white and black males and white females, but the relationship was not significant for black females. Additionally, among subjects who did not exhibit elevated depression at baseline, little or no recreational activity was a significant predictor of depressive symptoms only in white women. However, results by race should be interpreted cautiously as the sample size was small for both black men (N=64) and women (N=79).

Though inconsistencies have been exhibited both between and within studies, there do not appear to be any systematic differences (i.e., males vs. females; young vs. old) that would limit the generalizability of results. In addition, varied definitions of physical activity including walking, lifestyle and occupational activity, sports participation, and energy expenditure have yielded significant results. Because most studies have relied on self-report questionnaires to measure symptoms of depression, it is not known whether the type of depression assessment influences results. However, this issue requires further investigation, as two of the four studies that measured clinically defined depressive disorders failed to show a significant relationship.

Dose-Response. Additional research is also needed to determine whether the association between exercise and depressive symptoms is best described as an inverse dose-response relationship or a threshold effect. For example, results from the study of male Harvard alumni suggest a does-response relationship between physical activity and reductions in symptoms of depression. Whereas men who reported expending 1,000-2,499 kilocalories per week had a reduction in risk for depression compared to men expending less than 1,000 kilocalories per week, those with an energy expenditure of 2,500 kilocalories or more per week had an even greater reduction in risk (Paffenbarger et al., 1994). Data from a study of elderly women also support a dose-response relationship, as women who reported the highest levels of energy expenditure exhibited fewer symptoms of depression (Yaffe et

al., 1999). A similar relationship was found between physical activity level and depression scores among European college students (Steptoe et al., 1997).

In contrast, data from other studies suggest a threshold effect of exercise in which both moderate and high levels of physical activity are associated with lower levels of depression, but there is no additional benefit of high compared to moderate activity. For example, Foreyt, Brunner, Goodrick, St. Jeor, & Miller (1995) classified individuals by change in physical activity over a 4-year period. Participants who reported moderate increases in activity exhibited larger reductions in depressive symptoms relative to individuals reporting the largest increase in physical activity, although both groups reported improvements in depression. Data from a longitudinal study of elderly participants suggests that moderate walking is sufficient to protect against increases in depression. Though increases in depressive symptoms were observed for individuals reporting a decrease in physical activity, both maintenance of walking and an increase in physical activity appeared to protect against increases in depression (Lampinen et al, 2000).

The dose-response relationship between physical activity and depression is difficult to interpret in the epidemiological literature because most studies have classified participants into two or three activity levels or used regression techniques to analyze data. Research examining the systemic health benefits of exercise has demonstrated that individuals who increase activity from the lowest category incur significant health benefits (Blair, Well, Weathers, & Paffenbarger, 1994), and further research is needed to determine whether a similar effect exists for mental health. Because most individuals prefer to exercise at a low or moderate intensity (Dishman, Farquhar, & Cureton, 1994) and current guidelines for exercise prescription recommend a broad range of exercise intensities (ACSM, 1998), this issue is of clinical significance for the development of recommendations to promote mental health in the general population.

Summary and Recommendations

Despite inconsistencies among studies, population-based research supports an inverse association between levels of physical activity and symptoms of depression. However, methodological limitations common to epidemiological literature impede interpretation of results. First, the majority of population data is derived from studies in which mental health is not the primary focus (e.g., cardiovascular risk factor, aging, etc.), and the purpose of the study generally dictates the variables that are measured and reported as well as the statistical methods employed. In addition, several studies that have measured both physical activity and depression did not report the data in a manner that permits examination of the relationship (e.g., Brill, Kohl, & Blair, 1992; Ruuskanen & Parkatti, 1994; Strawbridge, Cohen, Shema, & Kaplan, 1996). Thus, there is a selectivity bias in the reporting of results based on the purpose of the study.

Second, if the aim of the study is to evaluate the relationship between physical activity and depression, then research should be conducted on high-risk populations (e.g., female, young adult, positive family history of depression, previous depressive episode, etc.). Moreover, the singular contribution of future epidemiological studies, beyond that of experimental research, is the determination of the role of physical activity in the prevention of depression as it naturally occurs in the population.

Third, further examination of the dose-response relationship between physical activity and depression is needed. The identification of a dose-response gradient would provide additional evidence for a causal effect of physical activity on reductions in depression. In addition, resolution of this issue is required to establish evidence-based exercise prescription guidelines for the promotion of both physical and mental health.

Finally, epidemiological research should employ valid measures of both physical activity and depression that are appropriate for the population of interest. Because population-based studies attempt to gather as much information as possible while minimizing the burden on participants and investigators, measures are often modified

versions of validated assessments or may just consist of a few untested items. Therefore, when the psychometric properties of the instruments are unknown, the validity of the inferences derived from the measures is also questionable. At minimum, validated self-report instruments should be used to measure physical activity and depression, and additional studies are needed that use objective measures to assess physical activity and that apply clinical criteria for diagnosis of depression. From an epidemiological perspective, there are three possible explanations to account for the inverse relationship between physical activity and depression: (1) exercise prevents and/or reduces symptoms of depression; (2) depressed individuals are less likely to exercise; or (3) exercise and depression are both associated with a third variable which is responsible for the observed effects (Stephens, 1988). Thus, randomized controlled intervention studies are needed to determine the nature of the relationship between exercise and depression to establish causality.

Intervention Research: a quantitative synthesis

Results from meta-analyses by North et al. (1990) and Craft & Landers (1998) suggest a moderate effect of exercise training in reducing symptoms of depression for most individuals. Although meta-analytic procedures will never yield an exact estimate of the actual population effect, a quantitative synthesis does provide a systematic method to examine an area of research and identify directions for future study (Ioannidis & Cappelleri, 1998). We recognize that conclusions derived from the aggregation of effects obtained from studies of diverse samples, using varied exercise interventions and depression measures are not a substitute for large controlled experiments that examine the effect of exercise interventions in individuals with a clinical diagnosis of depression. To date, few clinical trials have been conducted in this area. Therefore, the current meta-analysis was conducted to provide a revised summary of the antidepressant effects of exercise from the existing literature and present evidence for further justification of large randomized controlled trials which are considered to be the gold standard for determining

the efficacy of interventions in a clinical setting (LeLorier, Gregoire, Benhaddad, Lapierre, & Derderian, 1997).

Moreover, the present review differs from earlier meta-analyses in several ways to expand upon previous conclusions. First, we attempted to control for natural remission of depression by including in the overall analysis only studies that permitted the calculation of an effect size relative to a comparison group. Second, we excluded studies that measured symptoms of depression after an acute bout of exercise to avoid confounding the measurement of transient mood states with an enduring depressive condition. Third, although the use of multiple dependent measures permits the calculation of more than one effect size from the same group of people, we excluded redundant effects to avoid artificial inflation of effect sizes for a more accurate estimate of the population effect. Fourth, because studies vary in the use of exercise interventions, comparison groups, and sample characteristics, we expected effects to be heterogeneous, and a random effects model was used for analyses (Hedges & Olkin, 1985). Finally, in the years since prior meta-analytic reviews were conducted, additional studies have been published that include more representative samples (e.g., elderly), employ varied exercise interventions (e.g., resistance training), and utilize clinically meaningful comparison groups (e.g., pharmacological treatment).

Methods

Eighty-three studies published from 1970 through December 31, 1999 were located by searches of literature published in the English language using Medline, PubMed, PsychINFO, ERIC, and Current Contents computer searches with *exercise*, *physical activity*, *fitness*, *depression*, *psychological*, *psychiatric*, and *mental health* as key words. Reference lists from published articles were used to supplement computer searches. Dissertations, master's theses, and studies of acute exercise were excluded from analysis. Criteria for including a study consisted of the following: (1) The dependent variable was a measure of depression or depressive symptoms. (2) The independent variable was an exercise

intervention, physical activity measure, or standard measure of physical fitness that served as a surrogate measure of physical activity. (3) The dependent variable was quantified in a manner that allowed an effect size to be calculated from means and standard deviations, correlation coefficients, graphs, t-tests, F-tests with a single *df*, and exact p-values. (4) The design included a non-exercise comparison group. Eighteen located studies were excluded because they failed to use a standard measure of depression, reported results in a manner that did not allow the calculation of an effect size, implemented a combined intervention that prevented evaluation of the independent effect of exercise, or used an exercise comparison group (Appendix 1).

A quantitative synthesis was performed using Meta 5.3 (Schwarzer, 1991), DSTAT 1.10 (Johnson, 1993) and SPSS Windows version 9.0 (SPSS, Inc., Chicago, IL) statistical software. When possible, effect sizes were calculated by subtracting the mean change for a comparison group from the mean change for the experimental group and dividing the difference by the initial pooled standard deviation. Based on guidelines suggested by Cohen (1988), effect sizes of 0.2, 0.5, and 0.8 were considered to be small, moderate, and large effects, respectively. Composite effect sizes (d) were obtained using the random effects model to aggregate effect sizes because effects were expected to be heterogenous (Hedges & Olkin, 1985). Homogeneity of each effect was tested using a random effects model of variance (Hunter, Schmidt, & Jackson, 1982), and an effect was judged heterogeneous if sampling error was less than 75% of the observed variance. Factors that were hypothesized to moderate the effectiveness of exercise interventions in reducing symptoms of depression were examined by coding sample characteristics, research design, intervention characteristics, and outcome measures (Table 2). Levels of a moderating variable with fewer than 5 effects were collapsed or omitted to minimize sampling error. A one-way analysis of variance (ANOVA) was performed for each moderating variable to determine whether there were significant differences among levels of moderators, and focused contrasts were subsequently conducted to identify features that might account for variability

in mean effect size. Moderators with more than two nonordinal levels were dichotomized based on the results of the contrasts, and a multiple linear regression analysis was conducted for significant (p<.10) moderating variables to determine whether the variables had an independent influence on the overall estimate of effect size.

Two hundred sixty-three effects were retrieved from 92 studies involving approximately 17,500 people. Multiple effects were obtained for studies that reported separate results for gender, included more than one exercise intervention, provided results for multiple outcome measures, allowed calculation of effect sizes both within the exercise condition and between the exercise and comparison groups, and presented results obtained from multiple time points. However, only the primary effect size(s) were used for the main analyses. When a study reported multiple measures of depression using the same methodology (e.g., self-report questionnaires), the mean of the effects was used in the overall analysis. In addition, if a study used both clinician-rated and self-report measures, only the clinical rating was used in the main analyses to avoid artificially inflating an effect by redundant inclusion of data. Because the purpose of the quantitative synthesis is to examine the effectiveness of exercise in reducing symptoms of depression, only the 58 controlled studies that used a comparison group were included in the primary analyses for a total of 88 effects from 4,126 people. Within-subject and redundant effects were included for appropriate moderator analyses. Twenty-seven follow-up effects obtained from 9 studies (Appendix 2) were analyzed separately.

Results

A stem-and-leaf display for the 88 primary effects is presented in Figure 2. The distribution of effects was negatively skewed (-1.94, SE=.26) and leptokurtic (3.56, SE=.51). The mean overall effect of exercise was moderate, d= -0.57 (95% CI, -0.79 to -0.34).

Moderating Variables

      Study design and measurement. Effect sizes obtained from controlled studies using a non-randomized design did not differ from studies using a randomized design (p=.54). The ANOVA for comparison group was not significant (p=.64). However, because few studies used psychotherapy or pharmacotherapy as control conditions, these levels were eliminated from the moderator analyses. No significant differences were observed for recruitment source (p=.12). The ANOVA for depression measure was significant ($F_{1,92}$=8.80, p=.004, η^2=.09). Effect sizes from studies that used clinical ratings of depressive symptoms were larger than effects obtained from self-report questionnaires.

      Subject characteristics. Effect sizes did not differ by gender (p=.54) or age (p=.69). To minimize sampling error, categories of disease were collapsed for analysis, and effect sizes obtained from studies that included participants with a chronic health condition were significantly larger than effects obtained from studies of healthy individuals ($F_{1,81}$=5.43, p=.02, η^2=.06). The ANOVA for mental status approached significance ($F_{1,82}$=3.18, p=.08, η^2=.04), and studies that included subjects with elevated levels of depressive symptoms at baseline yielded larger effects than studies of mentally-healthy individuals.

      Exercise features. No significant differences were found among levels of exercise mode (p=.27), intensity (p=.62), frequency (p=.80), or duration (p=.86). There were also no significant differences in effects obtained from studies that administered the exercise intervention in groups or individually (p=.20). Effects also were not different across exercise settings (p=.37). The ANOVA revealed significant differences among effects obtained from supervised versus unsupervised exercise sessions ($F_{1,86}$=5.81, p=.02, η^2=.06) with studies using unsupervised exercise exhibiting larger effects. Effects also differed among studies by length of the intervention, and contrasts indicated a nearly significant quadratic trend (t_{88}=1.88, p=.06). Effect sizes from interventions that were

between 8 and 24 weeks in length were larger than effect sizes from interventions of less than 8 weeks or more than 24 weeks.

Multiple Regression Analysis. A multiple linear regression model including health status (healthy vs. chronic disease; $\beta=-.32$, t=4.06, p=.001), mental health status (normal vs. depressed; $\beta=-.21$, t=2.20, p=.03), exercise measure (intervention or assessment vs. self-report; $\beta=-.27$, t=2.65, p=.01), exercise supervision (supervised vs. unsupervised, $\beta=-.24$, t=2.4, p=.02), and depression measure (clinical rating vs. self-report; $\beta=.24$, t=2.49, p=.02) was significantly related to effect size d ($F_{5,77} = 7.81$, p<.001, adjusted $R^2=.29$).

Discussion

Results from this quantitative synthesis suggest that exercise interventions for reducing depression have a moderate effect that is statistically different from zero. These findings are in agreement with those obtained from prior analyses by North et al. (1990) and Craft and Landers (1998) who reported an overall effect size (d) of -0.53 and -0.72, respectively. The effects were heterogeneous, indicating that more than one population effect was estimated by the studies. The moderator analyses of effects suggests that some features of the studies may yield larger effects and provide direction for future research. However, low statistical power resulting from a limited number of effects and heterogeneity within levels of moderators provided weak tests of the variables in many instances. Because not all studies reported information on the moderators included in the analyses, it was not possible to conduct statistically powerful analyses of interactions. Therefore, well-designed studies that experimentally manipulate exercise features and control for subject characteristics are needed to accurately identify moderators of the effect of exercise on depressive symptoms.

Most analyses pertaining to study design failed to reach statistical significance. However, studies that used clinical ratings to assess depression exhibited larger effects than studies that used self-report questionnaires. Though results were not significant,

studies characterized by a nonrandomized design and lack of comparison group tended to produce larger effects than randomized, controlled studies. Types of comparison groups could not be analyzed due to the small number of independent effects within each level. The two studies that used a pharmacotherapy comparison (Blumenthal et al., 1999; Wearden et al.,1998) yielded zero or slightly positive effects, supporting the efficacy of exercise relative to a drug intervention in reducing depressive symptoms. Examination of effects from studies using psychotherapy control conditions suggest greater efficacy of exercise, and though such observations give credence to the effect of exercise in reducing depression, the results also call into question the success of implementing the comparison condition, as psychotherapy is a proven antidepressant intervention. In order to demonstrate that exercise is effective for reducing symptoms of depression, features of the intervention that are unrelated to the exercise stimulus (e.g., group dynamics, social support, placebo effects) must be controlled.

Results of the present analyses suggest that the effects of exercise do not differ by age or gender. However, few effects were available from adolescent populations to allow adequate examination of age. Though effect sizes tended to be smaller for studies of adolescents, a recent review examining correlates of physical activity in youth supports an inverse relationship between exercise and depression in younger populations (Sallis, Prochaska, & Taylor, 2000). As observed in previous analyses (North et al., 1990), studies of participants who exhibited chronic health conditions produced larger effects than studies of apparently healthy individuals. Effect sizes from studies that recruited subjects identified as exhibiting higher levels of depression tended to be larger than effects obtained from studies of nondepressed individuals. However, only four studies used clinical interviews to screen participants, and eight studies included participants with elevated levels of self-reported depression. Thus, the small number of effects from studies of depressed individuals likely contributed to the inability to detect a significant difference according to mental health status.

Generally, features of the exercise intervention did not significantly influence size of the effects. However, the number of effects per level of each moderator was often too small to permit a powerful analysis of the relationship among categories. For example, the data suggest that resistance training is at least as effective as aerobic exercise in reducing depression (Doyne et al., 1987; Martinsen et al., 1989), but only seven independent effects for strength training were available for analysis. The data also suggest that low intensity, flexibility-related exercise does not produce significant reductions in depression, but because few effects are available, interpretation is limited. Furthermore, studies have used both resistance training and low intensity exercise as control conditions, preventing the inclusion of the effects in the overall analysis and precluding the evaluation of these conditions as antidepressant interventions. Given the current popularity of resistance training and the success of lifestyle physical activity interventions, this omission represents a pressing research need.

As noted in the review of the epidemiological literature, the failure to adequately manipulate and quantify the exercise stimulus prevents examination of the dose-response relationship between exercise and reductions in depression. It remains to be determined whether there is a threshold for antidepressant effects or whether the degree of improvement in depression is dependent upon amount of exercise in terms of intensity, frequency, type, and/or duration. From a pharmacological perspective, it is possible that there is a dose-dependent treatment response to exercise until the exercise "dose" reaches an optimal level at which there is no benefit of additional exercise. The demonstration of dose-response relationship that follows a pharmacological model would provide additional evidence for causality. The issue of appropriate dose to enhance the efficacy of exercise in alleviating depression can be resolved by experimental studies that manipulate discrete features of exercise stimulus while controlling for other characteristics of the exercise.

Further investigation is also needed to determine the influence of length of training on the effects of exercise. This variable was excluded as an independent predictor due to its non-linear relationship with effect size. Though results from previous meta-analyses (North et al., 1990) suggest that there is a dose-response relationship between intervention length and reductions in depression scores, results from the present analysis suggest that effects are reduced for studies greater than six months in length. However, to adequately evaluate the effect of length of training, repeated measure designs are needed. Only eight studies included in the overall analysis assessed depression at multiple time points during the intervention, and nine studies included follow-up assessments upon conclusion of the intervention. Repeated measurement of depressive symptoms is important for monitoring improvements in depressive symptoms to ensure participant safety and is required to evaluate treatment response to exercise and enable comparisons with standard antidepressant therapies. Additional research using follow-up measures is also needed to evaluate the persistence of effects after completion of the exercise program.

Pre-experimental cohort designs were excluded from our overall quantitative analysis to prevent the confounding of the effects of exercise with the natural remission of depressive symptoms with the passage of time. However, this resulted in the elimination of fifteen of the studies retrieved. Studies that used a form of exercise as a control condition were also excluded from the overall analyses, as it has not been demonstrated that antidepressant effects of exercise are specific to type of physical activity. Fifteen additional studies were excluded because data were incomplete or reported in a manner that prevented the calculation of an effect size. Also, eleven studies were omitted because the results were reported elsewhere or the design did not enable interpretation of the effect of exercise. Excluded studies are presented in Appendix 1.

Though all studies with non-exercise comparison groups were included in the analysis, few studies had equivalent, randomized control groups. Furthermore, studies

included in the analysis often failed to provide detailed information in the reporting of procedures and/or results making it difficult, and in some cases impossible, to generate effect sizes. In addition to inadequate reporting of results, study characteristics were often poorly described, limiting the effectiveness of our moderator analyses.

Though useful in summarizing the literature, meta-analytic techniques have inherent limitations. A primary disadvantage is the aggregation of studies of varying degrees of scientific quality. Unfortunately, several of the studies with the best designs and methods could not be included in the analyses due to limitations with the reported statistics. For example, Martinsen and colleagues (1985, 1989a, 1989b) conducted a series of studies that examined the effect of exercise interventions on depression in hospitalized psychiatric patients. In the initial study, patients diagnosed with major depression were randomly assigned to aerobic exercise or occupational therapy in addition to their standard treatment protocol. The interventions were conducted in groups that met for 1 hour, 3 times a week. After 9 weeks of treatment, the exercise group exhibited significantly greater reductions in depressive symptoms relative to the control group (Martinsen, Medhus, & Sandvik, 1985). After discharge from the hospital, these patients were followed for 1 to 2 years. Approximately 90% of patients assigned to the exercise group continued regular exercise. Although not statistically significant, there was an inverse relationship between level of physical activity and depression scores at follow-up. In addition, patients who participated in the exercise intervention subjectively evaluated the exercise program as the most important component of their inpatient treatment (Martinsen & Medhus, 1989a). Martinsen, Hoffart, & Solberg (1989b) also compared aerobic (walking/jogging) and nonaerobic (strength, flexibility, relaxation) exercise in the treatment of clinical depression in psychiatric inpatients. The interventions were administered in small groups, and individuals trained for 1 hour, 3 times a week for 8 weeks. At the conclusion of treatment, both groups exhibited significant reductions in depression scores, but there were no differences between groups.

Perhaps the most convincing evidence for the effectiveness of exercise in the treatment of depression emerged from a recent randomized controlled clinical trial that compared exercise and antidepressant medication (Blumenthal et al., 1999). Approximately 150 older patients diagnosed with major depression were randomly assigned to one of 3 groups: aerobic exercise (walking/jogging, 3 times/week, 30 minutes), antidepressant medication (sertraline hydrochloride, i.e., Zoloft), or exercise plus medication. Although the medication group exhibited the fastest initial response to treatment, all groups exhibited significant reductions in depression scores, but there were no significant differences between groups after the 16-week intervention period. This study was excluded from the quantitative analyses because the zero effect size calculated from the comparison with a proven antidepressant therapy supports the efficacy of exercise in treating depression. In addition, it can be argued that a primary goal of meta-analysis is to provide a summary of the literature to provide a rationale for more rigorous clinical trials (LeLorier et al., 1997). The results of this clinical trial are encouraging given the stringent research design, particularly the use of standard diagnostic criteria and clinically meaningful comparison conditions. However, a placebo condition was not included in the study to control for attention or spontaneous recovery, and additional randomized clinical trials are needed to confirm these findings and identify intervention features that moderate the antidepressant effects of exercise.

Over twenty years have passed since experimental studies first demonstrated an antidepressant effect of exercise in individuals with a clinical diagnosis of depression (Greist, Klein, Eischens, Faris, Gurman, & Morgan, 1978), and the preponderance of evidence including anecdotal reports, epidemiological research, intervention studies, qualitative reviews of literature, and meta-analyses suggests that exercise can result in statistically significant and clinically meaningful reductions in symptoms of depression. Furthermore, reductions in depressive symptoms are often comparable to standard antidepressant therapies (Blumenthal et al. 1999; Fremont & Craighead, 1987; Greist et

al., 1978), and results from quantitative reviews of exercise studies are similar to meta-analyses of standard pharmacological and psychotherapy interventions. For example, Greenberg, Bornstein, Zborowski, Fisher, & Greenberg (1994) conducted a meta-analysis examining the efficacy of the antidepressant fluoxetine in double-blind, placebo-controlled clinical trials and found a moderate overall effect (d = -0.40). Also, a recent meta-analysis of studies using cognitive psychotherapy in treating patients with mild to moderate depression demonstrated an effect size (d) of -0.82 compared to wait-list or placebo comparison groups (Gloaguen, Cottraux, Cucherat, & Blackburn, 1998). However, comparisons of meta-analytic results should be approached cautiously as the quality of studies and decisions involved in the analyses could influence the accuracy of the effect size estimation (Juni, Witschi, Bloch, & Egger, 1999). Furthermore, effect sizes derived from meta-analyses may not accurately predict results of large randomized controlled trials implemented in a clinical setting (LeLorier et al, 1997).

Although it is inappropriate to view an effect size value as a precise representation of the efficacy of an intervention in the population, meta-analytic summaries can serve as useful a guide for prediction of intervention effects and direction of future research (Ioannidis & Cappelleri, 1998). However, the utility of qualitative reviews of literature should not be abandoned for quantitative syntheses (Bailar, 1997). Ideally, results from quantitative and qualitative reviews of literature are conducted in tandem and produce complementary results. The present review of the literature, both quantitative and qualitative, supports an antidepressant effect of exercise, and conclusions are generally consistent with prior analyses. Thus, randomized controlled trials examining the efficacy of exercise interventions are warranted. Further research is required to determine whether the estimated effects of exercise are reliably and meaningfully produced in a clinical setting.

Given the tremendous burden that depression poses to society, if exercise is indeed effective for the prevention and treatment of depression, the potential public

health impact would substantial. The lack of utilization of standard antidepressant interventions, the absence of uniformity in treatment response, and the health care costs associated with pharmacotherapy and psychotherapy call for examination of alternative therapies for treating depression. In addition to its apparent effectiveness in reducing symptoms of depression, exercise is affordable and widely accessible with proven systemic health benefits. Furthermore, in contrast to other antidepressant treatments, exercise interventions are ideally continued throughout the life span. However, despite abundant evidence that supports an antidepressant effect of physical activity and recognition of the established systemic health benefits associated with exercise, prominent mental health organizations have yet to recognize exercise as a viable alternative or adjunctive treatment for depression (Anonymous, 2000). In order to promote exercise as a sanctioned treatment for depressive disorders, researchers must raise the standard for research design and evolve toward a clinical trial paradigm that will meet the evaluative demands put forth by public health officials (LeLorier et al.,1997). Based on our findings from both the qualitative review of literature and meta-analytic procedures we offer the following recommendations for future research:

(1) Interventions should use established diagnostic criteria to identify depressed individuals for subject recruitment, as a primary limitation of this body of literature is the failure to study depressed individuals. The overall effects of exercise are likely attenuated by this design limitation, because mentally healthy individuals are far less likely to exhibit significant reductions in depression. Furthermore, results from studies of normal individuals cannot be generalized to clinical populations. When possible, it is also advantageous to identify depression subtype and the presence of comorbid psychological disorders because features of the depressive disorder may influence the effectiveness of interventions and predict treatment response (Anonymous, 2000).

(2) Researchers should determine intervention effects using validated assessments specifically designed to measure depression, and measures should be appropriate to the

population studied. When possible, clinical diagnostic interviews should be used in conjunction with self-report measures to enhance reliability and specificity of symptom assessment.

(3) Investigators should use fully randomized designs and include appropriate comparison groups. At minimum, studies should control for natural remission of depressive symptoms and placebo effects. Thorough descriptions of the components of the control condition as well as the rationale for the comparison should be included. In order to establish causality, large randomized clinical trials using meaningful comparison conditions are needed.

(4) Various features of physical activity (e.g., mode, intensity, duration, frequency, intervention length) should be manipulated to determine the stimulus required to produce antidepressant effects. The use of valid assessments of physical activity and appropriate measures of muscular strength or cardiorespiratory fitness are needed to verify the effectiveness of an exercise intervention in achieving the desired physiological stimulus. Improved descriptions of intervention characteristics and experimental confirmation are therefore needed to identify the exercise stimulus required for meaningful reductions in depression and to establish guidelines for exercise prescription in depressed populations.

(5) Authors should report detailed information on samples, interventions, measures, and statistical analyses. Results should be presented in a form that permits the calculation of an effect size, including means, standard deviations, and sample size before and after the intervention for all groups studied.

(6) Studies should include repeated measures of depressive symptoms to determine of the course of effects and to permit comparisons with standard antidepressant therapies. Follow-up measures of depression and physical activity should also be performed to evaluate persistence of effects after the conclusion of the intervention.

(7) Scientists should explore objective physiological measures that can be used to confirm results of psychological assessments or to identify mechanisms for the antidepressant effects of exercise. For example, brain imaging procedures administered before and after an intervention (Kalin et al., 1997) could provide evidence for improvements in brain function after exercise training and reveal biologically plausible explanations for the effect of exercise on depression. Other physiological measures that have been used in the study of depression include assessment of heart rate and blood pressure (e.g., Carney et al., 1999), peripheral measures of neuroendocrine function (e.g., Westrin, Ekman, & Traskman-Bendz, 1999), and examination of circadian rhythms (e.g., Steiger & Holsboer, 1997). Animal studies have also investigated neurobiological adaptations to exercise (e.g., Dishman et al., 1997; Yoo, Tackett, Crabbe, Bunnell, & Dishman, 2000; O'Neal, Van Hoomissen, Holmes, & Dishman, 2000) and provide additional evidence for biological plausibility warranting further investigation.

(8) Researchers must promote collaboration among scientists, mental health professionals, physicians, and public health officials to rigorously evaluate exercise interventions for the treatment of depression. Randomized controlled trials are needed to establish exercise as a viable alternative and/or adjunctive antidepressant therapy, and the available evidence provides justification for extensive study of the antidepressant effect of exercise in clinical populations.

References

* Ades, P.A., Maloney, A., Savage, P., Carhart, R.L. (1999). Determinants of physical functioning in coronary patients. Archives of Internal Medicine, 159, 2357-2360.

American College of Sports Medicine. (1998). The recommended quantity and quality of exercise for developing and maintaining cardiorespiratory and muscular fitness, and flexibility in healthy adults. Medicine and Science in Sports and Exercise, 30(6), 975-991.

Anonymous. (2000). Practice guideline for the treatment of patients with major depressive disorder (revision). The American Journal of Psychiatry, 157(4), 1-45.

* Auchus, M.P. & Kaslow, N.J. (1994). Weight lifting therapy: a preliminary report. Psychosocial Rehabilitation Journal, 18(2), 99-101.

Bailar, J.C. (1997). The promise and problems of meta-analysis. The New England Journal of Medicine, 337(8), 559-561.

* Beniamini, Y., Rubenstein, J.J., Zaichkowsky, L.D., & Crim, M.C. (1997). Effects of high-intensity strength training on quality-of-life parameters in cardiac rehabilitation patients. American Journal of Cardiology, 80, 841-846.

* Bennett, J., Carmack, M.A., & Gardner, V.J. (1982). The effect of a program of physical exercise on depression in older adults. The Physical Educator, 39, 21-24.

* Berger, B.G. & Owen, D.R. (1983). Mood alteration with swimming – Swimmers really do "feel better." Psychosomatic Medicine, 45(5), 425-433.

Blair, S.N., Wells, C.L., Weathers, R.D., & Paffenbarger, R.S. (1994). Chronic disease: the physical activity dose-response controversy. In R.K. Dishman (Ed.), Advances in Exercise Adherence (pp. 31-54). Champaign, IL: Human Kinetics.

* Blumenthal, J.A., Emery, C.F., Madden, D.J., Schniebolk, S., Walsh-Riddle, M., George, L.K., McKee, D.C., Higginbotham, M.B., Cobb, F.R., & Coleman, R.E. (1991). Long-term effects of exercise on psychological functioning in older men and women. Journal of Gerontology, 46(6), P352-361.

*   Blumenthal, J.A., Babyak, M.A., Moore, K.A., Craighead, W.E., Herman, S., Khatri, P., Waugh, R., Napolitano, M.A., Forman, L.M., Appelbaum, M., Doraiswamy, P.M., & Krishnan, K.R. (1999). Effects of exercise training on older patients with major depression. Archives of Internal Medicine, 159, 2349-2356.

*   Bosscher, R.J. (1993). Running and mixed physical exercises with depressed psychiatric patients. International Journal of Sport Psychology, 24, 170-184.

*   Brandon, J.E. & Loftin, J.M. (1991). Relationship of fitness to depression, state and trait anxiety, internal health locus of control, and self-control. Perceptual and Motor Skills, 73, 563-568.

**   Brill, P.A., Kohl, H.W., & Blair, S.N. (1992). Anxiety, depression, physical fitness, and all-cause mortality in men. Journal of Psychosomatic Research, 36(3), 267-273.

**   Brown, D.R., Croft, J.B., Anda, R.F., Barrett, D.H., & Escobedo, L.G. (1996). Evaluation of smoking on the physical activity and depressive symptoms relationship. Medicine and Science in Sports and Exercise, 28(2), 233-240.

*   Brown, D.R., Wang, Y., Ward, A., Ebbeling, C.B., Fortlage, L., Puleo, E., Benson, H., & Rippe, J.M. (1995). Chronic psychological effects of exercise and exercise plus cognitive strategies. Medicine and Science in Sports and Exercise, 27(5), 765-775.

*   Brown, J.D. & Lawton, M. (1986). Stress and well-being in adolescence: the moderating role of physical exercise. Journal of Human Stress, Fall, 125-131.

*   Brown, R.S., Ramirez, D.E., & Taub, J.M. (1978). The prescription of exercise for depression. Physician and Sportsmedicine, December,6, 35-45,

*   Brown, S.W., Welsh, M.C., Labbe, E.E., Vitulli, W.F., & Kulkarni, P. (1992). Aerobic exercise in the psychological treatment of adolescents. Perceptual and Motor Skills, 74, 555-560.

*   Buchman, B.P. Sallis, J.F., Criqui, M.H., Dimsdale, J.E., & Kaplan, R.M. (1991). Journal of Psychosomatic Research, 35(2/3), 197-208.

Burton, R. (1632). The Anatomy of Melancholy. Oxford: Printed by Ion Lichfield for Henry Cripps.

Byrne, A. & Byrne, D.G. (1993). The effect of exercise on depression, anxiety and other mood states: a review. Journal of Psychosomatic Research, 37, 565-574.

** Camacho, T.C., Roberts, R.E., Lazarus, N.B., Kaplan, G.A., & Cohen, R.D. (1991). Physical activity and depression: Evidence from the Alameda County Study. American Journal of Epidemiology, 134, 220-231.

Carney, R.M., Freedland, K.E., Veith, R.C., Cryer, P.E., Skala, J.A., Lynch, T., & Jaffe, A.S. (1999). Major depression, heart rate, and plasma norepinephrine in patients with coronary heart disease. Biological Psychiatry, 45, 458-463.

* Carney, R.M., Harter, H.R., Templeton, B.A., Schechtman, K.B., & Goldberg, A.P. (1983). Exercise improves depression and psychosocial functioning in hemodialysis patients. Clinical Research, 31(2), 294A.

* Carney, R.M., McKevitt, P.M., Goldberg, A.P., Hagberg, J., Delmez, J.A., Harter, H.R. (1983). Psychological effects of exercise training in hemodialysis patients. Nephron, 33, 179-181.

* Carney, R.M., Wetzel, R.D., Hagberg, J., Goldberg, A.P. (1986). The relationship between depression and aerobic capacity in hemodialysis patients. Psychosomatic Medicine, 48(1/2), 143-147.

* Carney, R.M., Templeton, B., Hong, B.A., Harter, H.R., Hagber, J.M., Schechtman, K.B., Goldberg, A.P. (1987). Exercise training reduces depression and increases the performance of pleasant activities in hemodialysis patients. Nephron, 47, 194-198.

Cohen, J. (1988). Statistical power analysis for the behavioral sciences (2nd ed.). Hillsdale, NJ: Lawrence Erlbaum Associates.

** Cohen, S., Phil, D., Schwartz, J.E., Bromet, E.J., & Parkinson, D.K. (1991). Mental health, stress, and poor health behaviors in two community samples. Preventive Medicine, 20, 306-315.

**   Cooper-Patrick, L., Ford, D.E., Mead, L.A., Chang, P.P., & Klag, M.J. (1997). Exercise and depression in midlife: a prospective study. American Journal of Public Health, 87, 670-673.

*   Coyle, C.P. & Santiago, M.C. (1995). Aerobic exercise training and depressive symptomatology in adults with physical disabilities. Archives of Physical Medicine and Rehabilitation, 76, 647-652.

Craft, L.L, & Landers, D.M. (1998). The effect of exercise on clinical depression and depression resulting from mental illness: a meta-analysis. Journal of Sport & Exercise Psychology, 20, 339-357.

*   Cramer, S.R., Nieman, D.C., & Lee, J.W. (1991). The effects of moderate exercise training on psychological well-being and mood state in women. Journal of Psychosomatic Research, 35(4/5), 437-449.

*   DiLorenzo, T.M., Bargman, E.P., Stucky-Ropp, R., Brassington, G.S., Frensch, P.A., & LaFontaine, T. (1999). Long-term effects of aerobic exercise on psychological outcomes. Preventive Medicine, 28, 75-85.

*   Dimeo, F.C., Stieglitz, R., Novelli-Fischer, U., Fetscher, S., & Keul, J. (1999). Effects of physical activity on the fatigue and psychologic status of cancer patients during chemotherapy. Cancer, 85, 2273-2277.

Dishman, R.K. (1998). Physical activity and mental health. In H. Friedman (Ed.), Encyclopedia of Mental Health, Vol. 3 (pp. 171-188).

Dishman, R.K., Farquhar, R.P., & Cureton, K.J. (1994). Responses to preferred intensities of exertion in men differing in activity levels. Medicine and Science in Sports and Exercise, 26, 783-790.

Dishman, R.K., Renner, K.J., Youngstedt, S.D., Reigle, T.G., Bunnell, B.N., Burke, K.A., Yoo, H.S., Mougey, E.H., & Meyerhoff, J.L. (1997). Activity wheel running reduces escape latency and alters brain monoamine levels after footshock. Brain Research Bulletin, 42(5), 399-406.

* Doyne, E.J., Ossip-Klein, K.J., Bowman, E.D., Osborn, K.M., McDougall-Wilson, I.B., & Neimeyer, R.A. (1987). Running versus weight lifting in the treatment of depression. Journal of Consulting and Clinical Psychology, 55(5), 748-754.

* Dua, J. & Hargreaves, L. (1992). Effect of aerobic exercise on negative affect, positive affect, stress, and depression. Perceptual and Motor Skills, 75, 355-361.

* Dugmore, L.D., Tipson, R.J., Phillips, M.H., Flint, E.J., Stentiford, N.H., Bone, M.F., & Littler, W.A. (1999). Changes in cardiorespiratory fitness, psychological wellbeing, quality of life, and vocational status following a 12 month cardiac exercise rehabilitation programme. Heart, 81, 359-366.

* Dyer, J.B. & Crouch, J.G. (1987). Effects of running on moods: a time series study. Perceptual and Motor Skills, 64, 783-789.

* Emery, C.F. & Gatz, M. (1990). Psychological and cognitive effects of an exercise program for community-residing older adults. Gerontologist, 30(2),184-188.

* Emery, C.F., Schein, R.L., Hauck, E.R., & MacIntyre, N.R. (1998). Psychological and cognitive outcomes of a randomized trial of exercise among patients with chronic obstructive pulmonary disease. Health Psychology, 17(3), 232-240.

* Engles, H.J., Drouin, J., Zhu, V., Kazmierski, J.F. (1998). Effects of low-impact, moderate-intensity exercise training with and without wrist weights on functional capacities and mood states in older adults. Gerontology, 44, 239-244.

** Farmer, M.E., Locke, B.Z., Moscicki, E.K., Dannenberg, A.L., Larson, D.B., & Radloff, L.S. (1988). Physical activity and depressive symptoms: the NHANES I epidemiologic follow-up study. American Journal of Epidemiology, 128, 1340-1351.

* Fetsch, R.J. & Sprinkle, R.L. (1983). Effects of running on depressed adults. American Mental Health Counselors Association Journal, 5, 75-84.

* Folkins, C.H. (1976). Effects of physical training on mood. Journal of Clinical Psychology, 32(2), 385-388.

**   Foreyt, J.P., Brunner, R.L., Goodrick, G.K., St. Jeor, S.T., & Miller, G.D. (1995). Psychological correlates of reported physical activity in normal-weight and obese adults: the Reno diet-heart study. International Journal of Obesity and Related Disorders, 19, S69-S72.

*   Francis, K.T. & Carter, R. (1982). Psychological characteristics of joggers. Journal of Sports Medicine, 22, 386-391.

*   Frazier, S.E. & Nagy, S. (1989). Mood state changes of women as a function of regular aerobic exercise. Perceptual and Motor Skills, 68(1), 283-287.

*   Fremont, J. & Craighead, L.W. (1987). Aerobic exercise and cognitive therapy in the treatment of dysphoric moods. Cognitive Therapy and Research, 11(2), 241-251.

**   Garrison, C.Z., McKeown, R.E., Valois, R.F., & Vincent, M.L. (1993). Aggression, substance use, and suicidal behaviors in high school students. American Journal of Public Health, 83(2), 179-184.

        Gloaguen, V., Cottraux, J., Cucherat, M., & Blackburn, I. (1998). A meta-analysis of the effects of cognitive therapy in depressed patients. Journal of Affective Disorders, 49, 59-72.

*   Gondola, J.C. & Tuckman, B.W. (1982). Psychological mood state in "average" marathon runners. Perceptual and Motor Skills, 55, 1295-1300/

*   Gondola, J.C. & Tuckman, B.W. (1983). Extent of training and mood enhancement in women runners. Perceptual and Motor Skills, 57, 333-334.

        Greenberg, P.E., Stiglin, L.E., Finkelstein, S.N., & Berndt, E.R. (1993). The economic burden of depression in 1990. Journal of Clinical Psychiatry, 54(11), 405-418.

        Greenberg, R.P, Bornstein, R.F., Zborowski, Fisher, & Greenberg, M.D. (1994). A meta-analysis of fluoxetine outcome in the treatment of depression. Journal of Nervous and Mental Disorders, 182(10), 547-551.

        Greist, J.H., Klein, M.H., Eischens, R.R., Faris, J., Gurman, A.S., & Morgan, W.P. (1978). Running through your mind. Journal of Psychosomatic Research, 22, 259-294.

**   Hainer, B.L. & Palesch, Y. (1998). Symptoms of depression in residents: a South Carolina Family Practice Research Consortium study. Academic Medicine, 73(12), 1305-1310.

*   Hannaford, C.P., Harrell, E.H., & Cox, K. (1988). Psychophysiological effects of a running program on depression and anxiety in a psychiatric population. Psychological Record, 38, 37-48.

*   Hassmen, P. & Koivula, N. (1997). Mood, physical working capacity and cognitive performance in the elderly as related to physical activity. Aging: Clinical and Experimental Research, 9, 136-142.

**   Hassmen, P., Koivula, N., & Uutela, A. (2000). Physical exercise and psychological well-being: a population study in Finland. Preventive Medicine, 30, 17-25.

*   Hayden, R.M. & Allen, G.J. (1984). Relationship between aerobic exercise, anxiety, and depression: Convergent validation by knowledgeable informants. Journal of Sports Medicine, 24, 69-74.

Hedges, L.V. & Olkin, I. (1985). Statistical methods for meta-analysis. San Diego: Academic Press.

Hirschfeld, R.M.A., Keller, M.B, Panico, S., Arons, B.S., Barlow, D., Davidoff, F., Endicott, J., Froom, J., Goldstein, M., Gorman, J.M., Guthrie, D., Marek, R.G., Maurer, T.A., Meyer, R., Phillips, K., Ross, J., Schwenk, T.L., Sharfstein, S.S., Thase, M.E., & Wyatt, R.J. (1997). The National Depressive and Manic-Depressive Association consensus statement on the undertreatment of depression. Journal of the American Medical Association, 277, 333-340.

*   Hughes, J.R., Casal, D.C., & Leon, A.S. (1986). Psychological effects of exercise: a randomized cross-over trial. Journal of Psychosomatic Research, 30(3), 355-360.

Hunter, J.E., Schmidt, F.L., & Jackson, G.B. (1982). Meta-analysis: cumulating research findings across studies. Beverly Hills, CA: Sage Publications.

Ioannidis, J.P.A. & Cappelleri, J.C. (1998). Meta-analyses and large randomized, controlled trials [Letter to the editor]. The New England Journal of Medicine, 338(1), 59.

*   Jasnoski, M.L. & Holmes, D.S. (1981). Influence of initial aerobic fitness, aerobic training and changes in aerobic fitness on personality functioning. Journal of Psychosomatic Research, 25(6), 553-556.

*   Jette, A.M., Harris, B.A., Sleeper, L., Lachman, M.E., Heislein, D., Giorgetti, M., & Levenson, C. (1996). A home-based exercise program for nondisabled older adults. Journal of the American Geriatric Society, 44, 644-649.

Johnson, B.T. (1993). DSTAT 1.10: Software for the meta-analytic review of research literatures. Hillsdale, NJ: Lawrence Erlbaum Associates, Inc.

Juni, P., Witschi, A., Bloch, R., & Egger, M. (1999). The hazards of scoring the quality of clinical trials for meta-analysis. Journal of the American Medical Association, 282, 1054-1060.

Kalin, N.H., Davidson, R.J., Irwin, W., Warner, G., Orendi, J.L., Sutton, S.K., Mock, B.J., Sorenson, J.A., Lowe, M., & Turski, P.A. (1997). Functional magnetic resonance imaging studies of emotional processing in normal and depressed patients: Effects of venlafaxine. Journal of Clinical Psychiatry, 58(Suppl 16), 32-39.

*   Kavanagh, T., Shephard, R.J., Tuck, J.A., & Qureshi, S. (1977). Depression following myocardial infarction: the effects of distance running. Annals of the New York Academy of Sciences, 301, 1029-1038.

*   Keats, M.R., Courneya, K.S., Danielsen, S., & Whitsett, S.F. (1999). Leisure-time physical activity and psychosocial well-being in adolescents after cancer diagnosis. Journal of Pediatric Oncology Nursing, 16(4), 180-188.

*   King, A.C., Taylor, C.B., Haskell, W.L., & DeBusk, R.F. (1989). Influence of regular aerobic exercise on psychological health: a randomized, controlled trial of healthy middle-aged adults. Health Psychology, 8(3), 305-324.

*   King, A.C., Taylor, C.B., & Haskell, W.L. (1993). Effects of differing intensities and formats of 12 months of exercise training on psychological outcomes in older adults. Health Psychology, 12(4), 292-300.

*   King, A.C. & Brassington, G. (1997). Enhancing physical and psychological functioning in older family caregivers: the role of regular physical activity. Annals of Behavioral Medicine, 19(2), 91-100.

*   Klein, M.H., Greist, J.H., Gurman, A.S., Neimeyer, R.A., Lesser, D.P., Bushnell, N.J., Smith, R.E. (1985). A comparative outcome study of group psychotherapy vs. exercise treatments for depression. International Journal of Mental Health, 13(3-4), 148-177.

*   Kouidi, E., Iacovides, A., Iordanidis, P., Vassiliou, S., Deligiannis, A., Ierodiakonou, C., & Tourkantonis, A. (1997). Exercise renal rehabilitation program: psychosocial effects. Nephron, 77, 152-158.

**   Krause, N., Goldenhar, L., Liang, J., Jay, G., & Maeda, D. (1993). Stress and exercise among the Japanese elderly. Social Science Medicine, 36, 1429-1441.

*   Kurtze, N., Gundersen, K.T., & Svebak, S. (1999). Quality of life, functional disability and lifestyle among subgroups of fibromyalgia patients: the significance of anxiety and depression. British Journal of Medical Psychology, 72, 471-484.

**   Lampinen, P., Heikkinen, R-L., & Ruoppila, I. (2000). Changes in intensity of physical exercise as predictors of depressive symptoms among older adults: an eight-year follow-up. Preventive Medicine, 30, 371-380.

*   LaPerriere, A.R., Antoni, M.H., Schneiderman, N., Ironson, G., Klimas, N., Caralis, P., & Fletcher, M.A. (1990). Exercise intervention attenuates emotional distress and natural killer cell decrements following notification of positive serologic status for HIV-1. Biofeedback and Self-Regulation, 13(3), 229-242.

*   Lavie, C.J., Milani, R.V., Cassidy, M.M., & Gilliland, Y.E. (1999). Effects of cardiac rehabilitation and exercise training programs in women with depression. American Journal of Cardiology, 83, 1480-1483.

LeLorier, J., Gregoire, G., Benhaddad, A., Lapierre, J., & Derderian, F. (1997). Discrepancies between meta-analyses and subsequent large randomized, controlled trials. The New England Journal of Medicine, 337(8), 536-542.

*   Lobstein, D.D., Mosbacher, B.J., & Ismail, A.H. (1983). Depression as a powerful discriminator between physically active and sedentary middle-aged men. Journal of Psychosomatic Research, 27(1), 69-76.

*   MacMahon, J.R & Gross, R.T. (1988). Physical and psychological effects of aerobic exercise in delinquent adolescent males. American Journal of Diseases of Children, 142, 1361-1366.

Martinsen, E.W. (1990). Benefits of exercise for the treatment of depression. Sports Medicine, 9, 380-389. Martinsen, E.W. (1994). Physical activity and depression: Clinical experience. Acta Psychiatrica Scandinavica, 377, 23-27.

Martinsen, E.W., Hoffart, A. & Solberg, O. (1989). Comparing aerobic with nonaerobic forms of exercise in the treatment of clinical depression: A randomized controlled trial. Comprehensive Psychiatry, 30, 324-331.

Martinsen, E.W. & Medhus, A. (1989). Adherence to exercise and patients' evaluation of physical exercise in a comprehensive treatment programme for depression. Nordisk Psykiatrisk Tidsskrift, 43, 411-415.

Martinsen, E.W., Medhus, A., & Sandvik, L. (1985). Effects of aerobic exercise on depression: a controlled study. British Medical Journal, 291, 109.

Mausner, J.S. & Kramer, S. (1985). Epidemiology: An introductory text. Philadelphia: Saunders.

*   McCann, I.L. & Holmes, D.S. (1984). Influence of aerobic exercise on depression. Journal of Personality and Social Psychology, 46(5), 1142-1147.

*   McNeil, J.K., LeBlanc, E.M., & Joyner, M. (1991). The effect of exercise on depressive symptoms in the moderately depressed elderly. Psychology and Aging, 6(3), 487-488.

*   Melamed, S., Kushnir, T., Strauss, E., & Vigiser, D. (1997). Negative association between reported life events and cardiovascular disease risk factors in employed men: the CORDIS study. Journal of Psychosomatic Research, 43(4), 247-258.

*   Milani, R.V, Lavie, C.J., & Cassidy, M.M. (1996). Effects of cardiac rehabilitation and exercise training programs on depression in patients after major coronary events. American Heart Journal, 132, 726-32.

**   Milligan, R.A.K., Burke, V., Beilin, L.J., Richards, J., Dunbar, D., Spencer, M., Balde, E., & Gracey, M.P. (1997). Health-related behaviours and psycho-social characteristics of 18 year-old Australians. Soc Sci Med, 45(10), 1549-1562.

**   Mobily, K.E., Rubenstein, L.M., Lemke, J.H., O'Hara, M.W., & Wallace, R.B. (1996). Walking and depression in a cohort of older adults: the Iowa 65+ Rural Health Study. Journal of Aging and Physical Activity, 4, 119-135.

*   Morgan, W.P., Roberts, J.A., Brand, F.R., & Feinerman, A.D. (1970). Psychological effect of chronic physical activity. Medicine and Science in Sports, 2(4), 213-217.

          Morgan, W.P. (1994). Physical activity, fitness, and depression. In C. Bouchard, R.J. Shephard, & T. Stephens (Eds.), Physical activity, fitness and health (pp. 851-867). Champaign, IL: Human Kinetics Publishers.

**   Murray, N.G., Keider, S.H., & Meyers, M.C. (1998). Type of physical activity and level of depression in suburban 9th and 10th grade students (abstract). Medicine and Science in Sports and Exercise, 30(Suppl. 5), S117.

          Murray, C.J.L. & Lopez, A.D. (1996). Evidence-based health policy – lessons from the Global Burden of Disease Study. Science, 274, 740-743.

*   Netz, Y., Yaretzki, A., Salganik, I., Jacob, T., Finkeltov, B., & Argov, E. (1994). The effect of supervised physical activity on cognitive and affective state of geriatric and psychogeriatric in-patients. Clinical Gerontologist, 15(1), 47-56.

Nicoloff, G. & Schwenk, T.L. (1995). Using exercise to ward off depression. Physician and Sports Medicine, 23(9), 44-58.

*   Norris, R., Carroll, D., & Cochrane, R. (1992). The effects of physical activity and exercise training on psychological stress and well-being in an adolescent population. Journal of Psychosomatic Research, 36(1), 55-65.

North, T.C., McCullagh, P., & Tran, Z.V. (1990). Effect of exercise on depression. Exercise and Sport Sciences Reviews, 18, 379-415.

*   Norvell, N. & Belles, D. (1993). Psychological and physical benefits of circuit weight training in law enforcement personnel. Journal of Consulting and Clinical Psychology, 61(3), 520-527.

*   Norvell, N., Martin, D., & Salamon, A. (1991). Psychological and physiological benefits of passive and aerobic exercise in sedentary middle-aged women. Journal of Nervous and Mental Disease, 179(9), 573-574.

O'Connor, P.J., Aenchbacher, L.E., & Dishman, R.K. (1993). Physical activity and depression in the elderly. Journal of Aging and Physical Activity, 1, 34-58.

O'Neal, H.A., Dunn, A.L., & Martinsen, E.W. (2000). Depression and exercise. International Journal of Sport Psychology, 31, 110-135.

O'Neal, H.A., Van Hoomissen, J.D., Holmes, P.V., & Dishman, R.K. (2000). Prepro-galanin mRNA levels are increased in rat locus coeruleus after treadmill exercise training. Manuscript submitted for publication.

**   Paffenbarger, R.S., Lee, I.M., & Leung, R. (1994). Physical activity and personal characteristics associated with depression and suicide in American college men. Acta Psychiatrica Scandinavica, 377(Suppl.), 16-22.

**   Palinkas, L.A., Wingard, D.L., & Barrett-Connor, E. (1990). Chronic illness and depressive symptoms in the elderly: a population-based study. Journal of Clinical Epidemiology, 43(11), 1131-1141.

**   Palinkas, L.A., Wingard, D.L., & Barrett-Connor, E. (1996). Depressive symptoms in overweight and obese older adults: a test of the "jolly fat" hypothesis. Journal of Psychosomatic Research, 40(1), 59-66.

*   Palmer, J.A., Palmer, L.K., Michiels, K., & Thigpen, B. (1995). Effects of type of exercise on depression in recovering substance abusers. Perceptual and Motor Skills, 80, 523-530.

         Paluska, S.A. & Schwenk, T.L. (2000). Physical activity and mental health. Sports Medicine, 29(3), 167-180.

*   Pappas, G.P., Golin, S., & Meyer, D.L. (1990). Reducing symptoms of depression with exercise. Psychosomatics, 31, 112-113.

*   Partonen, T., Leppamaki, S., Hurme, J., & Lonnqvist, J. (1998). Randomized trial of physical exercise alone or combined with bright light on mood and health-related quality of life. Psychological Medicine, 28, 1359-1364.

**   Penninx, B.W.J.H., Leveille, S., Ferrucci, L., van Eijk, J.T.M., & Guralnik, J.M. (1999). Exploring the effect of depression on physical disability: Longitudinal evidence from the established populations for epidemiologic studies of the elderly. American Journal of Public Health, 89(9), 1346-1352.

*   Penny, G.D. & Rust, J.O. (1980). Effect of a walking-jogging program on personality characteristics of middle-aged females. Journal of Sports Medicine, 20, 221-226.

*   Perri, S. & Templer, D.I. (1984-85). The effects of an aerobic exercise program on psychological variables in older adults. International Journal of Aging and Human Development, 20(3), 167-172.

*   Petajan, J.H., Gappmaier, E., White, A.T., Spencer, M.K., Mino, L., & Hicks, R.W. (1996). Impact of aerobic training on fitness and quality of life in multiple sclerosis. Annals of Neurology, 39(4), 432-441.

*   Peters, M.L., Turner, S.M., & Blanchard, E.B. (1996). The effects of aerobic exercise on chronic tension-type headache. Headache Quarterly, 7(4), 330-334.

*   Pierce, T.W., Madden, D.J., Siegel, W.C., & Blumenthal, J.A. (1993). Effects of aerobic exercise on cognitive and psychosocial functioning in patients with mild hypertension. Health Psychology, 12(4), 286-291.

*   Prosser, G., Carson, P., Phillips, R., Gelson, A., Buch, N., Tucker, H., Neophytou, M., Lloyd, M. & Simpson, T. (1981). Morale in coronary patients following an exercise programme. Journal of Psychosomatic Research, 25(6), 587-593.

**   Rajala, U., Uusimaki, A., Keinanen-Kiukaanniemi, S., & Kivela, S.L. (1994). Prevalence of depression in a 55-year-old Finnish population. Society of Psychiatry and Psychiatric Epidemiology, 29, 126-130.

*   Rape, R.N. (1987). Running and depression. Perceptual and Motor Skills, 64, 1303-1310.

**   Ruuskanen, J.M. & Parkatti, T. (1994). Physical activity and related factors among nursing home residents. Journal of the American Geriatric Society, 42(9), 987-991.

        Sallis, J.F., Prochaska, J.J., & Taylor, W.C. (2000). A review of correlates of physical activity of children and adolescents. Medicine and Science in Sports and Exercise, 32(5), 963-975.

        Sargeant, J.K., Bruce, M.L., Florio, L.P., & Weissman, M.M. (1990). Factors associated with 1-year outcome of major depression in the community. Archives of General Psychiatry, 47, 519-526.

        Schwarzer, R. (1991). Meta: programs for secondary data analysis, 5.3. Berlin: Free University of Berlin.

*   Segar, M.L., Katch, V.L., Roth, R.S., Garcia, A.W., Portner, T.I., Glickman, S.G., Haslanger, S., & Wilkins, E.G. (1998). The effect of aerobic exercise on self-esteem and depressive and anxiety symptoms among breast cancer survivors. ??? 25(1), 107-113.

*   Sexton, H., Maere, A., & Dahl, N.H. (1989). Exercise intensity and reduction in neurotic symptoms. Acta Psychiatrica Scandinavica, 80, 231-235.

*   Simons, C.W. & Birkimer, J.C. (1988). An exploration of factors predicting the effects of aerobic conditioning on mood state. Journal of Psychosomatic Research, 32(1), 63-75.

*   Singh, N.A., Clements, K.M., & Fiatarone, M.A. (1997). A randomized controlled trial of progressive resistance training in depressed elders. Journal of Gerontology, 52A(1), M27-M35.

*   Sorensen, M., Anderssen, S., Hjerman, I., Holme, I, & Ursin, H. (1999). The effect of exercise and diet on mental health and quality of life in middle-aged individuals with elevated risk factors for cardiovascular disease. Journal of Sports Sciences, 17, 369-377.

*   Sothmann, M.S. & Ismail, A.H. (1984). Relationships between urinary catecholamine metabolites, particularly MHPG, and selected personality and physical fitness characteristics in normal subjects. Psychosomatic Medicine, 46(6), 523-533.

         Steiger, A. & Holsboer, F. (1997). Nocturnal secretion of prolactin and cortisol and the sleep EEG in patients with major endogenous depression during an acute episode and after full remission. Psychiatry Research, 72(2), 81-88.

**   Stephens, T. (1988). Physical activity and mental health in the United States and Canada: Evidence from four population surveys. Preventive Medicine, 17, 35-47.

*   Steptoe, A., Edwards, S., Moses, J., & Mathews, A. (1989). The effects of exercise training on mood and perceived coping ability in anxious adults from the general population. Journal of Psychosomatic Medicine, 33(5), 537-547.

**   Steptoe, A., Wardle, J., Fuller, R., Holte, A., Justo, J., Sanderman, R., & Wichstrom, L. (1997). Leisure-time physical exercise: Prevalence, attitudinal correlates, and behavioral correlates among young Europeans from 21 countries. Preventive Medicine, 26, 845-854.

*   Stern, M.J. & Cleary, P. (1981). National Exercise and Heart Disease Project: Psychosocial changes observed during a low-level exercise program. Archives of Internal Medicine, 141, 1463-1467.

*   Stetson, B., Schlundt, D.G., Sbrocco, T., Hill, J.O., Sharp, T., & Pope-Cordel, J. (1992). The effects of aerobic exercise on psychological adjustment: a randomized study of sedentary obese women attempting weight loss. Women & Health, 19(4), 1-14.

**   Stewart, A.L., Hays, R.D., Wells, K.B., Rogers, W.H., Spritzer, K.L., & Greenfield, S. (1994). Long-term functioning and well-being outcomes associated with physical activity and exercise in patients with chronic conditions in the Medical Outcomes Study. Journal of Clinical Epidemiology, 47(7), 719-730.

*   Stewart, A.L., Mills, K.M., Sepsis, P.G., King, A.C., McLellan, B.Y., Roitz, K., & Ritter, P.L. (1997). Evaluation of CHAMPS, a physical activity promotion program for older adults. Annals of Behavioral Medicine, 19(4), 353-361.

**   Strawbridge, W.J., Cohen, R.D., Shema, S.J., & Kaplan, G.A. (1996). Successful aging: Predictors and associated activities. American Journal of Epidemiology, 144, 135-141.

*   Taylor, C.B., Houston-Miller, N., Ahn, D.K., Haskell, W., & DeBusk, R.F. (1986). The effects of exercise training programs on psychosocial improvement in uncomplicated postmyocardial infarction patients. Journal of Psychosomatic Research, 30(5). 581-587.

*   Terry, L., Sprinz, E. & Ribeiro, J.P. (1999). Moderate and high intensity exercise training in HIV-1 seropositive individuals: a randomized trial. International Journal of Sports Medicine, 20, 142-146.

Tkachuk, G.A. & Martin, G.L. (1999). Exercise therapy for patients with psychiatric disorders: Research and clinical implications. Professional Psychology: Research and Practice, 30(3), 275-282.

* Valliant, P.M. & Asu, M.E. (1985). Exercise and its effects on cognition and physiology in older adults. Perceptual and Motor Skills, 61, 1031-1038.

* Veale, D., Le Fevre, K., Pantelis, C., de Souza, V., Mann, A., & Sargeant, A. (1992). Aerobic exercise in the adjunctive treatment of depression: a randomized controlled trial. Journal of the Royal Society of Medicine, 85, 541-544.

* Wearden, A.J., Morriss, R.K., Mullis, R., Strickland, P.L., Pearson, D.J., Appleby, L., Campbell, I.T., & Morris, J.A. (1998). Randomised, double-blind, placebo-controlled treatment trial of fluoxetine and graded exercise for chronic fatigue syndrome. British Journal of Psychiatry, 172, 485-492.

Weissman, M.M., Bland, R.C., Canino, G.J., Faravelli, C., Greenwald, S., Hwu, H-G., Joyce,P.R., Karam, E.G., Lee, C-K., Lellouch, J., Lepine, J-P., Newman, S.C., Rubio-Stipec, M., Wells, J.E., Wickramaratne, P.J., Wittchen, H-U., & Yeh, E-K. (1996). Cross-national epidemiology of major depression and bipolar disorder. Journal of the American Medical Association, 276(4), 293-299.

Westrin, A., Ekman, R., & Traskman-Bendz, L. (1999). Alterations of corticotropin releasing hormone (CRH) and neuropeptide Y (NPY) plasma levels in mood disorder patients with a recent suicide attempt. European Neuropsychopharmacology, 9, 205-211.

** Weyerer, S. (1992). Physical inactivity and depression in the community: Evidence from the Upper Bavarian Field Study. International Journal of Sports Medicine, 13(6), 492-496.

* Wigers, S.H., Stiles, T.C., & Vogel, P.A. (1996). Effects of aerobic exercise versus stress management treatment in fibromyalgia. Scandinavian Journal of Rheumatology, 25, 77-86.

*   Wilfley, D. & Kunce, J. (1986). Differential physical and psychological effects of exercise. Journal of Counseling Psychology, 33(3), 337-342.

*   Williams, J.M. & Getty, D. (1986). Effect of levels of exercise on psychological mood states, physical fitness, and plasma beta-endorphin. Perceptual and Motor Skills, 63, 1099-1105.

*   Williams, P. & Lord, S.R. (1997). Effects of group exercise on cognitive functioning and mood in older women, Australian and New Zealand Journal of Public Health, 21(1), 45-52.

*   Wilson, V.E., Morley, N.C., & Bird, E.I. (1980). Mood profiles of marathon runners, joggers and non-exercisers. Perceptual and Motor Skills, 50, 117-118.

**   Yaffe, K., Blackwell, T., Gore, R., Sands, L., Reus, V., & Browner, W.S. (1999). Depressive symptoms and cognitive decline in nondemented elderly women. Archives of General Psychiatry, 56, 425-430.

        Yoo, H.S., Tackett, R.L., Crabbe, J.B., Bunnell, B.N., & Dishman, R.K. (2000). Antidepressant-like effects of physical activity vs. imipramine: Neonatal clomipramine model. Psychobiology, in press.

*   Studies included in the overall meta-analysis.
** Studies included in the review of epidemiological literature.

Table 1. Epidemiological studies examining the relationship between exercise and depression.

STUDY	SAMPLE	RESEARCH DESIGN	EXERCISE MEASURE	DEPRESSION MEASURE	POST-TEST	EFFECT
Camacho et al. (1991)	n=3,789 (2-wave) n=1,275 (3-wave) men and women aged 20 years and older	Cross-sectional and longitudinal study of Alameda County Study participants; 3 time points: 1965, 1974, and 1983	Self-report of frequency and intensity of leisure physical activity and change in physical activity (2 levels)	Self-report of depressive symptoms; depression indicated by cut-off score	1974 depression by activity level: low active moderate active	OR = 4.22 (3.17-5.62) OR = 2.14 (1.61-2.86)
					1965 activity / 1974 depression: (men) low active moderate active (women) low active moderate active	OR = 1.76 (1.06-2.92) OR = 1.46 (0.91-2.34) OR = 1.7 (1.06-2.70) OR = 1.0 (0.63-1.59)
					1965-74 activity change / 1974 depression: low – low low – high high – low	OR = 3.76 (2.57-5.50) OR = 2.12 (1.35-3.34) OR = 2.93 (1.92-4.46)
					1965-74 activity change / 1983 depression: low-low low-high high-low	RR = 1.22 (0.62-2.38) RR = 1.11 (0.53-2.21) RR = 1.61 (0.80-3.22)

Cohen et al. (1991)	n=404 men and 764 women recruited from communities in Pennsylvania	Cross-sectional data from a study of the psychological effects of the Three Mile Island accident	Self-report of frequency of exercise; "lack of exercise" defined as exercising less than 1-2 times per week in a typical month	Self-report of history of major depressive episodes according to RDC criteria	Lack of exercise / depressive episode men women	β = 0.02 ns β = -0.25 ns
Cooper-Patrick et al. (1997)	n=752 former medical students	Longitudinal study of John Hopkins University medical students; data collected in 1978 and 1993	Self-report of frequency of exercising to a sweat in 1978 (3 levels)	Self-report of previous diagnosis of clinical depression and review of medical records through 1993	1978 exercise / 1993 depression: 0 times/wk 1-2 times/wk	RR = 1.18 (0.53-2.64) RR = 1.08 (0.48-2.45)

Farmer et al. (1988)
 	
see also Brown et al. (1996)
	n=1900 men and women aged 25-77 years	Cross-sectional and longitudinal study of participants from the National Health and Nutrition Examination Survey (NHANES I); 1971-75, 1982-84	Self-report of recreational and nonrecreational physical activity (2 levels)	CES-D self-report questionnaire; cut-off score used to indicate depression	Low activity / depression at baseline:
					white men
					black men
					white women
					black women
					
					Low activity at baseline/ depression at follow-up by depression at baseline:
					   (white men)
					low depression
					high depression
					   (white women)
					low depression
					high depression
						OR = 2.2 (1.2-4.2)
						OR = 16.5 (2.1-128)
						OR = 1.7 (1.1-2.5)
						OR = 1.2 (0.3-4.1)
						
						RR = 1.3 (0.5-3.1)
						RR = 12.9 (1.7-98.9)
						
						RR = 1.9 (1.1-3.2)
						RR = 2.0 (0.8-14.5)

Foreyt et al. (1995)	n=381 obese and normal-weight adults	Cross-sectional and longitudinal study of Reno Diet-Heart Study participants; assessments conducted at year 1 and year 5	Self-report of frequency and beliefs regarding recreational physical activity	CES-D self-report questionnaire	Activity / depression at (Year 1) normal obese (Year 5) normal obese	r = -0.02 ns r = -0.15 r = -0.05 ns r = -0.21
					Actvity change / depression at follow-up: normal weight obese	r = -0.17 r = 0.11 ns
Hainer & Palesch (1998)	n=284 male and female medical residents	Cross-sectional analysis of data obtained from the South Carolina Family Practice Research Consortium Study	Self-report of perception of physical fitness	BDI self-report questionnaire; POMS depression-dejection scale score	Poor fitness / depression score BDI POMS	p<.01 ns
Hassmen et al. (2000)	n=3,403 Finnish men and women aged 25-64 years	Cross-sectional study of Finnish cardiovascular risk factor survey participants	Self-report of frequency of recreational exercise (6 levels)	BDI self-report questionnaire	Exercise frequency / depression	p<.001
Krause et al. (1993)	n=1,351 Japanese men and women aged 60 and older	Cross-sectional study of Japanese elderly randomly selected in a nationwide survey	Self-report of frequency of recreational physical activity	Self-report of depressive symptoms based on CES-D items	Exercise frequency / depression	β = -0.16 (p<.01)

Lampinen et al. (2000)	n=663 Finnish men and women aged 65 and older	Cross-sectional and longitudinal study of Finnish elderly participating in the Evergreen Project; assessments conducted in 1988 and 1996	Self-report of intensity of current physical activity; classified into 3 levels: necessary chores, regular walking, strenuous exercise	Self-report of depressive symptoms using modified BDI questionnaire	Change in intensity / 1996 depression: (chores) no change (walking) decreased no change (exercise) decreased	OR = 0.96 (0.25-3.7) OR = 10.56 (2.4-47.4) OR = 2.21 (0.53-9.3) OR = 1.23 (0.32-5.03)
Melamed et al. (1997)	n=1,837 male employees aged 20-64 years	Cross-sectional study of participants in the Cardiovascular Occupational Risk Factors Determination in Israel (CORDIS) study	Level of occupational physical work (4 levels)	Self-report of depressive symptoms using modified SDS	Physical work / depression	$\beta = -0.11$ (p=.0001)
Milligan et al. (1997)	n=583 Australian men and women aged 18 years	Cross-sectional analysis of data obtained from a longitudinal cardiovascular risk factor study of Australian youth	Self-report of weekly frequency, duration, type and intensity of physical activity; summed to yield total score (2 levels)	Zung SDS self-report questionnaire	(male) Activity / depression Fitness / depression (female) Activity / depression Fitness / depression	p=.0003 r = ns p = ns r = -0.14, (p=.03)

Mobily et al. (1996)	n=2,084 men and women aged 65 and older	Cross-sectional and longitudinal study of Iowa 65+ Rural Health Study participants; baseline and 3-year follow-up	Self-report of frequency of walking (2 levels)	Self-report of depressive symptoms based on CES-D items; cut-off score used to indicate depression	Depression at baseline: walkers vs nonwalkers	OR = .825 (0.64-1.06)
					Low depression at baseline / depression at follow-up: walkers vs nonwalkers	RR = 1.1 (0.77-1.60)
					High depression at baseline / depression at follow-up: walkers vs nonwalkers	RR = .379 (0.18-0.79)
Murray et al. (1998)	n=688 male and female high school students	Cross-sectional study of ninth and tenth grade students	Self-repot of frequency of vigorous exercise, frequency of strength training, number of sport teams, and perception of exercise in relation to peers	Self-report of depressive symptoms	Physical activity / depression (males) exercise strength training sports teams perception (females) exercise strength training sports teams perception	p=.013 ns ns p=.006 p<.001 p=.036 p=.001 p=.001

Paffenbarger et al. (1994)	n=10,201 men	Longitudinal study of Harvard Alumni; 1962-1988	Self-report of physical activity habits including sports play hr/week and kcal/wk (3 levels)	Self-report of diagnosis of clinical depression	Baseline activity / depression through 1988: (Sports play) 1-2 hr/wk 3+ hr/wk (Activity Index) 1000-2499kcal 2500+kcal	RR = 0.96 RR = 0.73 RR = 0.83 RR = 0.72
Palinkas et al. (1990)	n=741 men and 874 women aged 65 years and older	Cross-sectional follow-up data from the Lipid Research Clinic Prevalence Study of heart disease risk factors	Self-report of exercise frequency with a target of 3 times/wk (2 levels); self-report of change in exercise behavior during the past 10 years	BDI self-report questionnaire; cut-off score used to indicate depression	Activity / mean depression score (male) Little Exercise Decrease (female) Little Exercise Decrease	p<.001 p<.001 p<.001 p<.001
					Activity / percent reporting depression (male) Little Exercise Decrease (female) Little Exercise Decrease	p<.001 p<.001 ns p<.05

Palinkas et al. (1996)	n=2,245 men and women aged 50-89 years	Cross-sectional follow-up data from heart disease risk factor study	Self-report of exercise frequency with a target of 3 times/wk (2 levels); self-report of change in exercise behavior during the past 10 years	BDI self-report questionnaire; cut-off score used to indicate depression	Low activity / depression: males females	RR = 1.69 (0.79-3.6) RR = 1.26 (0.76-2.08)
Penninx et al. (1999)	n=6,247 men and women aged 65-103 years	Cross-sectional analysis of data from the Established Populations for Epidemiologic Studies of the Elderly	Self-report of frequency of walking, gardening, and vigorous exercise	CES-D self-report questionnaire; cut-off score used to indicated depression	Low activity / depression	p<.001
Rajala et al. (1994)	n=780 men and women aged 55 years	Cross-sectional study of depression in Finnish adults	Self-report of exercise during leisure and when commuting to/from work (3 levels)	Zung SDS self-report questionnaire	Activity / depression: (men) little moderate	OR = 2.0 (0.7-5.8) OR = 0.7 (0.2-1.9)
					(women) little moderate	OR = 1.1 (0.4-2.9) OR = 0.9 (0.5-1.5)
Stephens (1988)	n=3,011 men and women aged 20-64 years	Cross-sectional analysis of data from the 1979 National Survey of Personal Health Practices and Consequences (NSPHPC I)	Self-report of frequency leisure physical activity (5 levels)	Self-report of frequency of negative mood using the Blue-cheer index	Activity / mood: (men) < 40 years > 40 years (women) < 40 years > 40 years	p=.05 p<.001 p<.001 p<.001

Stephens (1988)	n=14,118 men and women aged 15 years and older	Cross-sectional analysis of data from the 1978-79 Canada Health Survey (CHS)	Self-report of frequency, intensity, and duration of leisure physical activity used to calculate energy expenditure during the past 2 weeks (5 levels)	Self-report of frequency of negative feelings using the Negative Affect Scale	Energy expenditure / negative affect (men) < 40 years > 40 years (women) < 40 years > 40 years	p<.001 p<.001 p=.005 p<.001
Stephens (1988)	n=15,239 males and females aged 10 years and older	Cross-sectional analysis of data from the 1981 Canada Fitness Survey (CFS)	Self-report of frequency, duration, and type of leisure physical activity during the past 12 months used to calculate energy expenditure (5 levels)	Self-report of frequency of negative feelings using the Negative Affect Scale	Energy expenditure / negative affect (men) < 40 years > 40 years (women) < 40 years > 40 years	ns ns ns p<.001
Steptoe et al. (1997)	n=5,529 male and female university students aged 18-30 years	Cross-sectional analysis of data obtained from the European Health and Behavior Survey	Self-report of frequency of exercise (2 levels)	Self-report of depressive symptoms based on items from the BDI	Exercise frequency / depression	p<.0001
Stewart et al. (1994)	n=1758 male and female chronic disease patients	Longitudinal analysis from the Medical Outcomes Study	Self-report of total time spent exercising, total time spent walking, and perceived level of physical activity	Self-report of time during the past month an individual experienced symptoms of depression	Activity at baseline / depression at follow-up: total exercise total walking perceived level	p<.01 n..s. p<.01

Weyerer (1992)	n=1,536 men and women aged 15 years and older	Cross-sectional and longitudinal study of Upper Bavarian Field Study participants; 1975-79, 1980-84	Self-report of frequency of leisure exercise (3 levels)	ICD diagnosis of depression by Clinical Interview Schedule	Activity / depression at baseline: none occasional	OR = 3.15 (1.84-5.38) OR = 1.55 (0.82-2.92)
					Activity at baseline / depression at follow-up: (men) none occasional (women) none occasional	RR = 1.15 (0.3-1.62) RR = .27 (0.03-2.35) RR = .70 (0.3-1.62) RR = .65 (0.26-1.61)
Yaffe et al. (2000)	n=5,781 women aged 65 years and older	Cross-sectional analysis of data from the Study of Osteoporotic Fractures	Self-report of physical activity during the past week (kcal)	Self-report of depressive symptoms using the GDS (3 levels)	Energy expenditure / depression	p<.001

Note. Beck Depression Inventory (BDI), Center for Epidemiological Studies-Depression Scale (CES-D), Geriatric Depression Scale (GDS), International Classification of Diseases (ICD), Profile of Mood States (POMS), Research Diagnostic Criteria (RDC), Zung Self-Rating Depression Scale (SDS)

Figure 1. Relative risk for depressive symptoms for individuals reporting low levels of physical activity.
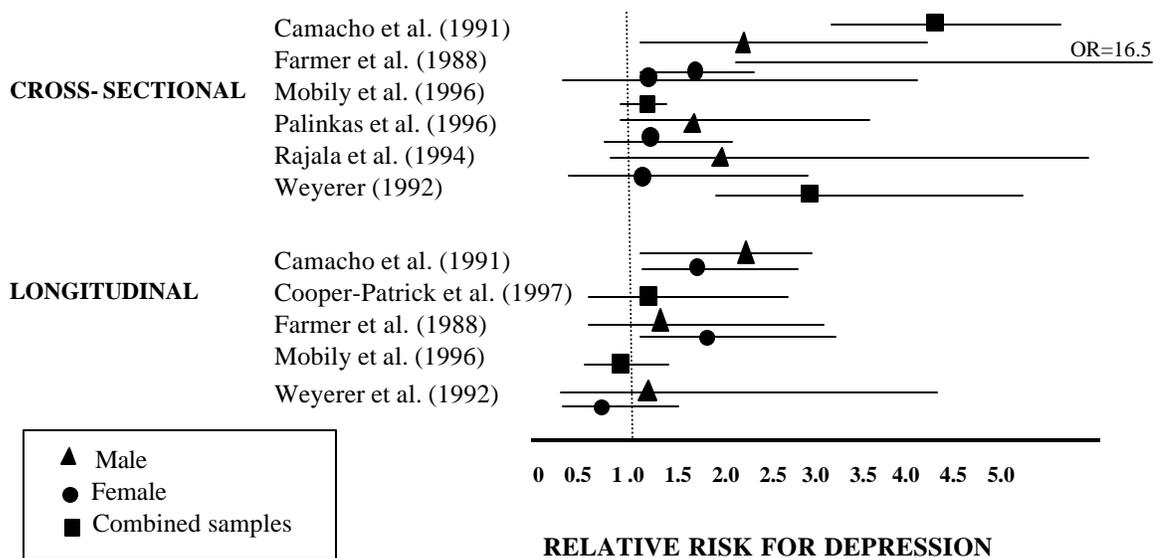
Table 2. Moderating variables influencing effects of exercise in reducing symptoms of depression.

GENDER	**Males:** Only males were studied or a separate analysis was performed for males **Females:** Only females were studied or a separate analysis was performed for females **Males & Females:** Evaluation of the intervention was not separated according to gender
AGE	**Adult:** Subjects were 18 years or older, not including college students **Elderly:** Study specified that subjects were older adults, usually over age 65 **College:** Subjects were university students **Youth:** Subjects were younger than 18 years of age
SOURCE	**Hospital/Medical Registry:** Subjects were recruited from inpatient or outpatient facilities or identified through medical registries **University**: Subjects were recruited from university populations including students, faculty and staff **Community**: Subjects were recruited from the general population
HEALTH STATUS	**Healthy:** Subjects were apparently healthy **Obese:** Subjects were classified as significantly overweight **CVD:** Subjects were identified as being at risk for or currently diagnosed with cardiovascular disease **Cancer:** Subjects had current or previous diagnosis of cancer **Disabled:** Subjects were identified as having some type of chronic disability, including fibromyalgia **Dialysis:** Subjects were undergoing current dialysis treatment
MENTAL STATUS	**Non-depressed:** Subjects were not identified as having elevated symptoms of depression or a diagnosis of clinical depression **Depressed:** Subjects were diagnosed with a depressive disorder or exhibited elevated depression scores **Psychiatric:** Subjects were diagnosed with a comorbid psychological disorder or were recruited from a general psychiatric facility
DESIGN	**Non-randomized:** Pre- or Quasi-experimental study including case study, cohort with no control group, and nonequivalent control group **Randomized:** Factorial design with a no-treatment, attention control, or traditional antidepressant intervention comparison group
COMPARISON GROUP	**None**: No control group was used or effect sizes could only be computed within the exercise group **No-treatment/Wait List Control:** Subjects in the comparison group received no intervention and participated only in pre and post-assessments **Psychotherapy:** Subjects in the control group received individual or group counseling **Attention Control/Placebo:** Subjects received a minimal intervention including health education, stress management, or relaxation training **Drug:** Subjects in the control group received an antidepressant medication

EXERCISE MEASURE	**Exercise Intervention:** An exercise program was implemented and change in depression scores was obtained pre to post-training **Self-report of physical activity:** The independent variable was level of physical activity as reported by the subjects **Fitness Correlation:** The independent variable reflected the relationship between physical fitness and depression score
EXERCISE SETTING	**Medical Facility:** Exercise sessions were conducted in a hospital or rehabilitation center **University:** Exercise sessions were part of a college course or were conducted in a laboratory setting **Home:** Exercise sessions were performed in the participant's home or surrounding area
LENGTH OF TRAINING	**Intervention Length:** Number of weeks the exercise intervention spanned
EXERCISE ADMINISTRATION	**Group:** Intervention was implemented in a group setting **Individual:** Intervention was conducted one-on-one with little or no contact with other subjects **Individual & Group:** Intervention was conducted using both individual contact and group activities
EXERCISE MODE	**Aerobic:** Prescribed physical activity was primarily aerobic, including walking, jogging, and aerobic dance. **Active leisure:** No mode of activity was specified or the goal involved increasing the level of physical activity or active games **Strength:** Program involved muscular strength and endurance training, with no aerobic component **Flexibility/Yoga:** Program involved stretching or other low intensity exercises **Combined exercise:** Program included a varied training regime, including flexibility, resistance, and/or aerobic activities
EXERCISE INTENSITY	Percent of aerobic capacity or heart rate reserve. If intensity was not specified, estimates were made based on information about the mode and sample. For example, walking was categorized as low (below 50%) and jogging as moderate (50% - 70%).
EXERCISE FREQUENCY	Number of days per week that physical activity was scheduled or prescribed.
EXERCISE DURATION	Length of each exercise session in minutes.
EXERCISE SUPERVISION	**Supervised:** Subjects met with a member of the research/ intervention team on a regular basis individually or in a group to receive the specific intervention and/or to exercise. **Not supervised:** Intervention was provided once or twice individually or in a group, after which subjects exercised on their own.
DEPRESSION MEASURE	**Clinical Rating:** The dependent variable was a clinician-administered questionnaire or interview **Self-Report Questionnaire:** The dependent variable was a self-report assessment of depressive symptoms
FOLLOW-UP	When the study included a follow-up period, effect sizes were computed for depression scores after the conclusion of the intervention

Table 3. Moderators of intervention effects. The number of effects for each level is indicated by k.

	k	N	d	95% CI
Gender				
Male	25	1107	-0.74	-1.28, -0.20
Female	20	817	-0.40	-0.73, -0.07
Male/Female	42	2185	-0.49	-0.78, -0.19
Age				
Adult	55	2453	-0.60	-0.90, -0.31
Elderly	18	834	-0.39	-0.90, 0.11
College	9	645	-0.66	-1.12, -0.20
Youth	5	177	-0.19	-0.49, 0.11
Recruitment Source				
Hospital	19	739	-0.96	-1.61, -0.31
University	19	1088	-0.29	-0.58, 0.00
Community	41	1898	-0.53	-0.88, -0.18
Health Status				
Healthy	60	2892	-0.39	-0.61, -0.17
Chronic Disease	23	914	-0.92	-1.45, -0.40
Mental Status				
Non-depressed	69	3383	-0.47	-0.73, -0.22
Depressed	15	638	-1.07	-1.68, -0.47
Research Design (Controlled Studies)				
Non-randomized	29	1419	-0.63	-1.03, -0.23
Randomized	59	2707	-0.54	-0.81, -0.26
Comparison Group				
None	107	9704	-0.67	-0.86, -0.48
No Treatment	73	3563	-0.53	-0.75, -0.31
Attention Control	13	579	-0.50	-1.20, 0.22
Exercise Measure				
Exercise Intervention	79	3847	-0.49	-0.72, -0.27
Self-report	9	279	-1.25	-2.20, -0.30
Fitness Correlation	10	1034	-0.57	-0.84, -0.30
Exercise Setting				
Medical Facility	7	209	-0.49	-0.77, -0.21
University	24	1247	-0.29	-0.58, 0.00
Home	11	553	-0.35	-0.85, 0.15
Intervention Length				
4-8 weeks	21	703	-0.38	-0.69, -0.08
9-12 weeks	29	1436	-0.52	-0.87, -0.17
13-16 weeks	20	831	-0.18	-0.36, -0.01
17-24 weeks	8	317	-2.18	-3.12, -1.24
25-52 weeks	14	1045	-0.26	-0.49, -0.04

Exercise Administration

Group	39	1991	-0.26	-0.50, -0.11
Individual	25	1190	-0.53	-0.84, -0.23
Individual/Group	6	410	-0.81	-2.26, 0.65

Exercise Mode

Aerobic	69	3037	-0.56	-0.82, -0.31
Strength	7	295	-1.15	-2.42, 0.11
Flexibility/Yoga	6	160	-0.05	-0.50, 0.40
Combined	6	369	-0.25	-0.46, -0.05

Exercise Intensity

Low (<50%)	15	449	-0.18	-0.53, 0.17
Moderate (50-70%)	45	2110	-0.42	-0.68, -0.15
High (>70%)	12	636	-0.48	-1.17, 0.22

Exercise Frequency

1-2 days/week	11	488	-0.41	-0.77, -0.02
3-4 days/week	58	2744	-0.41	-0.65, -0.16
5+ days/week	8	440	-0.20	-0.47, 0.07

Exercise Duration

20-30 minutes	18	951	-0.32	-0.56, -0.09
31-45 minutes	24	859	-0.38	-0.87, 0.10
>45 minutes	21	1339	-0.29	-0.53, -0.04

Exercise Supervision

Supervised	60	2977	-0.41	-0.65, -0.17
Unsupervised	28	1149	-0.93	-1.41, -0.45

Depression Measure

Clinical Rating	9	431	-1.75	-2.84, -0.67
Self-report	85	3985	-0.53	-0.75, -0.31

Follow-up Assessment

12 weeks	5	269	-1.18	-2.11, -0.25
13-26 weeks	9	459	-0.59	-1.02, -0.16
27-52 weeks	6	282	-1.03	-1.29, -0.78
>52 weeks	3	80	-0.92	-1.83, -0.01
overall	23	1090	-0.91	-1.21, -0.61

Figure 2. Stem-and-Leaf Plot of Effect Sizes (d) Included in the Overall Analysis.

```
-1 .  67
-1 .  00111223
-0.   55555667777889
-0 .  00001111112222223333334444444444
 0 .  0000000111222233444
 0 .  5557
```

Appendix 1. Located studies not included in the meta-analysis.

Anderson, R.E., Wadden, T.A., Bartlett, S.J., Zemel, B., Verde, T.J., & Franckowiak, S.C. (1999). Effects of lifestyle activity vs structured aerobic exercise in obese women. Journal of the American Medical Association, 281(4), 335-340.

Anshel, M.H. & Russell, K.G. (1994). Effect of aerobic and strength training on pain tolerance, pain appraisal and mood of unfit males as a function of pain location. Journal of Sport Sciences, 12, 535-547.

Blumenthal, J.A., Emery, C.F., Madden, D.J., George, L.K., Coleman, R.E., Riddle, M.W., McKee, D.C., Reasoner, J., & Williams, R.S. (1989). Cardiovascular and behavioral effects of aerobic exercise training in healthy older men and women. Journal of Gerontology, 44(5), M147-M157.

Blumenthal, J.A., Schocken, D.D., Needels, T.L., & Hindle, P. (1982). Psychological and physiological effects of physical conditioning on the elderly. Journal of Psychosomatic Research, 26(5), 505-510.

Brus, H.L.M., Taal, E., van de Laar, M.A.F.J., Rasker, J.J., & Wiegman, O. (1997). Patient education and disease activity: a study among rheumatoid arthritis patients. Arthritis Care and Research, 10(5), 320-324.

De Geus, E.J.C., Van Doornen, L.J.P., & Orlebeke, J.F. (1993). Regular exercise and aerobic fitness in relation to psychological make-up and physiological stress reactivity. Psychosomatic medicine, 55, 347-363.

Doyne, E.J., Chambless, D.L., & Beutler, L.E. (1983). Aerobic exercise as a treatment for depression in women. Behavior Therapy, 14, 434-440.

Dustman, R.E., Ruhling, R.O., Russell, E.M., Shearer, D.E., Bonekat, H.W., Shigeoka, J.W., Wood, J.S., & Bradford, D.C. (1984). Aerobic exercise training and improved neuropsychological function of older adults. Neurobiology of Aging, 5, 35-43.

Folkins, C.H., Lynch, S., & Gardner, M.M. (1972). Psychological fitness as a function of physical fitness. Archives of Physical Medicine & Rehabilitation, 53(11), 503-508.

Gondola, J.C. & Tuckman, B.W. (1982). Psychological mood states in "average" marathon runners. Perceptual and Motor Skills, 55, 1295-1300.

Gondola, J.C. & Tuckman, B.W. (1983). Extent of training and mood enhancement in women runners. Perceptual and Motor Skills, 57, 333-334.

Greist, J.H., Klein, M.H., Eischens, R.R., Faris, J., Gurman, A.S., & Morgan, W.P. (1978). Running through your mind. Journal of Psychosomatic Research, 22, 259-294.

Greist, J.H., Klein, M.H., Eischens, R.R., Faris, J., Gurman, A.S., & Morgan, W.P. (1979). Running as treatment for depression. Comprehensive Psychiatry, 20(1), 41-54.

Hannaford, C.P., Harrell, E.H., & Cox, K. (1988). Psychophysiological effects of a running program on depression and anxiety in a psychiatric population. The Psychological Record, 38, 37-48.

Hart, G.W., Wallace, W.L., & Cayton, T.G. (1982). Effect of aerobic conditioning upon mood in clinically depressed men and women: a preliminary investigation. Perceptual and Motor Skills, 55, 1217-1218.

Laukkanen, P., Era, P., Heikkinen, R-L., Suutama, T., Kauppinen, M., & Heikkinen, E. (1994). Factors related to carrying out everyday activities among elderly people aged 80. Aging: Clinical and Experimental Research, 6(6), 433-443.

Lennox, S.S., Bedell, J.R., & Stone, A.A. (1990). The effect of exercise on normal mood. Journal of Psychosomatic Research, 34(6), 629-636.

Martinsen, E.W., Hoffart, A. & Solberg, O. (1989). Comparing aerobic with nonaerobic forms of exercise in the treatment of clinical depression: A randomized controlled trial. Comprehensive Psychiatry, 30, 324-331.

Martinsen, E.W., Medhus, A., & Sandvik, L. (1985). Effects of aerobic exercise on depression: a controlled study. British Medical Journal, 291, 109.
Mausner, J.S. & Kramer, S. (1985). Epidemiology: An introductory text. Philadelphia: Saunders.

McMurdo, M.E.T. & Burnett, L. (1992). Randomised controlled trial of exercise in the elderly. Gerontology, 38, 292-298.

Moses, J., Steptoe, A., Mathews, A., & Edwards, S. (1989). The effects of exercise training on mental well-being in the normal population: a controlled trial. Journal of Psychosomatic Research, 33(1), 47-61.

O'Rourke, A., Lewin, B., Whitecross, S., & Pacey, W. (1991). The effects of physical exercise training and cardiac education on levels of anxiety and depression in the rehabilitation of coronary artery bypass graft patients. International Disability Studies, 12, 104-106.

Robb-Nicholson, L.C., Daltroy, L., Eaton, H., Gall, V., Wright, E., Hartley, L.H., Schur, P.H., & Liang, M.H. (1989). Effects of aerobic conditioning in lupus fatigue: a pilot study. British Journal of Rheumatology, 28, 500-505.

Sime, W.E. & Sanstead, M. (1987). Running therapy in the treatment of depression: Implications for prevention. In R.F. Munoz (Ed.), Depression Prevention: Research Directions (pp. 125-138). New York: Hemisphere Publishing.

Singh, N.A., Clements, K.M., & Fiatarone, M.A. (1997). A randomized controlled trial of the effect of exercise on sleep. Sleep, 20(2), 95-101.

Slater, M.A., Doctor, J.N., Pruitt, S.D., & Atkinson, J.H. (1997). The clinical significance of behavioral treatment for chronic low back pain: an evaluation of effectiveness. Pain, 71, 257-263.

Stern, K.J. & Cleary, P. (1982). The national exercise and heart disease project: Long-term psychosocial outcome. Archives of Internal Medicine, 142, 1093-1097.

Tsutsumi, T., Don, B.M., Zaichkowsky, L.D., & Delizonna, L.L. (1997). Physical fitness and psychological benefits of strength training in community dwelling older adults. Applied Human Science, 16(6), 257-266.

Wagner, G., Rabkin, J., & Rabkin, R. (1998). Exercise as a mediator of psychological and nutritional effects of testosterone therapy in HIV+ men. Medicine and Science in Sports and Exercise, 30(6), 811-817.

Appendix 2. Studies included in the analysis of follow-up effects.

DiLorenzo, T.M., Bargman, E.P., Stucky-Ropp, R., Brassington, G.S., Frensch, P.A., & LaFontaine, T. (1999). Long-term effects of aerobic exercise on psychological outcomes. Preventive Medicine, 28, 75-85.

Doyne, E.J., Ossip-Klein, K.J., Bowman, E.D., Osborn, K.M., McDougall-Wilson, I.B., & Neimeyer, R.A. (1987). Running versus weight lifting in the treatment of depression. Journal of Consulting and Clinical Psychology, 55(5), 748-754.

Fremont, J. & Craighead, L.W. (1987). Aerobic exercise and cognitive therapy in the treatment of dysphoric moods. Cognitive Therapy and Research, 11(2), 241-251.

Partonen, T., Leppamaki, S., Hurme, J., & Lonnqvist, J. (1998). Randomized trial of physical exercise alone or combined with bright light on mood and health-related quality of life. Psychological Medicine, 28, 1359-1364.

Sexton, H., Maere, A., & Dahl, N.H. (1989). Exercise intensity and reduction in neurotic symptoms. Acta Psychiatrica Scandinavica, 80, 231-235.

Steptoe, A., Edwards, S., Moses, J., & Mathews, A. (1989). The effects of exercise training on mood and perceived coping ability in anxious adults from the general population. Journal of Psychosomatic Medicine, 33(5), 537-547.

Wigers, S.H., Stiles, T.C., & Vogel, P.A. (1996). Effects of aerobic exercise versus stress management treatment in fibromyalgia. Scandinavian Journal of Rheumatology, 25, 77-86.

PREPRO-GALANIN MRNA LEVELS ARE INCREASED IN RAT LOCUS

COERULEUS AFTER TREADMILL EXERCISE TRAINING[2]

---

[2]O'Neal, H.A., Van Hoomissen, J.D., Holmes, P.V., and Dishman, R.K. (2000) To be
submitted to Neuroscience Letters.

Abstract

The effects of treadmill exercise training on prepro-galanin (GAL) and tyrosine hydroxylase (TH) gene expression in the locus coeruleus (LC) were examined. Male Fischer-344 rats (N=9) were assigned to six weeks of treadmill running. An additional group of animals comprised the sedentary home cage control group (n=9). Levels of GAL and TH messenger RNA (mRNA) in the LC were measured using *in situ* hybridization histochemistry with autoradiography. Levels of GAL mRNA were higher in treadmill trained animals compared to sedentary animals, but there was no effect of chronic treadmill running on TH mRNA. These results suggest that gene expression for galanin is responsive to chronic exercise stress.

Keywords: *in situ* hybridization, noradrenergic, stress, tyrosine hydroxylase

Prepro-galanin mRNA Levels Are Increased in Rat Locus Coeruleus
After Treadmill Exercise Training

Noradrenergic neurons project extensively throughout the brain and originate predominantly from cell bodies within the locus coeruleus (LC) complex in the brain stem [3,10]. The LC noradrenergic system comprises a diffuse network that regulates attention, vigilance, and arousal [1,10], and noradrenergic neurons in the LC are particularly sensitive to stressful stimuli. Acute stress is accompanied by increased brain norepinephrine (NE) release [23], and prolonged exposure to a severe stressor may result in depletion of NE [9,24]. However, repeated exposure to a stressor blunts the depletion of NE, indicating the acquisition of a resistance to stress [24].

Mechanisms to explain such a resistance to repeated stress are, as yet, not fully known. Potential adaptations of the central noradrenergic response to stress include changes in gene expression for tyrosine hydroxylase (TH), the rate-limiting enzyme for NE synthesis [23,26], and the neuropeptide galanin [14,23,25]. Galanin is a 29-amino acid peptide neurotransmitter that coexists with norepinephrine in approximately 80% of LC neurons [13]. Galanin hyperpolarizes noradrenergic neurons and inhibits locus coeruleus firing in vitro [19,22]. Thus, a possible function of galanin is to provide feedback inhibition to LC neurons [15]. Increases in both TH and prepro-galanin (GAL) messenger RNA (mRNA) have been observed after exposure to chronic stress or reserpine-induced NE depletion [2,14]. Although effects have varied according to the type and severity of the stressor, alterations in gene transcription for TH and/or galanin represent plausible mechanisms for noradrenergic adaptations to stress.

Acute and chronic exercise influence noradrenergic activity. Acute treadmill running results in increased discharge by LC neurons [20], whereas increased levels of extracellular NE in brain frontal cortex observed after acute exercise [17] are lower after a familiar (i.e., repeated) compared to a novel duration of treadmill running [18]. Though forced treadmill running decreases NE levels in the brainstem [12], repeated exposure to

exercise by treadmill training or chronic activity wheel running is associated with elevated post-mortem basal levels of brain NE [7,8].

In addition, Soares et al. [23] reported that chronic activity wheel running led to blunted levels of extracellular NE in brain frontal cortex relative to sedentary animals in response to acute footshock stress. Although effects were not statistically significant, activity wheel animals also exhibited a 1 *SD* higher level of GAL mRNA after footshock compared to sedentary rats, but there was no group difference in TH mRNA. Because galanin appears to modulate noradrenergic activity [15], this difference in gene expression for galanin might be involved in the attenuation of NE release observed in exercise-trained animals.

The purpose of the present study was to examine whether treadmill exercise training induces elevations in gene expression for prepro-galanin and TH mRNA in the LC under basal resting conditions. Because treadmill running activates LC neurons, we reasoned that treadmill exercise training would augment the capacity for inhibitory modulation by galanin of LC noradrenergic response to treadmill running. Thus, we hypothesized that repeated bouts of running during treadmill training would result in elevated levels of the gene that encodes galanin. Based on our previous research [23], we did not expect alterations in gene expression for TH.

Methods and Procedures

Subjects. Young (45d) male Fischer-344 rats (N=18) were purchased from Charles River (Raleigh, NC) and allowed to adapt to the vivarium for 2 weeks. Animals were housed in individual polypropylene cages in a temperature (23 ± 1 °C) and humidity-controlled vivarium maintained on a 12 h light/dark schedule (light 0700-1900). Animals had free access to food and water and were handled and weighed daily throughout the study. All procedures were conducted in accordance with the American Psychological Association ethical standards in the care and use of animals in research and

were approved by the University of Georgia Animal Care and Use of Laboratory Animals Committee.

Treadmill Training. After acclimatization to the vivarium, animals were familiarized to the treadmill and screened for exercise trainability by gradually increasing treadmill speed and session duration over a 2-week accommodation period.   In our experience, 10-30% of albino rats will not perform well enough during treadmill running to induce an increase in physical fitness. To judge exercise trainability during the accommodation period, investigators rated running performance on a scale of 1 (worst) to 5 (best) [28]. Animals with a mean rating of 3 or higher were considered to be trainable and were assigned to the treadmill training group (n=9).   In our experience this selection procedure does not bias behavioral and hypothalamic-pituitary-adrenocortical responses to stress [4,28]. Remaining animals comprised the sedentary home cage control group (n=9). Treadmill training continued for six additional weeks (40 minutes/day, 25m/min, 0 degree incline, 6 days/week). Electric shock was not used to aid running performance. This protocol reliably increases physical fitness in the Fischer rat as indicated by the oxidative capacity of locomotory skeletal muscle [6]. Six animals completed the training protocol and comprised the treadmill-trained group for analyses.

In Situ Hybridization. Twenty-four hours after the final training session, animals were killed by rapid decapitation, and brains were removed and stored at –80° C. Levels of prepro-GAL mRNA in the LC were measured using *in situ* hybridization histochemistry with autoradiography. Twelve-micron sections of brain were cut using a Microm cryostat (Carl Zeiss, Walldorf, Germany) and thaw-mounted on gelatin-coated microscope slides. Approximately every tenth section was stained with 0.1% thionin to visualize anatomical landmarks for region identification. Four sections per probe were selected for hybridization. Sections were fixed in 4% formaldehyde in 0.12 M sodium phosphate-buffered saline (PBS, pH 7.4) for 5 min, rinsed twice in PBS, and placed in 0.25% acetic anhydride in 0.1M triethanolamine HCl/0.9% NaCL (pH 8.0) for 10 min. Sections were dehydrated using a

series of ethanol washes (70%, 80%, 95%, 100%), delipidated in chloroform for 5 min,

rinsed in ethanol (100%, 95%), and allowed to dry.   Oligonucleotide probes were obtained

from Oligos Etc. (Wilsonville, OR). The GAL probe was complementary to bases 115-153

of porcine prepro-GAL mRNA [21], and the TH probe was complementary to bases 1441-

1488 of rat TH mRNA [11]. Probes were labeled at the 3'-end with [^{35}S]-dATP (1000-1500

mCi/mmol; New England Nuclear, Boston, MA), terminal deoxynucleotidyl transferase

(TdT) (15 units/ml; Bethesda Research Lab, Gaithersburg, MD), and tailing buffer. Column

separation was used to separate unincorporated nucleotides from the probes. Sections were

hybridized with radiolabeled probes in solution containing 50% formamide, 600 mM NaCl,

80 mM Tris-HCl, 4 mM EDTA, 0.1% sodium pyrophosphate, 0.2% SDS, 0.2 mg/ml

heparin sulfate, and 10% dextran sulfate. Sections were incubated with the hybridization

solution for 20 hours in a humid chamber at 37°C. Sections then underwent a series of

washes (3 1xSSC, 1 2xSSC/50% formamide, 3 2xSSC/50% formamide for 20 min at 40°C,

2 1xSSC for 30 min at 22°C) to reduce nonspecific binding. Slides were rinsed in deionized

water and 70% ethanol and allowed to dry. Sections were exposed to autoradiographic film

(BioMax MR, Eastman Kodak, Rochester, NY) and developed with Kodak GBX

developer and fixer.

Data Analysis. Autoradiographic films were analyzed using a computerized

image analysis system (Image 1.38 software, Rasband, 1995, National Institute of Mental

Health; Power Macintosh 8100 computer, Apple Computer, Cupertino, CA; light box and

camera, Imaging Research, St. Catharines, Ontario; video interface, Data Translation,

Marlboro, MA) to determine optical density within the LC. The area was analyzed by

tracing the LC with a 1-mm diameter circle and measuring optical gray scale values

within the region. Investigators were blinded to subject condition at the time of

quantification. Data were analyzed by a 1-tailed independent samples t-test using SPSS

Windows version 9. (SPSS, Inc.,Chicago, IL). GAL mRNA data were not available for 1

subject, and missing data were not replaced for analyses. Effect sizes are reported as

Cohen's d (treatment mean-control mean/pooled SD) and Ω^2. Sample size provided a statistical power of .80 for detecting a group difference of 1.5 SD at an alpha of p <.05.

Results

Group differences in body mass (mean ± SD) were observed between treadmill-trained animals (212.7 ± 5.7 g) and sedentary animals (230.4 ± 10.8 g); trained animals had significantly lower body mass relative to sedentary animals, t (13) = 3.68, p=.0015, d=-1.93, Ω^2=-.45 . Level of GAL mRNA was higher in treadmill-trained animals compared to sedentary animals, t (11) = 2.05, p=.03, d=1.17, Ω^2=.20. (Fig. 1 and 2). There was no difference between groups in level of TH mRNA, t (13) = .26, p =.40, d = -.14, Ω^2= -.07 (Fig. 3).

Discussion

The increase in expression of GAL mRNA in the LC after treadmill exercise training observed in the present study is consistent with prior research demonstrating adaptations in the brain noradrenergic system after chronic exercise. For example, Dunn et al. [8] reported that treadmill training and chronic activity wheel running each increased NE levels in the pons which contains NE cell bodies. Exercise training has also been shown to blunt the depletion of NE in response to stress in rats. Dishman et al. [5] found that NE levels in the LC of activity wheel animals were 60% higher than sedentary control animals after uncontrollable footshock. The elevations in norepinephrine levels in the LC of exercise groups were consistent with a blunted release of NE.

We have reported previously that wheel runners exhibited blunted extracellular brain cortical NE levels and higher GAL mRNA levels in the LC after footshock compared to control animals [23]. However, there was no difference in TH mRNA levels between sedentary and trained animals. The alterations in gene expression for galanin after chronic exercise are consistent with studies that demonstrate changes in galanin activity with acute and chronic stress. Acute administration of the catecholamine-depleting drug reserpine has been shown to increase TH and GAL mRNA in LC neurons

[2]. Holmes et al. [14] examined the effect of chronic social stress on galanin gene expression in the LC and observed that subordinate rats exhibited elevated GAL mRNA levels compared to dominant and control animals. In addition, a positive correlation was found between GAL mRNA and severity of the stress as indicated by number of wounds. Increases in GAL mRNA levels in the LC have also been observed after lesioning of a terminal NE field by olfactory bulbectomy [16]. However, Austin et al. [2] found no change in either GAL or TH mRNA in the LC after mild swim stress. Thus, adaptations in the LC noradrenergic system and changes in gene expression for galanin and TH may be dependent upon the nature and severity of the stressor.

The results of the present study suggest that basal gene expression for galanin is responsive to chronic exercise stress. Our findings of increased GAL mRNA and no change in TH mRNA in the LC after chronic exercise are thus consistent with previous research using chronic stress models. However, previous studies examining GAL mRNA and voluntary exercise found no difference in basal levels between sedentary and activity wheel animals [23]. This discrepancy might be explained by the forced nature or higher relative intensity of the exercise stimulus associated with the treadmill training protocol [6,8]. Though it has been proposed that increases in galanin activity may result in deleterious changes in brain function associated with chronic stress [27], it is equally plausible that alterations in galanin within the LC may play an adaptive role in regulating responsiveness of the noradrenergic system to stress.

Additional research is needed to elucidate the role of galanin in brain neurotransmission to determine whether this peptide contributes to neuropathology associated with chronic stress or whether galanin serves as a mechanism for adaptive regulation of brain activity. Because galanin-positive neurons within the LC tend to be dendrodendritically arranged and galanin hyperpolarizes LC neurons [19], it is plausible that upregulation of galanin within the LC may serve as a negative feedback mechanism for inhibition of NE release.

The present study examined gene expression for two regulatory components of noradrenergic function, TH and galanin. However, it is not known whether the observed elevations in GAL mRNA resulted in post-translational changes, as neither galanin release nor concentration was measured. It is also possible that noradrenergic responsiveness after exercise training is altered by other mechanisms not examined in the present study (e.g., NE transporters, receptors, etc.). Additional studies are needed to examine whether the increase in GAL mRNA within the LC represents enhanced galanin activity and whether galanin released from LC neurons acts on autoreceptors to directly inhibit NE release. Further research is also needed to determine the characteristics of the stressor required to induce changes in galanin transmission and to determine the time course of such effects.

In conclusion, the increase in gene expression for galanin in treadmill exercise-trained animals might indicate an autoregulatory function of the neuropeptide in brain noradrenergic responsiveness during exercise stress. Provided that the increase in GAL mRNA induces post-translational increases in galanin activity within LC neurons, adaptations in galanin represent a plausible mechanism to explain the blunted noradrenergic response exhibited by trained animals during both homotypic (i.e., familiar acute exercise) and heterotypic (i.e., novel footshock) stress [23].

References

[1]   Aston-Jones, G., Rajkowski, J., & Cohen, J., Role of locus coeruleus in attention and behavioral flexibility, Biol. Psychiat., 46 (1999) 1309.

[2]   Austin, M.C., Cottingham, S.L., Paul, S.M., & Crawley, J.N., Tyrosine hydroxylase and galanin mRNA levels in the locus coeruleus neurons are increased following reserpine administration, Synapse, 6 (1990) 351.

[3]   Dahlstrom, A. & Fuxe, K., Evidence for the existence of monoamine-containing neurons in the central nervous system, Acta Physiol. Scand., 62(suppl. 232) (1964) 1.

[4]   Dishman, R.K, Armstrong, R.B., Delp, M.D., Graham, R.E., & Dunn, A.L., Open-field behavior is not related to treadmill performance in exercising rats, Physiol. Behav., 43 (1988) 541.

[5]   Dishman, R.K., Renner, K.J., Youngstedt, S.D., Reigle, T.G., Bunnell, B.N., Burke, K.A., Yoo, H.S., Mougey, E.H., & Meyerhoff, J.L., Activity wheel running reduces escape latency and alters brain monoamine levels after footshock, Brain Res. Bull., 42 (1997) 399.

[6]   Dishman, R.K., Warren, J.M., Hong, S., Bunnell, B.N., Mougey, E.H., Meyerhoff, J.L., Jaso-Friedmann, L., & Evans, D.L., Treadmill exercise training blunts Natural Killer Cell cytolysis after footshock, J. Appl. Physiol., (2000) in press.

[7]   Dunn, A.L. & Dishman, R.K., Exercise and the neurobiology of depression, Exerc. Sport Sci. Rev., 19 (1991) 41.

[8]   Dunn, A.L, Reigle, T.G., Youngstedt, S.D., Armstrong, R.B., & Dishman, R.K., Brain norepinephrine and metabolites after treadmill training and wheel running in rats, Med. Sci. Sports Exerc., 28 (1996) 204.

[9]   Fillenz, M., Noradrenergic Neurons, Cambridge University Press, New York, 1990.

[10] Foote, S.L., Bloom, F.E., & Aston-Jones, G., Nucleus locus ceruleus: New evidence of anatomical and physiological specificity, Physiol. Rev., 63 (1983) 844.

[11] Grima, B., Lamouroux, A., Blanot, F., Faucon-Biguet, N., & Mallet, J., Complete coding sequence of rat tyrosine hydroxylase mRNA, Proc. Natl. Acad. Sci., 82 (1985) 617.

[12] Heyes, M.P., Garnett, E.S., & Coates, G., Nigrostriatal dopaminergic activity is increased during exhaustive exercise stress in rats, Life Sci., 42 (1988) 1537.

[13] Holets, V.R., Hokfelt, T., Rokaeus, A., Terenius, L., & Goldstein, M., Locus coeruleus neurons in the rat containing neuropeptide Y, tyrosine hydroxylase or galanin and their efferent projections to the spinal cord, cerebral cortex and hypothalamus, Neuroscience, 24 (1988) 893.

[14] Holmes, P.V., Blanchard, D.C., Blanchard, R.J., Brady, L.S., Crawley, J.N., Chronic social stress increases levels of preprogalanin mRNA in the rat locus coeruleus, Pharmacol. Biochem. Behav., 50 (1995) 655.

[15] Holmes, P.V. & Crawley, J.N., Coexisting neurotransmitters in central noradrenergic neurons, In F.E. Bloom & D.J. Kupfer (Eds.), Psychopharmacology: The Fourth Generation of Progress, Raven Press, Ltd, New York, 1995.

[16] Holmes, P.V. & Crawley, J.N., Olfactory bulbectomy increases prepro-galanin mRNA levels in the rat locus coeruleus, Mol. Brain Res., 36 (1996) 184.

[17] Pagliari, R. & Peyrin, L., Norepinephrine release in the rat frontal cortex under treadmill exercise: a study with microdialysis, J. Appl. Physiol., 78 (1995) 2121.

[18] Pagliari, R. & Peyrin, L., Physical conditioning in rats influences the central and peripheral catecholamine responses to sustained exercise, Eur. J. Appl. Physiol., 71 (1995) 41.

[19] Pieribone, V.A., Xu, Z.Q., Zhang, X., Grillner, S., Bartfai, T., & Hokfelt, T., Galanin induces a hyperpolarization of norepinephrine-containing locus coeruleus neurons in the brainstem slice, Neuroscience, 64 (1995) 861.

[20] Rasmussen, K., Morilak, D.A., & Jacobs, B.L., Single unit activity of locus coeruleus neurons in the freely moving cat: I. during naturalistic behaviors and in response to simple and complex stimuli, Brain Res., 371 (1986) 324.

[21] Rokaeus, A. & Brownstein, M.J., Construction of a porcine adrenal medullary cDNA library and nucleotide sequence analysis of two clones encoding a galanin precursor, Proc. Natl. Acad. Sci., 83 (1986) 6287.

[22] Seutin, V., Verbanck, P., Massote, P., & Dresse, A., Galanin decreases the activity of locus coeruleus neurons in vitro, Eur. J. Pharmacol., 164 (1989) 373.

[23] Soares, J., Holmes, P.V., Renner, K.J., Edwards, G.L., Bunnell, B.N., & Dishman, R.K., Brain noradrenergic responses to footshock after chronic activity-wheel running., Behav. Neurosci., 113 (1999) 558.

[24] Stanford, S.C., Central noradrenergic neurones and stress, Pharmacol..Ther., 68 (1995) 297.

[25] Sweerts, B.W., Jarrott, B., & Lawrence, A.J., Expression of preprogalanin mRNA following acute and chronic restraint stress in brains of normotensive and hypertensive rats, Mol. Brain Res., 69 (1999) 113.

[26] Wang, P., Kitayama, I., & Nomura, J., Tyrosine hydroxylase gene expression in the locus coeruleus of depression-model rats and rats exposed to short- and long-term forced walking stress, Life Sci., 62 (1998) 2083.

[27] Weiss, J.M., Bonsall, R.W., Demetrikopoulos, M.K., Emery, M.S., & West, C.H.K., Galanin: a significant role in depression? Ann. NY Acad. Sci., 863 (1998) 364.

[28]   White-Welkley, J.E., Bunnell, B.N., Mougey, E.H., Meyerhoff, J.L., & Dishman, R.K., Treadmill exercise training and estradiol differentially modulate hypothalamic-pituitary-adrenal cortical responses to acute running and immobilization, Physiol. Behav., 57 (1995) 533.

Figure 1. Representative autoradiographs of rat brain sections hybridized for prepro-galanin mRNA in the locus coeruleus (LC). Rats were treadmill exercise trained (A) or sedentary home-cage control (B).
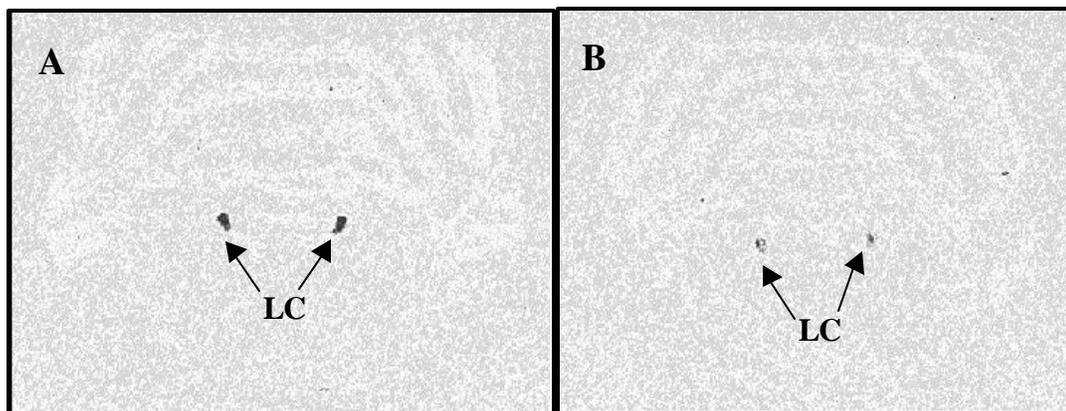
Figure 2. Levels of prepro-galanin (GAL) mRNA levels in the locus coeruleus were higher in treadmill exercise trained animals compared to sedentary home-cage control (*p=.03). Values are means ± SD. OGS= Optical Gray Scale
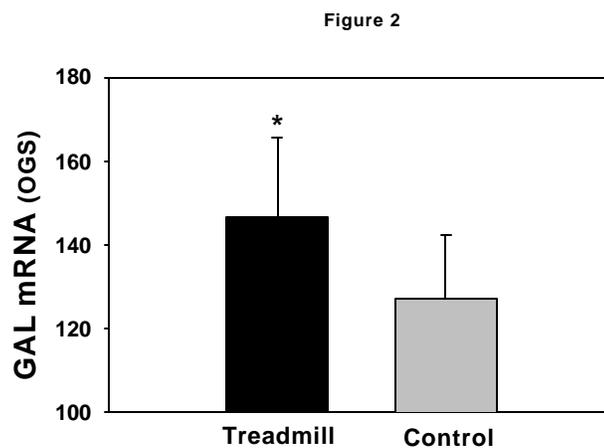
Figure 3. Levels of tyrosine hydroxylase (TH) mRNA in the locus coeruleus did not differ between treadmill exercise trained animals and sedentary home-cage control (p=.40). Values are means ± SD.   OGS= Optical Gray Scale
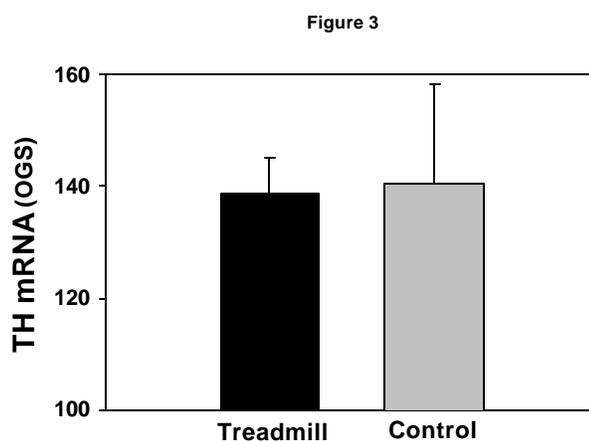
EFFECTS OF CHRONIC ACTIVITY WHEEL RUNNING OR ANTIDEPRESSANT

PHARMACOTHERAPY ON APPETITIVE BEHAVIOR AND GENE EXPRESSION

IN RAT LOCUS COERULEUS AFTER OLFACTORY BULBECTOMY[3]

---

[3]O'Neal, H.A., Van Hoomissen, J.D, Holmes, P.V., Bunnell, B.N. & Dishman, R.K.
2000. To be submitted to Behavioral Neuroscience.

Abstract

We examined the effects of chronic activity wheel running and imipramine administration on appetitive behavior and gene expression for noradrenergic modulatory proteins in neurons of the locus coeruleus after olfactory bulbectomy. Male Long-Evans rats were randomly assigned to the following conditions using a 2 x 2 x 2 design: (1) bilateral olfactory bulbectomy or sham surgery, (2) voluntary activity wheel running or sedentary home cage, and (3) daily imipramine or saline injections. After 21 days of treatment, animals underwent behavioral testing for sucrose preference and copulatory activity. *In situ* hybridization histochemistry with autoradiography was used to measure levels of tyrosine hydroxylase (TH), prepro-galanin (GAL), and prepro-neuropeptide Y (NPY) messenger RNA (mRNA). Bulbectomized animals exhibited reductions in sucrose intake and decrements in copulatory performance compared to sham animals. In addition, imipramine administration was associated with decrements in copulatory performance as evidenced by lower rates of copulation and reduced ejaculation frequency. However, activity wheel running in bulbectomized animals improved copulation rates, reduced ejaculation latency, and increased ejaculation frequency. Levels of GAL mRNA were increased after imipramine treatment. Activity wheel running after olfactory bulbectomy was associated with elevations in NPY mRNA. No differences were found in gene expression for TH. The results suggest that adaptations in noradrenergic function after three weeks of imipramine treatment and activity wheel running are not regulated by similar mechanisms of gene expression within the locus coeruleus.

Effects of Chronic Activity Wheel Running or Antidepressant Pharmacotherapy on

Appetitive Behavior and Gene Expression in Rat Locus Coeruleus

after Olfactory Bulbectomy

Though the etiology of depressive disorders has not been well-established, there is abundant evidence to suggest involvement of the central noradrenergic system in the pathology of depression (Leonard, 1997). Noradrenergic neurons originate primarily from cell bodies within the locus coeruleus (LC) complex in the brain stem and project extensively throughout the brain (Dahlstrom & Fuxe, 1964; Foote, Bloom, & Aston-Jones, 1983) to regulate attention and arousal in response to stressful stimuli (Aston-Jones, Rajkowski, & Cohen, 1999).

The present study examined the effects of chronic activity wheel running and imipramine antidepressant treatment on gene expression for tyrosine hydroxylase (TH), prepro-galanin (GAL), and prepro-neuropeptide Y (NPY) messenger RNA (mRNA) in LC neurons using the olfactory bulbectomy model of depression. Bilateral olfactory bulbectomy (OBX) in the rat results in a well-established syndrome of behavioral, biochemical, and physiological features that are reversed by chronic antidepressant treatment (Kelly, Wrynn, & Leonard, 1997). Characteristics of the OBX syndrome that have relevance for the study of depressive disorders include reduced brain cortical catecholamine concentrations (Cairncross, Schofield, & Bassett, 1975; Redmond, Kelly, & Leonard, 1997), increased density of β-adrenoreceptors (Leonard, 1997) altered circadian rhythms (Lumia, Teicher, Salchli, Ayers, & Possidente, 1992), impaired learning (Grecksch, Zhou, Franke, Schroder, Sabel, Becker, & Huether, 1997), elevated plasma corticosterone levels (Marcilhac, Maurel, Anglade, Ixart, Mekaouche, Hery, & Siaud, 1997), hyperactivity in an open field (Redmond et al., 1997), and attenuated exhibition of motivated behaviors (Calcagnetti, Quatrella, & Schechter, 1996).

Exercise interventions are associated with reductions in depressive symptoms in humans (O'Neal, Dunn, & Martinsen, 2000), and chronic physical activity in animals induces adaptations in central noradrenergic function that may have relevance for the treatment of depressive disorders (Dunn & Dishman, 1991). Acute treadmill running results in increased discharge by LC neurons (Rasmussen, Morilak, & Jacobs, 1986) and decreased brain NE levels (Heyes, Garnett, & Coates, 1988). However, repeated exposure to exercise by treadmill training or chronic activity wheel running is associated with elevated post-mortem levels of brain NE, including regional increases in the pons and LC cell bodies (Dishman, Renner, Youngstedt, Reigle, Bunnell, Burke, Yoo, Mourgey, & Meyerhoff, 1997; Dunn, Reigle, Youngstedt, Armstrong, & Dishman, 1996). Because commonly prescribed antidepressant drugs act to increase availability of brain norepinephrine (NE) (Charney, 1998), it is plausible that adaptations within the LC that facilitate noradrenergic transmission could modulate antidepressant effects of physical activity.

Although exercise has been postulated to exert antidepressant effects by modulating noradrenergic activity (Dunn & Dishman, 1991), to our knowledge, only one other study has used an endogenous animal depression model to examine neurobiological adaptations after chronic exercise that might explain the mental health benefits of exercise. Yoo, Tackett, Crabbe, Bunnell, & Dishman (2000) examined the effects of chronic activity wheel running, treadmill training, and imipramine treatment on brain noradrenergic responses in animals treated neonatally with clomipramine to model depression. Exercise-trained and imipramine-treated animals had higher brain cortical norepinephrine levels relative to control animals, and activity wheel runners and imipramine-treated animals exhibited downregulation of β-adrenoreceptors in frontal cortex. Thus, chronic exercise in rats has been shown to produce favorable adaptations in noradrenergic function, and effects were similar to chronic antidepressant administration. The present study was conducted to further investigate noradrenergic adaptations to

chronic exercise in a depression model by examining gene expression of modulatory proteins in the LC.

Within neurons of the LC, alterations in gene expression for synthetic enzymes and modulatory neuropeptides represent a plausible mechanism for noradrenergic adaptations throughout the brain. First, norepinephrine availability may be mediated directly by TH, the rate-limiting enzyme for NE synthesis; therefore, changes in synthetic capacity via TH mRNA may enhance norepinephrine transmission. In addition, the neuropeptides galanin and neuropeptide Y, which are colocalized with norepinephrine, may act on LC neurons to influence noradrenergic transmission. Galanin is a 29-amino acid peptide neurotransmitter that coexists with norepinephrine in approximately 80% of LC neurons (Holets, Hokfelt, Roakaeus, Terenius, & Goldstein, 1988). Neuropeptide Y is a 36-amino acid peptide colocalized with NE in around 40% of LC neurons (Holets et al., 1988; Xu, Shi, & Hokfelt, 1998). Both GAL and NPY inhibit locus coeruleus firing in vitro (Pieribone, Xu, Zhang, Grillner, Bartfai, & Hokfelt, 1995; Seutin, Verbanck, Massote, & Dresse, 1989; Finta, Regenold, & Illes, 1992; Illes & Regenold, 1990). Thus, a possible function of these neuropeptides is to provide feedback inhibition to LC neurons (Holmes & Crawley, 1995), and basal alterations in gene expression within LC neurons could contribute to noradrenergic adaptations to both physical activity and antidepressant treatment. Furthermore, a similarity in response of mRNA to exercise and imipramine administration would provide evidence for the biological plausibility of exercise as an antidepressant intervention.

Implications for the role of gene expression in the etiology and treatment of depression are limited, as most studies examining the effects of antidepressant treatment on gene expression have not used an animal model of depression. Moreover, we are unaware of studies that have examined the effect of voluntary exercise on mRNA levels using a depression model. The purpose of the present study was to examine gene expression for TH, GAL, and NPY in the LC after chronic antidepressant treatment and

voluntary activity wheel running in olfactory bulbectomized animals. Behavioral assessments, including measurement of sucrose preference and male copulatory activity, were also conducted to examine appetitive features of the OBX syndrome after chronic exercise and antidepressant administration. It was hypothesized that chronic activity wheel running would induce adaptations in gene expression within the LC and that effects would be similar to those of antidepressant treatment. Activity wheel runners were also expected to exhibit increases in appetitive behaviors. Both pharmacological and exercise interventions were hypothesized to reduce depressive-like features of olfactory bulbectomy. However, the effects of drug and physical activity were not expected to be additive.

Methods and Procedures

Subjects. Ninety-six male Long Evans rats (aged 45 d) were used in the experiment, and an additional group of 18 female Long Evans rats (aged 65 d) were obtained for sexual activity testing (Harlan Inc., Indianapolis, IN). A continuous weekly protocol of animal intake, treatment, and testing (12 animals/week, 8 weeks) was implemented to avoid aging effects. Animals were allowed to adapt to the animal facilities for 2 weeks prior to experimental manipulations. Animals were housed in individual polypropylene cages in a temperature ($23 \pm 1\ °C$) and humidity-controlled vivarium that was maintained on a 12 h light/dark schedule (light 0600-1800). Food and water were available *ad libitum*. Experimental animals were handled daily and weighed three times weekly throughout the study. All procedures were conducted in accordance with the American Psychological Association guiding principles in the care and use of animals and were approved by the University of Georgia Animal Care and Use Committee.

Research Design. A 2 x 2 x 2 factorial design was used in the study. The independent variables were 1) Group (olfactory bulbectomy vs. sham) 2) Condition (activity wheel running vs. sedentary) 3) Treatment (imipramine vs. saline). The

dependent measures included sucrose preference score, male copulatory behaviors, and levels of TH, GAL, and NPY mRNA in the LC.

Experimental Procedures

Group. Experimental male rats received bilateral olfactory bulbectomy (OBX) or sham surgery (SHAM). Rats were anesthetized with intraperitoneal injections of 25 mg/kg Nembutal (Abbott Laboratories) and 40 mg/kg ketamine hydrocholoride (Mallinckrodt). A 1-cm incision was made on the scalp midline, and burr holes 2-mm in diameter were drilled through the skull (coordinates: 6 mm anterior to bregma, 1 mm lateral to midline). Olfactory bulbs were aspirated using a curved plastic pipette tip 2-mm in diameter, and cavities were filled with gel-foam (Upjohn) to control bleeding. Animals in the sham surgery condition received burr holes only. A 1-cc subcutaneous injection of saline to replace fluids and 1.0 mg/kg intraperitoneal injection of Banamine for pain were administered after surgery. The incision was closed with vicryl sutures, and animals were placed under heating lamps until recovery from anesthesia. Removal of the olfactory bulbs was verified upon dissection, and OBX rats with remaining olfactory bulb tissue exceeding 30% of SHAM animals were excluded from the study.

Condition. Rats were randomly assigned to either the activity wheel (AW) or sedentary (SED) condition. Twenty-four hours after surgery, activity wheel animals were placed in shoebox cages with 24-hour access to a running wheel (circumference = 105 cm) connected to an electromagnetic revolution counter. Daily running distance for each subject was determined by multiplying wheel circumference by the number of revolutions recorded by the counter.

Treatment. Animals assigned to the imipramine treatment group (IMI) received daily intraperitoneal injections of 10 mg/kg imipramine hydrocholoride (Sigma). An equivalent volume of saline was injected into all other animals (SAL). Injections were initiated 24 hours post-surgery and continued for 21 days.

Behavioral Measures

     Sucrose Preference. After the 21-day treatment period, all animals underwent behavioral testing for sucrose consumption during the dark phase. Testing was initiated 12 hours after the final injection at the onset of the dark phase (1800h) and concluded 14 hours after the beginning of the dark phase (0800h). Each rat was given access to two bottles filled with 50 ml of liquid (1% sucrose and tap water). Bottles were fitted with special spouts to minimize leakage, and placement of bottles was counterbalanced to control for position preference. The amount of liquid remaining in each bottle was recorded at the end of the testing period. A sucrose preference score was calculated as percent of total intake (sucrose preference = sucrose intake/total intake x 100).

     Male Copulatory Behavior. All animals underwent copulatory testing 24 hours after the final injection and/or exercise session. Test sessions were conducted under dim red light illumination in a thermoneutral testing room during the dark phase (1800h – 2400h). Female receptivity was induced by intramuscular injection of estradiol (50 μg, 72 hr pre-test) and progesterone (0.5 mg, 3-6 hr pretest), and receptivity was confirmed using non-experimental male rats prior to testing. Each male rat was placed in a circular Plexiglass testing chamber (24 in diameter) and allowed to adapt to the testing environment for 5 minutes. The sexually receptive female was introduced to the chamber, and male copulatory behaviors were recorded by raters blinded to subject condition. Each session lasted 30 minutes after the first intromission or 20 minutes if no activity was observed.

     The copulatory behaviors that were measured include the following: mount latency (ML: period of time between the introduction of the female and the first mount); mount frequency (MF: total number of mounts observed); intromission latency (IL: period of time until the first intromission); intromission frequency (IF: total number of intromissions observed); ejaculation frequency (EF: total number of ejaculations; and post-ejaculatory interval (PEI: period of time from the first ejaculation until the next intromission). The

behaviors of ML, IL, and PEI were considered as indicators of sexual motivation or arousal while MF, IF, and EF were indicative of copulatory performance (Dewsbury, 1967).

In Situ Hybridization. Twenty-four to 72 hours after conclusion of behavioral testing, animals were killed by decapitation under Halothane inhalation anesthesia. Brains were removed, frozen in dry ice, and stored at -80°C. Twelve micrometer sections of brain containing the locus coeruleus were cut using a Microm cryostat (Carl Zeiss, Walldorf, Germany) and thaw-mounted on gelatin-coated microscope slides. Approximately every tenth section was stained with 0.1% thionin to allow visualization of anatomical landmarks for region identification. Four brain sections per probe were selected for hybridization. Sections were fixed in 4% formaldehyde in 0.12 M sodium phosphate-buffered saline (PBS, pH 7.4) for 5 min, rinsed twice in PBS, and placed in 0.25% acetic anhydride in 0.1M triethanolamine HCl/0.9% NaCL (pH 8.0) for 10 min. Sections were dehydrated using a series of ethanol washes (70%, 80%, 95%, 100%), delipidated in chloroform for 5 min, rinsed in ethanol (100%, 95%), and allowed to dry. Oligonucleotide probes were obtained from Oligos Etc. (Wilsonville, OR). The GAL probe was complementary to bases 115-153 of human prepro-GAL mRNA (Rokaeus & Brownstein, 1986), the TH probe was complementary to bases 1441-1488 of rat TH mRNA (Grima, Lamouroux, Blanot, Faucon-Biguet, & Mallet, 1985), and the NPY probe was complementary to bases 3146-3194 of rat prepro-NPY mRNA (Larhammer, Ericsson, & Persson, 1987). Probes were labeled at the 3'-end with [^{35}S]-dATP (1000-1500 mCi/mmol; New England Nuclear, Boston, MA), terminal deoxynucleotidyl transferase (TdT) (15 units/ml; Bethesda Research Lab, Gaithersburg, MD), and tailing buffer. Column separation was used to separate unincorporated nucleotides from the probes. Sections were hybridized with radiolabeled probes in solution containing 50% formamide, 600 mM NaCl, 80 mM Tris-HCl, 4 mM EDTA, 0.1% sodium pyrophosphate, 0.2% SDS, 0.2 mg/ml heparin sulfate, and 10% dextran sulfate. Sections were incubated with the hybridization solution for 20 hours in a humid chamber at 37°C. Sections then underwent a series of washes (3 1xSSC, 1

2xSSC/50% formamide, 3 2xSSC/50% formamide for 20 min at 40°C, 2 1xSSC for 30 min at 22°C) to reduce nonspecific binding. Slides were rinsed in deionized water and 70% ethanol and allowed to dry. Sections were exposed to autoradiographic film (BioMax MR, Eastman Kodak, Rochester, NY) and developed with Kodak GBX developer and fixer.

Data Analysis. Autoradiographic films were analyzed using a computerized image analysis system (Image 1.38 software, Rasband, 1995, National Institute of Mental Health; Power Macintosh 8100 computer, Apple Computer, Cupertino, CA; light box and camera, Imaging Research, St. Catharines, Ontario; video interface, Data Translation, Marlboro, MA) to determine optical density within the LC. Data were analyzed using SPSS Windows version 9.0 (SPSS, Inc., Chicago, IL). Group differences in weekly body weight, weekly kilometers run, copulatory behaviors, and mRNA levels were tested by 3-way ANOVA (p<.05), and effect sizes were estimated by η^2. Results of the ANOVA were decomposed using the method of contrasts. Interrater agreement for copulatory data was estimated by intraclass correlation (R_I), and a directional test of proportions (Ferguson, 1966) was conducted to examine differences in copulatory rates. A power analysis was used to determine sample size based on an expected mean effect size of 1.0 *SD.* In this study, 8 animals per cell provided a statistical power of .80 at an α of p<0.05. Values are reported as means ± SD in the text and means ± SE in the figures.

Results

Subject Characteristics. The ANOVA for average body weight indicated a significant main effect for Group, $F(1,73) = 21.13$, p=.001, η^2=.22. Body weight was higher for SHAM animals (286 ± 21 g) than OBX animals (265 ± 22 g). The mean daily distance run during the 3-week period was approximately 3.5 ± 2 km. There were no significant differences in average distance run for Group, Treatment, or their interactions (p>.45).

Sucrose Consumption. The ANOVA for sucrose preference score indicated that main effects and interactions were not significant (p>.11). However, there was a significant

positive correlation between average distance run and sucrose preference score (r=.32, p=.04). The ANOVA for total sucrose consumption indicated a significant main effect for Group, $F(1,72) = 10.04$, p=.002, η^2=.12 (Figure 1). OBX animals consumed less sucrose solution (22.2 ± 14 ml) than SHAM animals (32.8 ± 15 ml). There were no significant differences in water intake (p>0.16).

Male Copulatory Behaviors. Interrater agreement for copulatory variables was high (R_I>.80). The ANOVA for ejaculation frequency indicated a significant main effect for Group, $F(1,73) = 35.04$, p<.001, η^2=.32. OBX animals exhibited significantly fewer ejaculations (1.03 ± 1.1) relative to SHAM animals (2.37 ± .97). Twenty-three of the 40 (58%) OBX animals and 38 of the 42 (90%) SHAM animals achieved at least one ejaculation. Among SHAM animals, there was no effect of Condition or Treatment. However, 41% of OBX-IMI animals achieved ejaculation compared to 78% in the SAL group (p=.005). In addition, 90% of OBX-SAL animals with access to activity wheels successfully copulated compared to 63% of sedentary OBX-SAL animals (p=.045) (Figure 2). Because some animals failed to initiate copulation, the analysis of remaining copulatory variables included data from only those rats that achieved intromission. The ANOVA for MF and IF indicated that main effects and interactions were not significant (p>.32 and p>.10, respectively). However, a significant Group x Condition x Treatment interaction was found for ML, $F(1,53)=4.57$, p=.04, $\eta^2 = .08$ and IL, $F(1,53) = 6.45$, p =.01, η^2=.11.

The ANOVA for EL indicated a significant Group x Condition interaction $F(1,52)=5.88$, p=.02, $\eta^2 = .10$. OBX animals exhibited longer ejaculation latencies relative to SHAM animals, but activity wheel running significantly attenuated this effect (p=.04) (Figure 3). The ANOVA for PEI indicated a significant main effect for Group $F(1,52) = 4.27$, p=.04, $\eta^2 = .08$ with OBX animals exhibiting significantly longer post-ejaculatory intervals (17.1 ± 1 min) relative to SHAM animals (7.7 ± 1 min) (Figure 4). The ANOVA for total number of intromissions revealed a significant main effect for Condition $F(1,52) = 5.39$, p=.02, $\eta^2 = .09$, and AW animals had significantly more intromissions compared to

SED animals (20.3 ± 8 and 16.3 ± 5, respectively). The ANOVA for total number of ejaculations demonstrated a significant main effect of Group F(1,73) = 35.04, p<.001, η^2=.32 with OBX animals exhibiting fewer ejaculations than SHAM animals (1.0 ± 1 and 2.4 ± 1, respectively). In addition, IMI treatment was associated with a reduction in the number of ejaculations for bulbectomized animals (p=.02) (Figure 5).

mRNA for GAL, NPY, and TH. The ANOVA for GAL mRNA indicated a significant main effect for Treatment F(1,67) = 5.18, p = .03, η^2 = .072 with IMI animals exhibiting higher levels of GAL mRNA in the LC compared to SAL animals (Figures 6 and 9). The ANOVA for NPY revealed a significant Group (OBX vs. SHAM) x Condition (AW vs. SHAM) interaction F(1,62) = 4.06, p= .05, η^2 = .061. Activity wheel running after olfactory bulbectomy significantly increased expression of the gene that encodes NPY relative to the sham surgery condition (p=.04) (Figures 7 and 10). The ANOVA of TH mRNA data revealed no main effects for Group, Condition, Treatment or their interactions (p>0.32) (Figure 8). There were no differences in level of mRNA for GAL, NPY, or TH among copulators versus non-copulators (p>.23). There was also no effect of order of killing on level of mRNA (p>.31).

Discussion

The present study examined responses in appetitive behavior and gene expression to three weeks of chronic activity wheel running and imipramine antidepressant treatment after olfactory bulbectomy in rats. Measurement of sucrose preference and male copulatory activity was used to assess the tendency to engage in motivated behaviors, because a loss of pleasure, or anhedonia, is a core feature of major depression. In the present study, bulbectomized animals exhibited a reduction in sucrose intake, but neither imipramine treatment nor wheel running compensated for the behavioral decrements. However, running distance was positively correlated with sucrose preference score but unrelated to total volume of sucrose consumed. The large variability among animals likely contributed to the failure to detect a significant difference for sucrose preference

score. Additionally, the anosmic component of the olfactory bulbectomy model is problematic for implementation of this measure and may account for the failure to observe increases in consumption after treatment.

Olfactory bulbectomy also significantly attenuated copulation, an effect congruous with prior research (Lumia et al., 1992). It is unlikely that this effect was entirely due to anosmia, because bulbectomized, saline-treated animals with access to activity wheels achieved ejaculation as often as control animals. Not only did a higher proportion of activity wheel animals exhibit successful copulation, but activity wheel animals also had shorter ejaculation latencies and post-ejaculatory intervals as well as greater total intromissions and ejaculations. These results confirm previous findings of an enhanced rate of copulatory performance after activity wheel running in a depression model (Yoo et al., 2000). However, it is not known whether the attenuation of copulatory deficits after activity wheel running is due to physiological adaptations or reflects enhanced motivation. As observed in previous studies (Yoo et al., 2000), imipramine administration was associated with decrements in sexual performance including reduced copulation rates, longer post-ejaculatory intervals and fewer ejaculations. Such disturbances in copulatory behaviors are consistent with reports of sexual dysfunction in clinically depressed patients receiving antidepressant medications (Baldwin, 1995).

Because chronic exercise influences noradrenergic transmission and imipramine acts to inhibit NE reuptake, we examined gene expression for modulatory proteins within cell bodies of the LC. Although prior research has demonstrated an increase in gene expression for GAL in the LC after olfactory bulbectomy (Holmes & Crawley, 1996), we observed no significant differences in levels of GAL mRNA in bulbectomized animals. Gene expression for GAL is responsive to conditions that deplete brain NE such as reserpine treatment (Austin, Cottingham, Paul, & Crawley, 1990) and that activate noradrenergic systems such as chronic social stress (Holmes, Blanchard, Blanchard,

Brady, & Crawley, 1995); thus, the reasons for the lack of effect for GAL mRNA after lesioning of NE terminal fields after bulbectomy are unknown.

Activity wheel running did not alter basal levels of GAL mRNA, an effect that is consistent with prior research (Soares, Holmes, Renner, Edwards, Bunnell, & Dishman, 1999). However, there is evidence that adaptations in gene expression for GAL after chronic exercise may influence noradrenergic transmission. For example, Soares et al. (1999) found no basal differences in GAL mRNA level between activity wheel and sedentary animals, though activity wheel runners exhibited a 1 SD higher level of GAL mRNA after footshock compared to sedentary rats. In addition, six weeks of treadmill training resulted in significant elevations in basal levels of gene expression for GAL compared to sedentary animals (O'Neal, Van Hoomissen, Holmes, & Dishman, 2000). The discrepancy in gene expression for GAL after treadmill training and activity wheel running might be explained by the forced nature or increased intensity of the exercise stimulus associated with the treadmill training protocol (Dunn et al., 1996; White-Welkley, Bunnell, Mougey, Meyerhoff, & Dishman, 1995).

In the present study, only imipramine treatment was associated with increased gene expression for GAL, an effect that is consistent with the action of the drug. Imipramine increases availability of NE by inhibiting reuptake from the synapse, and acute and chronic imipramine administration has been shown to decrease the firing rate of LC neurons and increase NE concentration in frontal cortex (Linner, Arboerlius, Nomiko, Bertilsson & Svensson, 1999). Thus, it is plausible that the effects of imipramine could involve adaptations in galanin activity resulting in feedback inhibition to LC neurons.

Although NPY mRNA levels were unchanged by bulbectomy, activity wheel running, or antidepressant treatment, we observed a significant interaction between the bulbectomy and exercise conditions. These results suggest that activity wheel running after olfactory bulbectomy induces elevations in the gene that encodes NPY in LC

neurons. Adaptations in NPY are of interest as alterations in NPY transmission have been associated with depressive disorders. For example, a reduction in plasma NPY levels have been observed in individuals diagnosed with major depression (Hashimoto, Onishi, Koide, Kai, & Yamagami, 1996) and in patients hospitalized after a recent suicide attempt (Westrin, Ekman, & Traskman-Bendz, 1999). Chronic imipramine treatment also decreases NPY immunoreactivity in rat brain cortex (Smilowska & Legutka, 1991), and desipramine administration reduces NPY mRNA levels within LC neurons (Makino, Baker, Smith, & Gold, 2000). Furthermore, repeated intracerebroventricular administration of NPY may reverse some features of the OBX syndrome including normalization of circadian locomotory behaviors and elevation of NE concentration in the amygdala (Song, Earley, & Leonard, 1996).

The results of the present study are consistent with prior observations by Holmes & Crawley (1996) who found no significant differences in levels of NPY mRNA in rat locus coeruleus after olfactory bulbectomy although there was a tendency for reductions in NPY mRNA 14 days post-surgery. Because gene expression for NPY in the LC was also unchanged by activity wheel running, the interaction observed in the present study could reflect an additive response to olfactory bulbectomy and activity wheel running. Most NPY-immunoreactive cell bodies in the LC project to the hypothalamus (Holets et al., 1988), and a recent study suggests involvement of the hypothalamic-pituitary-adrenal (HPA) axis in the regulation gene expression for NPY. Makino et al. (2000) found that NPY mRNA levels in the LC were increased after 4 days of repeated immobilization stress but were unchanged after 14 days of repeated stress; however, adrenalectomized rats given corticosterone replacement exhibited elevated NPY mRNA levels after the 14-day stress protocol. Because exercise by chronic activity wheel running and treadmill training can modulate HPA function (Dishman, Bunnell, Youngstedt, Yoo, Mougey, & Meyerhoff, 1998; White-Welkley et al., 1995), it is possible that HPA responses to activity wheel running after olfactory bulbectomy could explain the observed effects on

NPY gene expression. Moreover, an upregulation of NPY mRNA levels could be an adaptive response that serves to attenuate noradrenergic activation within LC neurons. We are unaware of any studies that have previously examined the effects of chronic exercise on brain NPY mRNA levels, and additional research is needed to determine the effects of various exercise stimuli on gene expression in LC neurons.

We observed no differences in expression of the gene that encodes TH after olfactory bulbectomy, activity wheel running, or imipramine treatment. The lack of effect of olfactory bulbectomy and exercise on TH mRNA levels in the LC is consistent with prior research. Holmes & Crawley (1996) observed a transient increase in TH mRNA levels in LC neurons after olfactory bulbectomy, but effects were no longer apparent 14 days after surgery. Similarly, previous studies have found that TH mRNA levels are unchanged after chronic activity wheel running (Soares et al., 1999) and treadmill training (O'Neal et al., 2000). We also failed to observe an effect of imipramine on TH gene expression. Although chronic imipramine treatment has been found to decrease TH mRNA levels (Brady, Whitfield, Fox, Gold, & Herkenham, 1991), effects are most pronounced after 8 weeks of treatment. Thus, the 21-day treatment protocol used in the present study might have been insufficient to induce adaptations in gene expression for TH. In addition, it is difficult to predict the response of TH mRNA, as research examining the effects of chronic administration of antidepressants on TH gene expression within the LC has produced disparate results. Whereas a decrease in TH mRNA level has been found after chronic imipramine administration (Brady et al., 1991), treatment with the selective serotonin reuptake inhibitor, fluoxetine, and the monoamine oxidase inhibitor, phenelzine, have been shown to increase TH mRNA in the LC (Brady, Gold, Herkenha, Lynn, & Whitfield, 1992). Schultzberg, Austin, Crawley, & Paul (1991) observed no effect of chronic treatment with the tricyclic antidepressant, desipramine, on TH mRNA levels in the LC, but concurrent treatment with reserpine prevented a

reserpine-induced elevation in TH mRNA. Given the varied response of TH mRNA to antidepressant treatments and the lack of change with chronic exercise, the effects of pharmacological interventions and physical activity do not appear to be dependent on TH modulation at the level of gene expression within the LC.

Though this study was designed to compare exercise and antidepressant administration in an animal model of depression, we acknowledge the limitations of the experimental manipulations for generalizability to human populations. First, the olfactory bulbectomy model of depression utilized in the present study has inherent disadvantages, namely the anosmic component of the model and the etiological inconsistencies with some of the presumed precursors of depressive disorders in humans (e.g., separation loss or chronic stress). However, chronic but not acute administration of antidepressants is effective in reversing features of the model, and neurobiological consequences of bulbectomy are consistent with the pathology of depression (Kelly et al., 1997).

Second, we selected imipramine, a norepinephrine and serotonin reuptake inhibitor, for the pharmacological intervention. Numerous types of antidepressants have proved efficacious in treating depressive disorders, but not all antidepressant medications are effective for all individuals (Anonymous, 2000). Thus, it is possible that antidepressants with diverse mechanisms of action could produce different results than were obtained in the present study.

Lastly, the present study examined basal gene expression for modulatory components of noradrenergic function in LC neurons. However, it is not known whether the observed adaptations in neuropeptide gene expression resulted in enhanced post-translational activity, as neuropeptide levels were not measured. It is also likely that noradrenergic activity after antidepressant administration and exercise training is altered by mechanisms not examined in the present study (e.g., pre- and post-synaptic receptors, transporters, and 2nd messenger systems). Additionally, though the 21-day protocol was selected to simulate the period required for significant reductions in depressive symptoms

after antidepressant treatment, it is possible that the point of measurement was either premature or too late for detection of differences in mRNA levels. Though few differences were observed in basal levels of mRNA, it is also possible that a response in gene expression might have been found after administration of an acute stressor as observed in previous research (Soares et al., 1999).

To date, few studies have examined the neurobiological effects of exercise at the level of gene expression. The results of the present study suggest that previously observed noradrenergic adaptations to chronic exercise are not dependent upon basal regulation of genes that encode TH, GAL, or NPY in LC neurons. Hence, the hypothesis that chronic exercise and pharmacological treatment would influence noradrenergic function via similar mechanisms of gene expression was not supported by results of the present study. The finding of increased GAL mRNA levels in the LC after imipramine treatment and elevated NPY mRNA levels after activity wheel running in bulbectomized animals are consistent with a modulatory role of these neuropeptides in regulating noradrenergic transmission via feedback inhibition of LC neurons.

Because transcriptional changes that mediate the effects of exercise and/or antidepressant interventions could occur in various brain regions receiving noradrenergic projections, additional studies are needed to examine levels of mRNA in regions associated with the regulation of mood and emotion.   Moreover, gene expression may be dependent upon the nature, severity, and/or duration of the stressor, and further investigation is needed to determine the effects of various exercise protocols and antidepressant treatments on gene expression.

References

Anonymous. (2000). Practice guidelines for the treatment of patients with major depressive disorder (revision). The American Journal of Psychiatry, 157(4), 1-45.

Aston-Jones, G., Rajkowski, J., & Cohen, J. (1999). Role of locus coeruleus in attention and behavioral flexibility. Biological Psychiatry, 46, 1309-1320.

Austin, M.C., Cottingham, S.L., Paul, S.M., & Crawley, J.N. (1990). Tyrosine hydroxylase and galanin mRNA levels in the locus coeruleus neurons are increased following reserpine administration. Synapse, 6, 351-357.

Baldwin, D.S. (1995). Psychotropic drugs and sexual dysfunction. International Review of Psychiatry, 7(2), 261-274.

Brady, L.S., Gold, P.W., Herkenham, M., Lynn, A.B., & Whitfield, H.J. (1992). The antidepressant s fluoxetine, idazoxan and phenelzine alter corticotropin-releasing hormone and tyrosine hydroxylase mRNA levels in rat brain: therapeutic implications. Brain Research, 572, 117-125.

Brady, L.S., Whitfield, H.J., Fox, R.J., Gold, P.W., & Herkenham, M. (1991). Long-term antidepressant administration alters corticotropin-releasing hormone, tyrosine hydroxylase, and mineralocorticoid receptor gene expression in rat brain. Journal of Clinical Investigation, 87, 831-837.

Cairncross, K.D., Schofield, S.P.M., & Bassett, J.R. (1975). Endogenous brain norepinephrine levels following bilateral olfactory bulbectomy bulb ablation. Pharmacology Biochemistry and Behavior, 3, 425-427.

Calcagnetti, D.J., Quatrella, L.A., & Schechter, M.D. (1996). Olfactory bulbectomy disrupts the expression of cocaine-induced conditioned place preference. Physiology & Behavior, 59(4-5), 597-604.

Charney, D.S. (1998). Monoamine dysfunction and the pathophysiology and treatment of depression. Journal of Clinical Psychiatry, 59(Suppl. 14), 11-14.

Dahlstrom, A. & Fuxe, K. (1964). Evidence for the existence of monoamine-containing neurons in the central nervous system. Acta Physiologica Scandinavica, 62(Suppl. 232) 1-55.

Dewsbury, D.A. (1967). A quantitative description of the behavior of rats during copulation. Behaviour, 29, 154-178.

Dishman, R.K., Renner, K.J., Youngstedt, S.D., Reigle, T.G., Bunnell, B.N., Burke, K.A., Yoo, H.S., Mougey, E.H., & Meyerhoff, J.L. (1997). Activity wheel running reduces escape latency and alters brain monomine levels after footshock. Brain Research Bulletin, 42, 399-406.

Dishman, R.K., Bunnell, B.N., Youngstedt, S.D., Yoo, H.S., Mougey, E.H., & Meyerhoff, J.L. (1998). Activity wheel running blunts increased plasma adrenocorticotrophin (ACTH) after footshock and cage-switch stress. Physiology & Behavior, 63(5), 911-917.

Dunn, A.L. & Dishman, R.K. (1991). Exercise and the neurobiology of depression, Exercise and Sport Sciences Reviews, 19. 41-98.

Dunn, A.L, Reigle, T.G., Youngstedt, S.D., Armstrong, R.B., & Dishman, R.K. (1996). Brain norepinephrine and metabolites after treadmill training and wheel running in rats. Medicine and Science in Sports and Exercise, 28, 204-209.

Ferguson, G.A. (1966). Tests of significance: other statistics. In, Statistical Analysis in Psychology and Education (pp.176-181). New York: McGraw-Hill.

Finta, E.P., Regenold, J.T., & Illes, P. Depression by neuropeptide Y of noradrenergic inhibitory postsynaptic potentials of locus coeruleus neurons. Naunyn-Schmiedeberg's Archives of Pharmacology, 346, 472-474.

Foote, S.L., Bloom, F.E., & Aston-Jones, G. (1983). Nucleus locus ceruleus: New evidence of anatomical and physiological specificity. Physiological Reviews, 63, 844-903.

Grecksch, G., Zhou, D., Franke, C., Schroder, U., Sabel, B., Becker, A., & Huether, G. (1997). Influence of olfactory bulbectomy and subsequent imipramine treatment of 5-hydroxytryptaminergic presynapses in the rat frontal cortex: behavioural correlates. British Journal of Pharmacology, 122(8), 1725-1731.

Grima, B., Lamouroux, A., Blanot, F., Faucon-Biguet, N., & Mallet, J. (1985). Complete coding sequence of rat tyrosine hydroxylase mRNA. Proceedings of the National Academy of Sciences, 82, 617-621.

Hashimoto, H., Onishi, H., Koide, S., Kai, T., & Yamagami, S. (1996). Plasma neuropeptide Y in patients with major depressive disorder. Neuroscience Letters, 216, 57-60.

Heyes, M.P., Garnett, E.S., & Coates, G. (1988). Nigrostriatal dopaminergic activity is increased during exhaustive exercise stress in rats. Life Sciences, 42, 1537-1542.

Holets, V.R., Hokfelt, T., Rokaeus, A., Terenius, L., & Goldstein, M. (1988). Locus coeruleus neurons in the rat containing neuropeptide Y, tyrosine hydroxylase or galanin and their efferent projections to the spinal cord, cerebral cortex and hypothalamus. Neuroscience, 24, 893-906.

Holmes, P.V., Blanchard, D.C., Blanchard, R.J., Brady, L.S., Crawley, J.N. (1995). Chronic social stress increases levels of preprogalanin mRNA in the rat locus coeruleus. Pharmacology Biochemistry and Behavior, 50, 655-660.

Holmes, P.V. & Crawley, J.N. (1995). Coexisting neurotransmitters in central noradrenergic neurons. In F.E. Bloom & D.J. Kupfer (Eds.), Psychopharmacology: The Fourth Generation of Progress (pp. 347-353. New York: Raven Press, Ltd.

Holmes, P.V. & Crawley, J.N. (1996). Olfactory bulbectomy increases prepro-galanin mRNA levels in the rat locus coeruleus. Molecular Brain Research, 36, 184-188.

Illes, P. & Regenold, J.T. (1990). Interaction between neuropeptide Y and noradrenaline on central catecholamine neurons. Nature (London), 344, 62-63.

Kelly, J.P., Wrynn, A.S., & Leonard, B.E. (1997). The olfactory bulbectomized rat as a model of depression: an update. Pharmacology & Therapeutics, 74(3), 299-316.

Larhammer, D., Ericsson, A., & Persson, H. (1987). Structure and expression of the rat neuropeptide Y gene. Proceedings of the National Academy of Sciences, 84, 2068-2072.

Leonard, B.E. (1997). Noradrenaline in basic models of depression. European Neuropsychopharmacology, 7(Suppl. 1), S11-S16.

Linner, L., Arboreliu, L., Nomikos, G.G., Bertilsson, L., & Svensson, T.H. (1999). Locus coeruleus neuronal activity and noradrenaline availability in the frontal cortex of rats chronically treated with imipramine: Effect of $"_2$-adrenoceptor blockade. Biological Psychiatry, 46, 766-774.

Lumia, A.R., Teicher, M.H., Salchli, F., Ayers, E., & Possidente, B. (1992). Olfactory bulbectomy as a model for agitated hyposerotonergic depression. Brain Research, 587, 181-185.

Makino, S., Baker, R.A., Smith, M.A., & Gold, P.W. (2000). Differential regulation of neuropeptide Y mRNA expression in the arcuate nucleus and locus coeruleus by stress and antidepressants. Journal of Neuroendocrinology, 12, 387-395.

Marcilhac, A., Maurel, D., Anglade, G., Ixart, G., Mekaouche, M., Hery, F, & Siaud, P. (1997). Effects of bilateral olfactory bulbectomy on circadian rhythms of ACTH, corticosterone, motor activity and body temperature in male rats. Archives of Physiology and Biochemistry, 105(6), 552-559.

O'Neal, H.A., Dunn, A.L., & Martinsen, E.W. (2000). Depression and exercise. International Journal of Sport Psychology, 31, 110-135.

O'Neal, H.A., Van Hoomissen, J.D., Holmes, P.V., & Dishman, R.K. (2000). Prepro-galanin mRNA levels are increased in rat locus coeruleus after treadmill exercise training. Manuscript submitted for publication.

Pieribone, V.A., Xu, Z.Q., Zhang, X., Grillner, S., Bartfai, T., & Hokfelt, T. (1995). Galanin induces a hyperpolarization of norepinephrine-containing locus coeruleus neurons in the brainstem slice. Neuroscience, 64, 861-874.

Rasmussen, K., Morilak, D.A., & Jacobs, B.L. (1986). Single unit activity of locus coeruleus neurons in the freely moving cat: I. during naturalistic behaviors and in response to simple and complex stimuli. Brain Research, 371, 324-334.

Redmond, A.M., Kelly, J.P., & Leonard, B.E. (1997). Behavioural and neurochemical effects of dizocilpine in the olfactory bulbectomized rat model of depression. Pharmacology Biochemistry and Behavior, 58(2), 355-359.

Rokaeus, A. & Brownstein, M.J. (1986). Construction of a porcine adrenal medullary cDNA library and nucleotide sequence analysis of two clones encoding a galanin precursor. Proceedings of the National Academy of Sciences, 83, 6287-6291.

Schultzberg, M., Austin, M.C., Crawley, J.N., & Paul, S.M. (1991). Repeated administration of desmethylimipramine blocks the reserpine-induced increase in tyrosine hydroxylase mRNA in locus coeruleus neurons of the rat. Molecular Brain Research, 10, 307-314.

Seutin, V., Verbanck, P., Massote, P., & Dresse, A. (1989). Galanin decreases the activity of locus coeruleus neurons in vitro. European Journal of Pharmacology, 164, 373-376.

Smialowska, M. & Legutko, B. (1991). Influence of imipramine on neuropeptide Y immunoreactivity in the rat brain. Neuroscience, 41(2/3), 767-771.

Soares, J., Holmes, P.V., Renner, K.J., Edwards, G.L., Bunnell, B.N., & Dishman, R.K. (1999). Brain noradrenergic responses to footshock after chronic activity-wheel running. Behavioral Neuroscience, 113, 558-566.

Song, C., Earley, B., & Leonard, B.E. (1996). The effects of central administration of neuropeptide Y on behavior, neurotransmitter, and immune functions in the olfactory bulbectomized rat model of depression. Brain, Behavior, and Immunity, 10, 1-16.

Westrin, A., Ekman, R., Traskman-Bendz, L. (1999). Alterations of corticotropin releasing hormone (CRH) and neuropeptide Y (NPY) plasma levels in mood disorder patients with a recent suicide attempt. European Neuropsychopharmacology, 9, 205-211.

White-Welkley, J.E., Bunnell, B.N., Mougey, E.H., Meyerhoff, J.L., & Dishman, R.K. (1995). Treadmill exercise training and estradiol differentially modulate hypothalamic-pituitary-adrenal cortical responses to acute running and immobilization. Physiology & Behavior, 57, 533-540.

Xu, Z.D., Shi, T.S., & Hokfelt, T. (1998). Galanin/GMAP- and NPY-like immunoreactivities in locus coeruleus and noradrenergic nerve terminals in the hippocampal formation and cortex with notes on the galanin-R1 and –R2 receptors. Journal of Comparative Neurology, 392, 227-251.

Yoo, H.S., Tackett, R.L., Crabbe, J.B., Bunnell, B.N., & Dishman, R.K. (2000). Antidepressant-like effects of physical activity vs. imipramine: Neonatal clomipramine model. Psychobiology, in press.

Figure 1. Sucrose preference score expressed as percent of total fluid intake in activity wheel and sedentary animals after imipramine or saline administration by group. A significant main effect in sucrose consumption for group is depicted. N=8-11 animals per group.
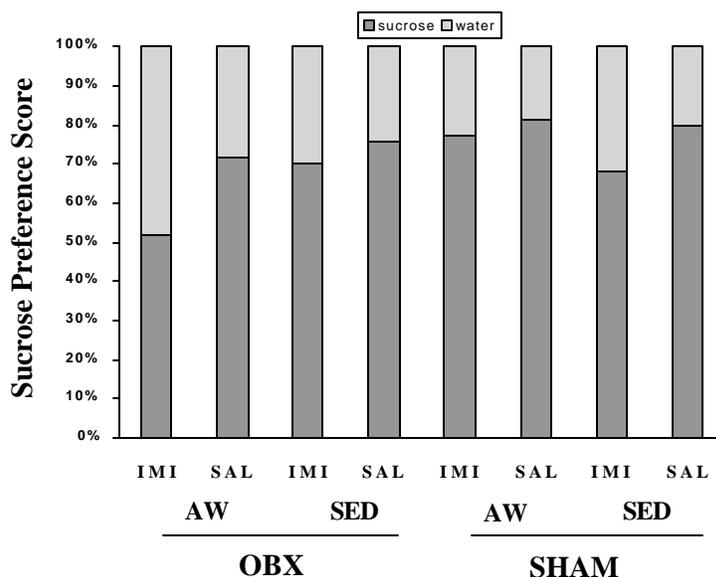
Figure 2. Copulation rates among male rats according to group assignment. Within the olfactory bulbectomy group, imipramine-treated animals exhibited lower copulation rates compared to saline-treated animals (p=.005). Sedentary, saline-treated animals exhibited lower copulation rates compared to activity wheel, saline-treated animals after olfactory bulbectomy (p=.045). Activity wheel runners receiving saline treatment after olfactory bulbectomy did not differ from sham animals. N=8-11 animals per group.
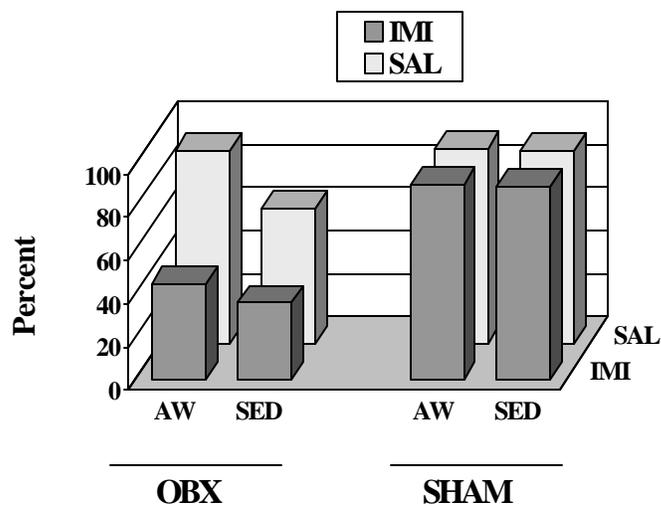
Figure 3. Ejaculation latencies in activity wheel and sedentary animals after imipramine or saline administration by group. Within the bulbectomy group, activity wheel runners exhibited significant reductions in ejaculation latency (p=.04). The number of animals per group is given in parentheses. Values are means ± SE.
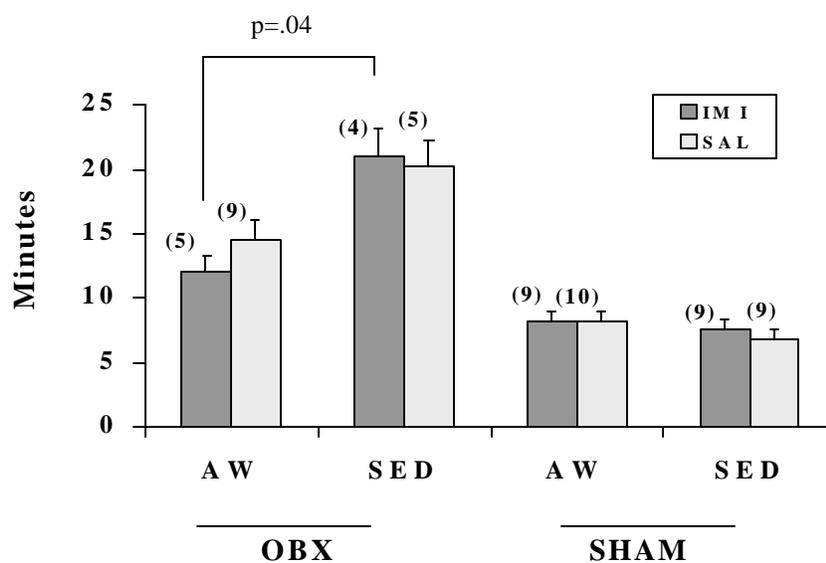
Figure 4. Post-ejaculatory intervals in activity wheel and sedentary animals after imipramine or saline administration by group. A significant main effect for group is depicted. Activity wheel running after olfactory bulbectomy was associated with longer intervals relative to sham animals (p=.04). The number of animals per group is given in parentheses. Values are means ± SE.
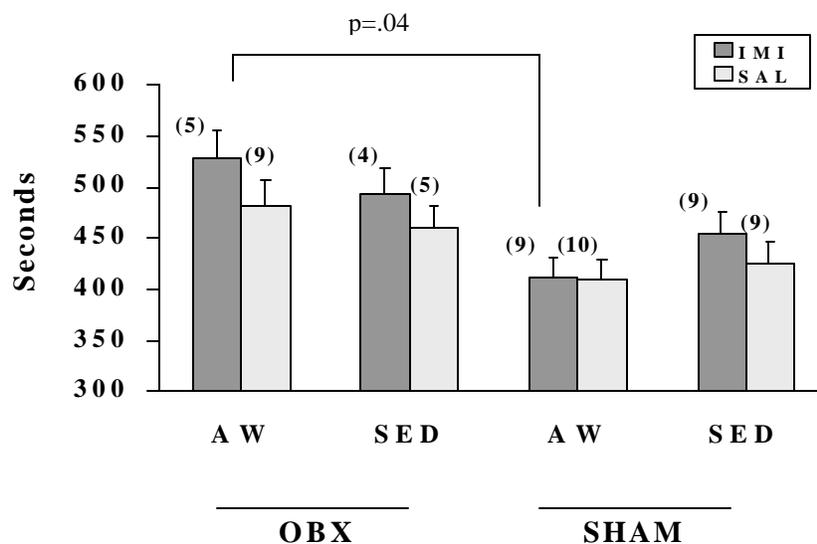
Figure 5.   Ejaculation frequency in activity wheel and sedentary animals after imipramine or saline administration by group. A significant main effect for group is depicted. Within the olfactory bulbectomy group, imipramine-treated animals exhibited significant decreases in ejaculation frequency (p=.02). N=8-11 animals per group. Values are means ± SE.
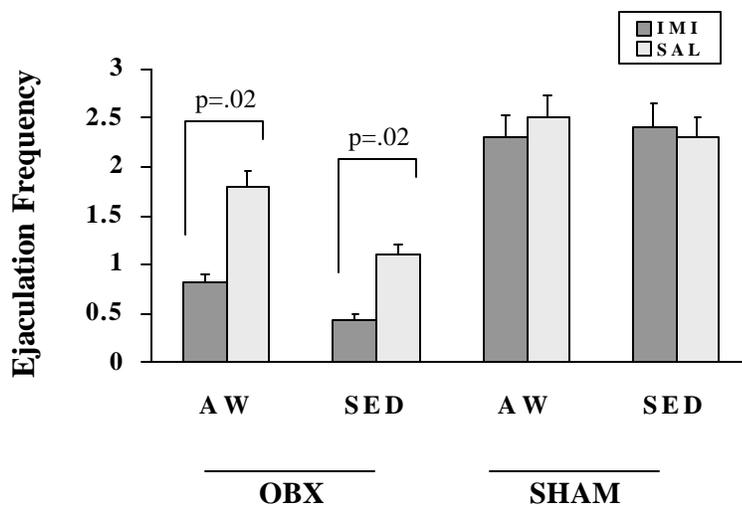
Figure 6. Levels of prepro-galanin mRNA in the locus coeruleus in activity wheel and sedentary animals after imipramine or saline administration by group. A significant main effect for treatment (imipramine vs saline) is depicted. N=8-10 animals per group. Values are means ± SE. OGS= Optical Gray Scale
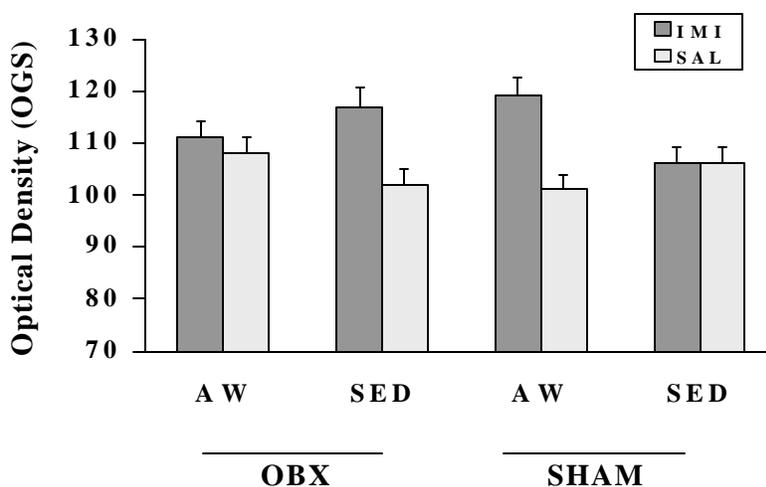
Figure 7. Levels of prepro-neuropeptide Y mRNA in the locus coeruleus in activity wheel and sedentary animals after imipramine or saline administration by group A significant group (olfactory bulbectomy vs sham) by condition (activity wheel vs sedentary) interaction is depicted (p=.04). N=7-10 animals per group. Values are means ± SE. OGS= Optical Gray Scale
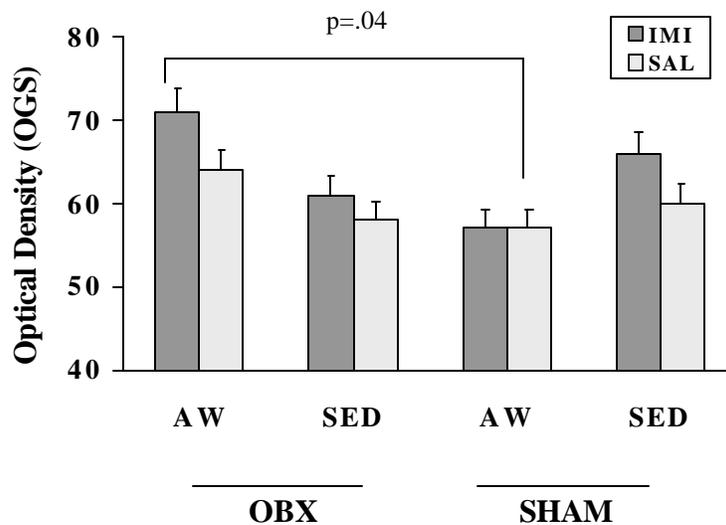
Figure 8. Levels of tyrosine hydroxylase mRNA in the locus coeruleus in activity wheel and sedentary animals after imipramine or saline administration by group. There were no significant differences among comparisons. N=8-11 animals per group. Values are means ± SE. OGS= Optical Gray Scale
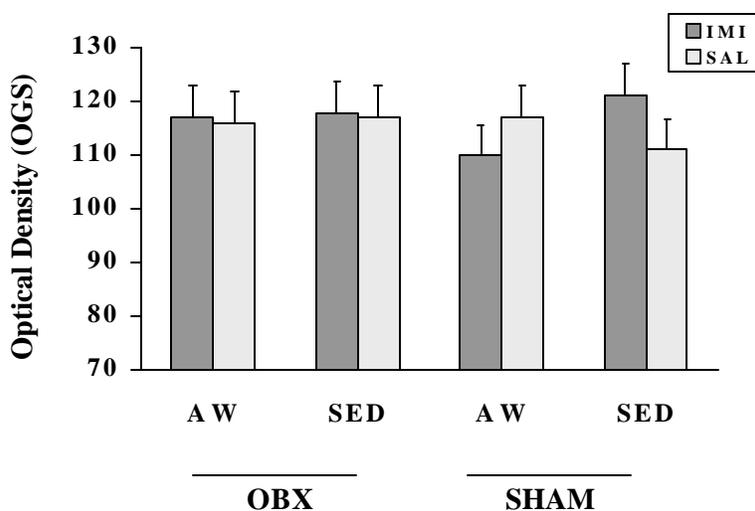
Figure 9. Representative autoradiographs of rat brain sections hybridized for prepro-galanin mRNA in the locus coeruleus (LC). Rats were activity wheel runners in the sham surgery group after treatment with imipramine (A) or saline (B).
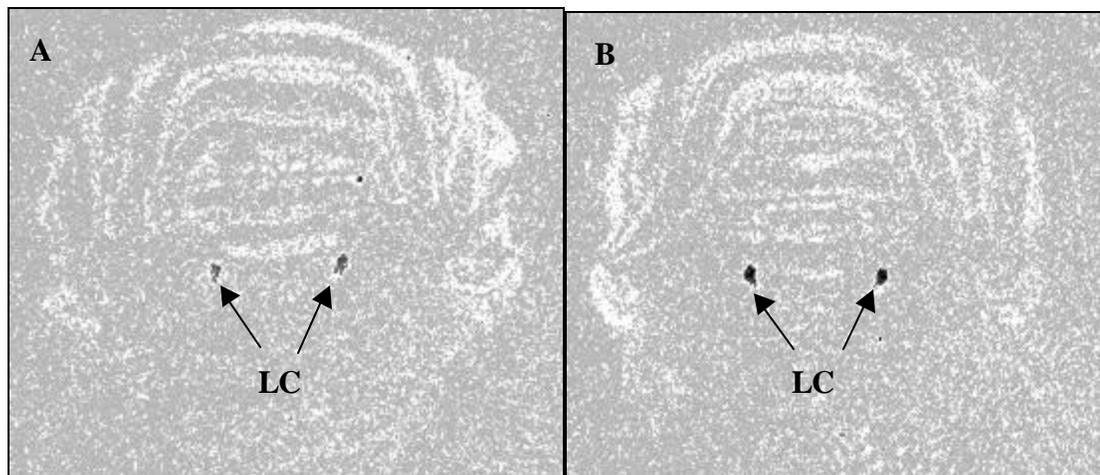
Figure 10. Representative autoradiographs of rat brain sections hybridized for prepro-neuropeptide Y mRNA in the locus coeruleus (LC). Rats were activity wheel runners after sham surgery (A) or olfactory bulbectomy (B).
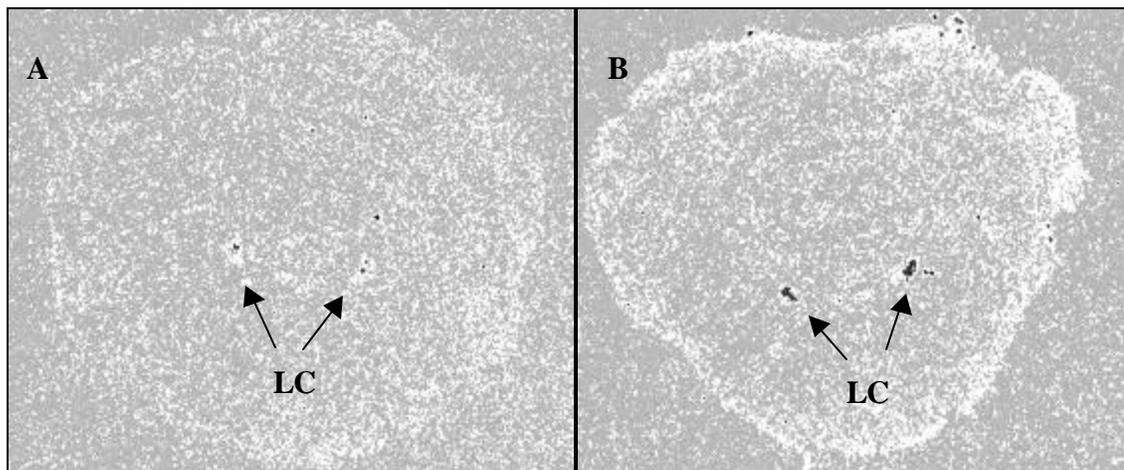
CONCLUSIONS

Research conducted during the past three decades suggests that exercise can be effective in reducing symptoms of depression for many individuals. Given the tremendous burden that depression poses to society, if exercise is indeed an effective antidepressant treatment, the potential impact on public health would be substantial. However, because this area of research is characterized by a lack of scientific rigor, exercise has yet to be recognized by prominent mental health organizations as a viable therapeutic intervention for depression.

The preponderance of evidence from population-based studies supports an inverse association between physical activity and symptoms of depression. However, methodological variations limit generalizability of results. In order to support a causal relationship between exercise and reductions in depression, additional population-based studies are needed that (1) include representative samples, (2) study high-risk populations, (3) use valid measures of both physical activity and depression, (4) control for additional risk factors, and (5) follow participants for a time period that permits observation of the progression of depressive disorders including onset, remission, and recurrence.

A quantitative synthesis of data from controlled intervention studies yielded a moderate overall effect (d=-0.57), suggesting that individuals who participate in exercise programs exhibit a ½ SD reduction in depressive symptoms relative to individuals in comparison groups. Moderator analyses revealed larger effects for studies that examined individuals with elevated symptoms of depression at baseline and that used clinical ratings to assess depression.

Based on our findings from both the qualitative review of literature and meta-analytic procedures, we offer the following recommendations for future research: (1) Studies should include participants meeting standard diagnostic criteria for depressive disorders in order to obtain meaningful reductions in depressive symptoms and establish exercise as an efficacious treatment for clinical depression. (2) Valid assessments of depression should be selected based on the psychometric properties and appropriateness for the population of interest. When possible, studies should include clinical ratings to verify data from self-report measures. (3) Comparison groups should, at minimum, control for placebo effects and spontaneous remission of depressive symptoms, and additional studies that include clinically relevant control conditions are needed to substantiate meaningful reductions in depression. (4) Studies that manipulate components of the exercise intervention (e.g., mode, intensity, duration, and frequency) are needed to identify the stimulus required to produce antidepressant effects. (5) Further examination of neurobiological adaptations to exercise is required to demonstrate the biological plausibility of exercise as an antidepressant intervention.

In order to investigate biologically plausible mechanisms for the antidepressant effects of exercise and expand upon previous research, we examined gene expression after chronic exercise in noradrenergic cell bodies within the locus coeruleus complex. The purpose of the first study was to determine the effect of treadmill exercise training on basal levels of the gene that encodes TH and GAL in the LC. Because chronic treadmill exercise results in increased brain NE concentration in the pons region containing the LC complex (Dunn, Reigle, Youngstedt, Armstrong, & Dishman, 1996), we hypothesized that an upregulation in gene expression for TH (via synthesis) or GAL (via feedback inhibition) could enhance availability of brain NE. Although no differences were observed in TH mRNA level after 6 weeks of treadmill training, GAL mRNA levels were higher in exercise-trained compared to sedentary animals. These results suggest that gene expression for GAL is responsive to chronic exercise stress. If the observed

increase in GAL mRNA results in enhanced post-translational activity, GAL could act on LC neurons to inhibit firing and preserve central NE levels. Because prominent biological theories of depression involve a deficiency in NE, exercise-induced increases in GAL within the LC may serve an adaptive role in the homeostatic regulation of noradrenergic activity.

Although exercise induces adaptations in noradrenergic function, the efficacy of exercise training in reversing disturbances in noradrenergic transmission has not been widely studied. However, prior research by Yoo, Tackett, Crabbe, Bunnell & Dishman (2000) using the neonatal clomipramine animal model of depression suggests that chronic exercise by activity wheel running produces beneficial noradrenergic adaptations including elevated NE levels and downregulation of β-adrenoreceptors in frontal cortex comparable to antidepressant pharmacotherapy. To continue this line of research, an animal depression model was used to examine mechanisms at the level of gene expression that might induce adaptations in noradrenergic function.

Therefore, the second study was designed to investigate the effects of chronic activity wheel running and imipramine administration on gene expression for noradrenergic modulatory proteins in neurons of the LC after olfactory bulbectomy. Behavioral assessments including sucrose preference and male copulatory activity were conducted to measure exhibition of motivated behaviors. Olfactory bulbectomy produced a decrease in consumption of sucrose solution, but neither activity wheel running nor imipramine treatment significantly altered this effect. In addition, olfactory bulbectomy produced deficits in male copulatory behavior including reduced copulation rates, prolonged ejaculation latencies, and increased post-ejaculatory intervals. However, consistent with prior research, activity wheel running improved copulatory performance in experimental animals (Yoo et al., 2000). Activity wheel running after olfactory bulbectomy resulted in improved copulation rates, reduced ejaculation latencies, and increased ejaculation frequencies. In addition, activity wheel running attenuated the

decrements in sexual activity associated with imipramine administration. These findings are clinically meaningful, as both symptoms of depression and side effects of commonly prescribed antidepressant medications include disturbances in sexual performance.

In contrast to previous studies, we failed to observe an effect of olfactory bulbectomy (Holmes & Crawley, 1996) or exercise (O'Neal, Van Hoomissen, Holmes, & Dishman., 2000) on GAL mRNA level. However, levels of GAL mRNA were increased by imipramine treatment. In addition, activity wheel running after olfactory bulbectomy was associated with elevations in NPY mRNA. There were no differences in gene expression for TH. Thus, adaptations in noradrenergic function after imipramine treatment and activity wheel running do not appear to be regulated via similar basal mechanisms at the level of gene expression within the LC, though both imipramine treatment and activity wheel running induced alterations that might influence noradrenergic activity.

To date, few studies have examined the neurobiological effect of exercise at the level of gene expression. These results suggest that previously observed adaptations to chronic activity wheel exercise are not dependent upon basal regulation of gene expression for TH, GAL, or NPY in LC neurons. Hence, the hypothesis that chronic exercise and pharmacological treatment would influence noradrenergic function via similar mechanisms was not supported. The finding of increased GAL mRNA level in the LC after imipramine treatment and elevated NPY mRNA level after activity wheel running in bulbectomized animals could indicate a modulatory role of these neuropeptides in regulating noradrenergic transmission via feedback inhibition. However, it is not known whether the observed adaptations in neuropeptide gene expression resulted in enhanced post-translational activity, as neuropeptide levels were not measured.

Though animal models possess inherent limitations for the study of human mental illness, such models are important for identifying mechanisms for the etiology and

treatment of depressive disorders. Therefore, additional studies conducted within depression models are needed to derive clinically relevant conclusions regarding the influence of exercise on malfunctioning brain systems. Because adaptations may be dependent upon the nature, severity, and/or duration of the stressor, research is also needed to determine the effects of various exercise protocols (e.g., treadmill vs. activity wheel) and antidepressant treatments (e.g., SSRI vs TCA) on gene expression. Furthermore, noradrenergic transmission after antidepressant administration and exercise training is likely altered by mechanisms not examined in the present study (e.g., pre- and post-synaptic receptors, transporters, and 2nd messenger systems), and such mechanisms warrant further investigation. Adaptations that mediate the effects of exercise and/or pharmacotherapy could also involve other monoaminergic systems (i.e., serotonin and dopamine) and occur in various brain regions associated with the regulation of mood and behavior (i.e., ventral tegmental area, nucleus accumbens, dorsal raphe, amygdala and hypothalamic nuclei). Thus, additional studies are needed to elucidate the effect of physical activity on neurobiological systems associated with the pathology of depression to establish the biological plausibility of exercise as an antidepressant intervention.

References

Dunn, A.L., Reigle, T.G., Youngstedt, S.D., Armstrong, R.B., & Dishman, R.K. (1996). Brain norepinephrine and metabolites after treadmill training and wheel running in rats. Medicine and Science in Sports and Exercise, 28, 204-209.

Holmes, P.V., & Crawley, J.N. (1996). Olfactory bulbectomy increases prepro-galanin mRNA levels in the rat locus coeruleus. Molecular Brain Research, 36, 184-188.

O'Neal, H.A., Van Hoomissen, J.D., Holmes, P.V., & Dishman, R.K. (2000). Prepro-galanin mRNA levels are increased in rat locus coeruleus after treadmill exercise training. Manuscript submitted for publication.

Yoo, H.S., Tackett, R.L., Crabbe, J.B., Bunnell, B.N., & Dishman, R.K. (2000). Antidepressant-like effects of physical activity vs. imipramine: Neonatal clomipramine model. Psychobiology, in press.