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DYSTHYMIA: BEHAVIORAL, NEUROENDOCRINE AND IMMUNE FACTORS

by

Jenna Griffiths

Dissertation presented to

the Faculty of Graduate Studies and Research

in partial fulfillment of the requirements for the degree of

Doctor of Philosophy

Department of Psychology

Carleton University

Ottawa, Ontario

November 23, 1998

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in partial fulfillment of the requirements
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Abstract

In this series of experiments we explore several behavioral and biological features associated with dysthymic disorder, a subtype of depressive illness characterized by chronic, low-grade depression. Although symptoms of dysthymia are less severe, albeit more prolonged, than those associated with major depressive disorder (MDD), dysthymic patients exhibited behavioral dysfunction in many ways similar to that of MDD patients. Specifically, both dysthymic and MDD patients exhibited heightened perception of stressors and diminished positive events relative to controls, while relying on emotion-focused coping strategies at the expense of problem-focused coping. Moreover, like MDD, dysthymic symptoms were successfully alleviated following treatment with a selective serotonin reuptake inhibitor (SSRI). While pharmacotherapy assuaged stressor perception and improved coping strategies, uplift perception was not influenced to the same degree, supporting the notion that despite symptom alleviation, residual functional impairments persisted.

In addition to behavioral differences relative to controls, the depressive patients exhibited elevated circulating lymphocyte subsets, particularly natural killer (NK) cells. While pharmacotherapy resulted in normalization of circulating NK cells in both MDD and dysthymic patients, a more sustained treatment interval was required to achieve this effect among the dysthymic patients. Interestingly, regardless of depressive subtype, significant gender differences were observed in the levels of circulating NK cells.

Although stressor perception was heightened among dysthymic and MDD patients relative to controls, the depressive patients could not be distinguished from controls with respect to their immunological responses to laboratory stressors, such as a cognitive challenge and a clinical interview. However, circulating lymphocytes were exquisitely sensitive to stressors of graded severity. Specifically, among nondepressed students exposure to the cognitive challenge and clinical interview resulted in a modest increase of circulating NK cells; however, the more intense stress associated with the anticipation of a

final academic examination was associated with a significantly larger NK response, as well as increases of several other lymphocyte subsets, cortisol, and ACTH.

In addition to their core diagnostic symptoms, dysthymia and MDD can be classified into typical and atypical subgroups based on various neurovegetative and behavioral features. Patients classified as either typical or atypical dysthymia or MDD were compared with respect to stressor perception, coping strategies and quality of life. Although, these behaviors did not differentiate the typical from the atypical patients, immunological, cytokine and neuroendocrine assessments revealed that these subgroups were biologically distinct from one another. In particular, the atypical MDD patients displayed elevated ACTH, but reduced cortisol, a profile reminiscent of that seen in other illnesses characterized by atypical symptoms. As well, it appeared that the altered NK levels were most apparent in male, typical MDD patients, suggesting a relationship with neurovegetative features of the illness. Of particular interest was the finding that heightened production of mitogen-stimulated interleukin (IL)-1 β occurred specifically among the dysthymic patients, irrespective of whether they exhibited typical or atypical features. This finding raised the possibility that the chronicity of dysthymia played a critical role in the functional status of this cytokine. In contrast to IL-1, production of IL-2 was significantly reduced in the two dysthymic groups and in the atypical major depressive patients relative to nondepressed controls. These data were taken to suggest that depressive illness may be associated with immune activation. A provisional model is offered to account for the mechanisms underlying dysthymia (as well as recurrent depression) based on cytokine and neuroendocrine alterations associated with stress and depressive affect.

Acknowledgments

I would like to express my sincere gratitude to Dr. Hymie Anisman, for providing guidance, inspiration, and tireless encouragement. It has been a privilege to work with you. I would also like to express my appreciation to Dr. A.V. Ravindran for supplying me the means and invaluable opportunities for such a rich learning experience, to Dr. Zul Merali for providing his expertise relating to various neuroendocrine processes, and to Dr. Jerzy Kulczycki for technical analyses and training.

Most importantly, I am indebted to my family for their enduring love and moral support. I like to think that I have combined a fraction of my mother's ability with people as a social worker with my father's inquisitiveness as a gifted scientist. I feel very blessed to have you as my parents. Thank-you Lizz & Ken, Chris & Maria for always being there for me. Finally, Matthew, Olivia, Emily and Andrew... you are my beacons of joy.

This work was supported in part by a grant from the Medical Research Council of Canada and the Pharmaceutical Manufacturers Association of Canada (MRC-PMAC) in conjunction with Pfizer Canada Inc.

To my parents

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GLOSSARY

Acute phase proteins: Serum proteins that increase during acute inflammation.

Acute phase response (APR): A nonspecific response by an individual stimulated by IL-1 β , IL-6, IFN and TNF- α . C-reactive protein may show a striking rise within a few hours. Infection, inflammation, tissue injury, and often neoplasm may be associated with APR. The liver produces acute-phase proteins at an accelerated rate, and the endocrine system is affected with elevated gluconeogenesis and impaired thyroid function.

Adrenocorticotrophic hormone (ACTH): Polypeptide hormone secreted by the anterior pituitary; stimulates cells of the adrenal cortex to secrete cortisol.

Antigen: Any foreign material that is specifically bound by a specific antibody or specific lymphocytes.

Complement: Regulatory proteins and receptors found on blood and tissue cells. The complement system is activated by the reaction between antigen and antibody. It consists of a series of serum enzymes, the end result of which is lysis of the target or enhanced phagocytosis by phagocytic cells. Complement proteins are significant, nonspecific mediators of humoral immunity.

Corticotropin releasing hormone (CRH): Hypothalamic hormone that stimulates ACTH secretion by the anterior pituitary.

Cytokines: Immune system proteins which are biological response modifiers. They coordinate antibody and T cell immune system interactions and amplify immune reactivity. Cytokines include monokines (e.g., IL-1, TNF α) synthesized by macrophages, and lymphokines (e.g., interleukins, gamma interferon, lymphotoxin) produced by activated T lymphocytes and natural killer cells.

Enzyme-linked immunosorbent assay (ELISA): An assay in which an enzyme is linked to an antibody and a coloured substrate is used to measure the activity of bound enzyme and, thus, the amount of bound antibody. This method is based on the sandwich or double-layer technique, in which an enzyme rather than a fluorochrome is used as the label. ELISA involves the solid-phase immunoassay technique, which employs the property of various plastics to adsorb (hold molecules to its surface, forming a thin film) monomolecular layers of proteins onto their surface. Although the adsorbed molecules may lose some of their antigenic determinants, enough remain unaltered and can still react with their corresponding antibodies. The presence of these antibodies, bound to antigen adsorbed onto the plastic, may be detected by anti-immunoglobulins labeled with an enzyme. These enzyme-labeled anti-immunoglobulins can be detected by the appearance of a color upon addition of a substrate. The ELISA test is read by incubating the reactants with an appropriate substrate to yield a colored product that is measured in a spectrophotometer.

Epinephrine: A hormone, secreted by the adrenal medulla, that produces physiological effects characteristic of the sympathetic division of the autonomic nervous system.

Fluorescence-Activated Cell-Sorting Analysis (FACSCAN): a tool developed around the use of a fluorescent antibody specific for cell surface antigens. A cell suspension labeled with specific fluorescent antibody is passed through an apparatus which forms a stream of small droplets each containing one cell. These droplets are passed

between a laser beam of ultraviolet light and a detector for picking up emitted fluorescence when a labeled cell is present in the droplet. This emitted signal is passed to an electrode which charges the droplet, leading to its deflection in an electromagnetic field. Thus as all droplets fall through they are collected and counted depending on whether they emit a signal. By sophisticated electronics the fluorescein-stained and unstained cells may be counted, as well as the intensity of fluorescence on each. With this type of apparatus a profile is developed of a pool of lymphocytes consisting of their type, the presence of differentiation markers, the relative amount of marker on each, and size distribution and numbers of each type. One of its many uses is to enumerate lymphocytes which are first reacted with a specific monoclonal antibody, then followed by interaction with fluorescein-labeled anti-Ig.

Glucocorticoid: One of a group of hormones (e.g., cortisol) of the adrenal cortex that are important in protein and carbohydrate metabolism, secreted especially in times of stress.

Interleukins: A group of cytokines synthesized by lymphocytes, monocytes and other cells, which promote growth of T and B cells and other hematopoietic stem cells, and have a variety of other biological functions.

Interleukin (IL)-1: Produced by monocytes and macrophages; it stimulates acute phase protein synthesis and promotes T-cell proliferation; it is also an endogenous fever-producing substance. IL-1 is comprised of two principal polypeptides: IL-1 α and IL-1 β .

IL-2: Secreted by T-helper cells. IL-2 has an autocrine effect, acting on the CD4 cells that produce it, but it mainly affects lymphocytes. IL-2 (formerly known as T-cell growth factor) is a factor required for the growth of some T cells, B cells, and promotes synthesis of antibody by B cells. IL-2 also promotes growth of NK cells, and potentiates their cytolytic action.

IL-6: (formerly known as B cell differentiation factor): produced by vascular endothelial cells, phagocytes, fibroblasts, activated T lymphocytes, and neoplasms. IL-6 is secreted in response to IL-1 or TNF. It mainly acts on B cells and hepatocytes. Its main function is to cause B lymphocytes to differentiate into cells that synthesize antibodies. IL-6 also induces hepatocytes to form acute-phase proteins including fibrinogen.

IL-2 receptor (IL-2R; also known as CD25, Tac antigen): Structure on the surface of T and B lymphocytes and NK cells which interacts with IL-2 molecules. IL-1, -2, -4, -6 and TNF can induce IL-2R expression.

Interferon-gamma (IFN- γ): Synthesized by activated T-helper cells. Activates killer T cells, NK cells and macrophages.

Lymphocytes: Small cell with virtually no cytoplasm, and bears antigen-specific receptors; found in blood, in all tissue, and in lymphoid organs, such as lymph nodes, spleen, Peyer's patches. Lymphocytes are divided into 2 principal groups: T and B lymphocytes. In addition, NK cells, which are large granular lymphocytes, comprise a small percentage of the lymphocyte population. Lymphocytes are distinguished by the expression of distinctive surface molecules that have precise roles in immune reaction.

Mitogen: A substance, often derived from plants, that causes DNA synthesis and induces blast transformation and division by mitosis. Lectins, representing plant-derived mitogens, are widely used in clinical and experimental immunology to evaluate T and B lymphocyte function in vitro. Phytohemagglutinin (PHA) and concanavalin A (Con A) are principally T cell mitogens. Staphylococcal protein A is the mitogen used to induce B lymphocyte

transformation. Pokeweed mitogen is a B and T cell mitogen.

Natural killer (NK) cells: Cells that attack and destroy tumor cells and certain virus-infected cells. NK cells induce lysis through the action of antibody. Immunologic memory is not involved, in that previous contact with the antigen is not necessary.

Norepinephrine (NE): A neurotransmitter found in the brain and in the terminal buttons of postganglionic fibers of the sympathetic division of the autonomic nervous system.

Plasma: Transparent yellow fluid that constitutes 50-55% of blood volume. It is 92% fluid, 7% protein, and 1% inorganic salts, sugars, lipids, hormones and gases.

Radioimmunoassay (RIA): A technique to assay either antigen or antibody which is based on radiolabeled antigen competitively inhibiting the binding of antigen, which is not labeled, to specific antibodies. Minute quantities of enzymes, hormones, or other immunogens can be assayed by RIA. Because of problems concerning radioisotope regulation and disposal, enzyme immunoassays are largely replacing RIAs.

Serum: Plasma from which fibrinogen and clotting factors have been removed.

Solid-phase immunoassay can be used to detect the presence of antibodies to the antigen that coats the plastic. Since the plastic wells are usually coated with relatively large amounts of antigen, the higher the concentration of antibodies bound with the antigen, the higher the amount of labeled anti-immunoglobulin that can bind to the antibodies. Thus, it is important to use excess labeled anti-ig for saturation. Solid-phase immunoassay can be used to qualitatively/quantitatively determine antigen by mixing antiserum with varying known amounts of antigen before adding the antiserum to the antigen-coated plastic wells. This results in the antibodies binding with antigen coating the plastic. The higher the concentration of soluble antigen that reacts with antibodies prior to addition of the antibody to the wells, the lower the number of antibodies that can bind with the antigen on the plate, and the lower the number of labeled anti-immunoglobulin that can bind to these antibodies. The decrease in the amount of bound label as a function of the concentration of antigen used to cause this decrease can be plotted, and the amount of antigen in an unknown solution can then be determined from the graph by a comparison of the decrease in bound label caused by the unknown solution to the decrease caused by known concentrations of pure antigen.

Tumor Necrosis Factor (TNF-) α : TNF α is a monokine produced by macrophages stimulated with bacterial endotoxin; it participates in inflammation, wound healing and tissue remodelling.

Note:

Benjamini E, Leskowitz S. *Immunology: A Short Course* (2nd Edition). Wiley-Liss: New York, 1991.

Carlson NR. *Physiology of Behavior* (3rd Edition). Allyn and Bacon: Boston, 1986.

Cruse JM, Lewis RR. *Illustrated Dictionary of Immunology*. CRC Press: Boca Raton, 1995.

Vander AJ, Serman JH, Luciano DS. *Human Physiology: The Mechanisms of Body Function* (3rd Edition). McGraw-Hill: New York, 1980.

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1. GENERAL INTRODUCTION

Although dysthymia, a chronic, low-grade form of depression, occurs in a substantial portion of the population, and increases the risk for major depressive disorder, there is limited information available concerning the behavioral concomitants, as well as the physiological mechanisms subserving this illness. While the view had long been accepted that depressive illnesses are subserved by neurochemical and neuroendocrine processes, increasing evidence has pointed to a potential role for immune factors in this respect. However, despite the numerous reports of immune dysregulation in depressive illness, inconsistent findings have been documented. Furthermore, while the position had been taken that depression promoted immunosuppression, it has recently been argued that the compromised immunity may be secondary to an initial immune activation (not unlike an acute phase reaction). According to this view, immune activation may come to promote central neurochemical changes, hence provoking a depressive episode (see Maes, 1995).

The immune activation notwithstanding, the disparate results concerning the immunosuppression associated with depressive illness may be due, in part, to the divergent features of the mood disorder. For example, one can subdivide depressive illness into numerous subtypes based on chronicity and severity of symptoms. Among other things, it is known that severity of illness contributes to the immune disturbances associated with depression (Maes, 1998), but it remains to be established whether chronicity of illness (as in the case of dysthymia), also influences immune outcome. Furthermore, it is possible that the symptom profile, including the presence or absence of neurovegetative features (as seen in typical vs atypical depressive illness) may influence immune factors (e.g., Moldofsky, 1995). Finally, while stressful events are known to influence immune dysfunction, it remains to be established whether the stress or the maladaptive coping styles associated with affective illness (either as an antecedent event or a consequence of the illness) contribute to the immune alterations. For that matter, there is limited information available as to whether the stress perception and coping styles of subtypes of depression are

comparable to one another.

The broad purpose of the present investigation was to assess the behavioral, neuroendocrine and immune characteristics of one subtype of depressive disorder, namely that of dysthymia. In so doing, it was necessary to evaluate other depressive subtypes for comparative purposes. In the present series of experiments we assessed whether (a) depressive illnesses, including major depression and dysthymia, with or without atypical features, are associated with altered stress perceptions and coping styles, (b) depressive illness is associated with alterations of circulating lymphocyte subsets, and whether changes of these subsets are more readily affected by laboratory stressors in depressive groups than in controls, (c) depressive illness is associated with variations of cytokine production (notably interleukin-1 and interleukin-2, primary cell signaling factors released by macrophages and T cells during an immune response), (d) cytokine variations are related to chronicity of illness and symptom characteristics (neurovegetative features), and (e) the immune alterations and psychosocial parameters associated with dysthymia are attenuated following antidepressant treatment.

1.1. Major depression and dysthymia: Clinical and epidemiological features

The literal meaning of dysthymia is 'being of bad mood' or 'ill-humored'. The illness is characterized by a number of affective and neurovegetative features (see Table 1). The diagnosis of dysthymic disorder was introduced in the third edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-III; American Psychiatric Association, 1980) to characterize chronic depression of two or more years and to encompass disorders which had previously been considered characterologically based, including neurotic depression, chronic minor depression, and characterological depression. While the severity of dysthymia is usually less profound than acute major depressive disorder, the symptom profiles of the two are very similar (see Table 1). However, it should be noted that dysthymic symptoms may fluctuate in intensity.

Table 1. DSM-IV (1994) Criteria for Dysthymic Disorder and Major Depressive Disorder

Dysthymic Disorder

- A. Over at least a 2-year period, depressed mood for most of the day, more days than not.
- B. Presence, while depressed, of at least 2 of the following:
- (1) Hyperphagia or poor appetite
 - (2) Increased or decreased sleep
 - (3) Fatigue or low energy
 - (4) Poor self-esteem
 - (5) Difficulty concentrating or making decisions
 - (6) Hopeless feelings
- C. Over the course of the 2-year period of depressed mood, remission of symptoms from A. and B. has not endured for more than 2 months at a time.
- D. During the first 2 years of the disturbance, no major depressive episode has occurred, in order that the condition is not better accounted for by a major depressive episode in partial remission, or chronic major depressive disorder.
- There may, however, have been a major depression prior to the development of the dysthymia, provided that there was full remission (no symptoms over 2 months).
- E. Symptoms of mania have never been present.
- F. The symptoms do not occur exclusively during a chronic Psychotic Disorder.
- G. The symptoms are not due to a general medical condition (e.g., hypothyroidism) or to the effects of a substance (e.g., medication or drug of abuse).
- H. The disturbance causes significant distress.

Major Depressive Disorder

- A. Presence of a major depressive episode:
- (A) Over a two-week period, five or more of the following symptoms (at least one of which is (1) or (2)) have been present nearly every day:
- (1) Depressed mood most of the day
 - (2) Reduced interest / pleasure in most activities
 - (3) 5% weight change over 1-month period, or change in appetite
 - (4) insomnia / hypersomnia
 - (5) psychomotor retardation / agitation
 - (6) fatigue
 - (7) sense of worthlessness or excessive guilt
 - (8) difficulties concentrating or making decisions
 - (9) suicidal ideations
- (B) Symptoms do not occur as part of a Mixed Episode
- (C) Disturbance causes significant distress
- (D) The symptoms are not the result of a substance or general medical condition.
- (E) The symptoms are not accounted for by bereavement (unless symptoms are subsequent to loss of a loved one, and persist beyond 2 months, and markedly impair functioning)
- B. The major depressive episode is not better accounted for by Schizoaffective Disorder, and is not superimposed on a psychotic disorder.
- C. No evidence of mania.

Like major depression, dysthymia is twice as common among women than men. It is one of the more common mood disorders, with a 1 year prevalence rate (for the United States) estimated as high as 5.4% (Regier et al., 1993). Regier et al.'s (1988) multi-site study results indicated that dysthymia was the most prevalent lifetime affective disorder with a prevalence rate of 3.3%. Despite its prevalence, a relatively small proportion of dysthymic individuals seek treatment for their illness, likely due to the insidious onset of symptoms. Indeed, the illness often appears in childhood and adolescence (Kovacs et al., 1984; Keller et al., 1995), and as a result of such long-term low-grade depression, dysthymia might not be perceived as differing from the individual's norm. In fact, Regier et al. (1993) reported that as few as 42.1% of their dysthymic sample had received treatment of any kind. Additionally, the dysthymic group had received the lowest level of mental health service among those with affective disorders, with 19.3% receiving treatment provided by specialists, and 21.2% receiving general medical care.

The characteristics of dysthymia in many ways overlap with those of major depression, although in dysthymia symptoms outnumber signs (i.e., objective characteristics such as vegetative symptoms and psychomotor changes are typically absent) (Akiskal, 1990). In many dysthymic patients a gradual emergence of major depression may occur (double depression), although the patient usually returns to the dysthymic state following remission of the major depression (Akiskal, 1990). Likewise, dysthymia may emerge as a residual syndrome of acute depression (Akiskal, 1983; Keller and Sessa, 1990). In fact, in Judd et al.'s (1997) analyses of the NIMH Epidemiological Catchment Area study, as many as 30% of their dysthymic sample (n=262) reported previously having experienced a major depressive episode, a significantly higher rate than those subjects belonging to the no-symptom or no-disorder groups. These findings support the view (Keller et al., 1983) that major depression and dysthymia have a clinically significant association.

Biological and clinical evaluations of dysthymia have given rise to the notion that several subtypes of the illness exist as determined by specific symptoms, family history, and

age of onset. Akiskal (1983) proposed dimensions on which to refine the classification of dysthymia, namely character spectrum and subaffective dysthymia (i.e., subaffective being of biological origin, while character spectrum is more personality-based). As a result of Akiskal's recommendations, the DSM-III-R distinguished primary vs. secondary dysthymia, and early vs. late age of onset. Primary dysthymia represents dysphoria resulting from an affective disorder, while secondary dysthymia constitutes dysphoria which is the result of a non-affective disorder. Age of onset was subdivided into early (occurring prior to the age of 21 years) and late onset (see McCullough et al., 1996).

Like major depression, dysthymia can be treated by pharmacological intervention, although the efficacy of such treatment is inferior to that seen in major depressive disorder, and may vary as a function of the subtype of dysthymia exhibited (Howland, 1991). Because dysthymia was initially thought to be more of a characterological disturbance than a biologically-based illness, and because of the oft-noted superiority of pharmacotherapy in major depression relative to that observed in dysthymia (Howland, 1991), the illness was typically treated using psychotherapy (including cognitive therapy) (Akiskal and Weise, 1992; Markowitz, 1994). Given the predominant use of psychotherapy over pharmacotherapy in the treatment of dysthymia, the lack of controlled studies comparing the relative efficacy of cognitive therapy and pharmacotherapy (either alone or in combination) is surprising. In fact, recent studies from our laboratory are the first to demonstrate that pharmacotherapy (using a selective serotonin reuptake inhibitor) is the treatment of choice for dysthymia (Ravindran et al., 1999). Of course, this does not exclude a role for cognitive therapy once the primary clinical symptoms are attenuated. As will be seen later, residual features associated with quality of life and coping styles may persist even after depressive symptoms have been alleviated in dysthymic patients by means of pharmacotherapy. Cognitive therapy may be effective in attenuating these residual characteristics and thus may serve to preclude relapse and recurrence of illness (Ravindran et al., 1999)

Despite the relatively high rate at which dysthymia occurs, and its significant relationship and comorbidity with other psychiatric illnesses, it is curious that there have been only limited attempts to identify the mechanisms underlying this disorder. In part, this may stem from the long-standing belief that dysthymia is a neurotic/characterological illness, unlikely to respond adequately to biological treatments. As already indicated, however, it has become clear that dysthymic disorder is responsive to pharmacological interventions. Moreover, there have been recent attempts made to characterize the mechanisms subserving this illness (e.g., Howland and Thase, 1991; Ravindran et al., 1997). In the present review relatively recent biological and pharmacological studies will be outlined, which assess the etiology and treatment of the disorder, and which offer a view as to the processes which may underlie dysthymia (see General Discussion).

1.2. Symptom and illness comorbidity

As indicated earlier, major depression is often superimposed on a dysthymic state (double depression), which may complicate treatment, and is associated with a high rate of illness recurrence (Keller and Shapiro, 1982; Keller et al., 1983). In their epidemiological study, Weissman et al. (1988) reported that 40% of dysthymic patients exhibited comorbid major depression, while more recent evidence suggested that as many as 62% of dysthymic patients met criteria for current major depression, and 80% for lifetime major depression (Keller et al., 1995). Upon remission of the major depression, the double depressive usually returns to the dysthymic state (Akiskal, 1990). The recovery rate for these patients is reduced (39% vs. 61% in double vs. major depression), and recurrence rates within 2 years are appreciably increased (58% vs. 39%) (Keller et al., 1983). It has been suggested that the chronology of occurrence of dysthymia relative to major depression may determine treatment response during subsequent depressive episodes. For example, Levitt et al. (1998) provided preliminary evidence that patients who had experienced dysthymia subsequent to their first episode of major depression, showed a reduced treatment response relative to those patients in

whom onset of dysthymia had preceded their first major depression. These data support the notion that dysthymia which follows an episode of major depression may represent a residual illness, and may indicate a worse prognosis (Keller et al., 1992) than that observed when dysthymia is a prodromal feature of a major depression (Akiskal, 1994). Other investigators have even offered the view that dysthymia may be a life-long disorder that may require sustained maintenance therapy (Ravindran et al., 1999).

As in major depression, anxiety disorder is often a comorbid feature of dysthymia. In fact, as many as 75% of dysthymic patients suffer from some comorbid psychiatric disorder, of which depression, anxiety, and substance abuse are the most common (Weissman et al., 1988). Indeed, approximately one half (50.7%) of Shelton et al's (1997) sample of 410 dysthymic patients reported a history of major depression, while 26.3% had a history of substance abuse, and as many as 68.2% of the sample were diagnosed with a comorbid personality disorder. These investigators also found that prior history of psychiatric disorder differentially influenced the likelihood of prior therapeutic intervention. Specifically, a history of major depression increased the rate of prior psychotherapy as opposed to pharmacotherapy (71% vs. 46% respectively) relative to those without a history of depression (37% vs. 41%). Comorbid personality disorder had somewhat higher rates of prior psychotherapy than those without (59% vs. 50%), while patients with comorbid substance abuse did not have different rates of prior treatment. Unfortunately, these authors did not determine whether a history of comorbid anxiety affected the likelihood of prior treatment.

With respect to comorbid anxiety, Pini et al. (1997) demonstrated that while social phobia and panic attacks were significantly more common among unipolar and bipolar depressives (respectively) than dysthymic patients, the latter exhibited a significantly higher prevalence of generalized anxiety disorder. As such, anxiety disorder comorbidity ought to be more closely scrutinized in dysthymia, as it might be associated with a more severe and enduring symptom profile, as observed in major depression with co-existing anxiety disorder

(Coryell et al., 1988). Furthermore, dysthymia with comorbid anxiety disorder might respond preferentially to adjunctive forms of pharmacotherapy.

In addition to comorbid anxiety, dysthymia often co-exists with personality disorder. Pepper et al. (1995) compared early-onset dysthymic patients (including double depressives) with episodic major depressives, with respect to axis II comorbidity. The data were obtained by means of independent assessment of the patient as well as by interviewing a knowledgeable informant. Both sets of results indicated that more double depressive patients than pure dysthymics met criteria for at least one personality disorder. Conversely, compared to the episodic major depressives, significantly more dysthymics met criteria for an axis II disorder. Interestingly, the dysthymic patients scored significantly higher than the major depressive patients on all of the 13 dimensions of the Personality Disorder Examination, with the most common axis II disorders being borderline, avoidant and histrionic (Pepper et al., 1995).

1.3. Biological aspects of dysthymia

It has been suggested that stressful events or failure experiences result in the formation of attributions, which give rise to negative expectancies of future performance and may result in the development of helplessness (e.g., Abramson et al., 1978; Lazarus, 1993; Seligman, 1975). While stressful events and coping styles (as well as factors such as chronicity, predictability, nature of the stressor experiences, as well as a host of organismic variables) may be associated with the provocation of depression (see Abramson et al., 1978; Brown and Harris, 1978; Brown et al., 1987; Robins and Block, 1988), such an outcome may not be related to the development of "helplessness". Instead depression may result from stressor-provoked deficiencies of norepinephrine (NE) (e.g., Schildkraut, 1965, 1978; Bunney and Davis, 1965), serotonin (5-HT) (Coppen et al., 1972; van Praag and Korf, 1971; van Praag, 1978) or dopamine (DA) (Brown and Gershon, 1993; Jimerson and Post, 1984; Jimerson, 1987; Post et al., 1984; Siever, 1987; Van Praag, 1984), or to variations of DA autoreceptors, 5-HT₂ receptors, α_1 -NE or β -NE receptors

(Antelman and Chiodo, 1983; Meltzer and Lowy, 1987; Sulser, 1984; Vetulani, 1984). Indeed, depression is likely a biochemically heterogeneous disorder, such that the neurochemical underpinnings for the illness, as well as the symptom profile exhibited, may vary dramatically across subjects (Agren, 1980; Banki et al., 1984; Maas, 1975; van Praag, 1984). Moreover, given the abnormal responses to various endocrine challenges (e.g., the dexamethasone suppression test (DST), corticotropin releasing hormone (CRH), as well as thyroid releasing hormone (TRH) challenges), there is reason to suppose that hormonal variations contribute to the provocation or expression of depressive symptoms, and that the contribution of these endocrine factors varies across subjects (Checkley and Arendt, 1984; Prange et al., 1984; Rubin and Poland, 1984; Joffe, 1990; Joffe et al., 1992).

It has been proposed that stressors provoke depressive symptoms owing to neurochemical changes engendered by environmental insults. Although stressors initiate a series of neurochemical changes that may be of adaptive significance, when these neurochemical alterations are insufficient to deal with environmental demands (or neurochemical adaptation does not occur readily), vulnerability to pathology is increased (Anisman et al, 1991). Indeed, animal studies have indicated that stressors will induce many of the central neurotransmitter alterations (e.g., including variations of NE, DA and 5-HT turnover and levels) that have been proposed to subserve the depressive syndrome in humans. Specifically, in response to acute stressors the increased utilization of NE. DA and 5-HT is ordinarily met by adequate synthesis and hence transmitter levels remain stable. However, under conditions that favor amine utilization exceeding synthesis (i.e., if the stressor is sufficiently severe and uncontrollable), a decline of NE levels occurs in several brain regions (Anisman et al, 1991; Weiss and Simson, 1989; Weiss et al. 1981). Such events are also associated with reductions of septal 5-HT and mesocortical DA (Anisman et al., 1989; Glavin, 1985; Heinsbroek et al, 1989; Petty and Sherman, 1982). These amine reductions typically persist for only a few hours, depending on the stressor

severity and organismic variables (e.g., age, species) (Ritter and Pelzer, 1978; Tanaka et al., 1982; Weiss et al., 1981). However, in acutely stressed animals, later reexposure to a mild stressor enhances the utilization of hypothalamic NE (Anisman and Sklar, 1979; Cassens et al., 1980) and mesocortical DA (sensitization effect) (Doherty and Gratton, 1992; Herman et al., 1982; Roth et al., 1988), even if the reexposure involves a different stressor (Nisenbaum and Abercrombie, 1992; Nisenbaum et al., 1991; see Antelman, 1988). It is significant that many of the variables deemed important in promoting human depression, are also those which appear to be fundamental in eliciting the stressor-provoked amine alterations in animal studies (e.g., stressor controllability, predictability, chronicity) (see Anisman et al., 1991; Weiss and Simson, 1985).

In contrast to the amine reductions induced by acute stressors, transmitter levels have been shown to equal or exceed control values following protracted or repeated stressors (Deutch et al., 1985; Herman et al., 1984; Irwin et al., 1986; Kvetnansky, 1980; Roth et al. 1988). These effects are due to increased amine synthesis and/or moderation of excessive utilization ordinarily associated with an acute stressor (Zacharko and Anisman, 1989; Herman et al. 1984). Moreover, chronic insults affect the amine variations engendered by later stressors. Specifically, in addition to a sensitization with respect to amine utilization (seen in the case of acute stress experiences), chronic stressors also induce sensitization of mechanisms associated with amine synthesis, thereby assuring adequate NE supplies upon later stressor encounters (Irwin et al., 1986). In addition, a chronic stressor may result in the down regulation of β -NE receptor activity and the NE sensitive cAMP response (Stone, 1987). However, upon application of a chronic unpredictable stressor the neurochemical adaptation was slower to develop (Anisman et al., 1991) and the β -NE subsensitivity was not evident (Molina et al., 1990). There is also reason to believe that chronic stress may influence both DA and NE autoreceptor sensitivity (Antelman and Chiodo, 1984; Stone, 1983). It has been suggested that under conditions where inadequate neurochemical coping mechanisms are generated, depressive-like characteristics may

evolve. In fact, Anisman and Merali (1997) indicated that a regimen of mild, unpredictable stressors may be precisely the antecedent events most closely aligned with dysthymic-like states.

Data regarding the neurochemical substrates of depressive disorder are limited, owing of course, to the inability to examine brain tissue other than those obtained post-mortem. These data have been reviewed in numerous recent reviews, and will not be reiterated at this juncture (see Holsboer, 1995; Maes and Meltzer, 1995; Plotsky et al 1995; Schatzberg and Schildkraut, 1995). Instead, attention will be devoted to a comparison of the supposed underpinnings of dysthymia relative to major depression.

Howland and Thase (1991) and Ravindran et al (1997) provided reviews of the biological studies of dysthymia or similar disorders. Both reviews indicated that the most consistent findings of the biochemistry of dysthymia originate from neuroendocrine studies related to the DST, as well as the thyrotropin stimulating hormone (TSH) and the growth hormone (GH) response to physiological challenges. In general, these reviews indicated that the rate of abnormal DSTs among major depressive patients differed from that seen in dysthymia. That is, the abnormal DST characteristic of a subset of major depressive patients is not apparent in dysthymics, and in fact, there is reason to believe that the latter is associated with hypocortisol responding (Ravindran et al., 1994a). Paralleling these alterations of pituitary-adrenal axis activity, are findings which revealed differences between major depression and dysthymia in growth hormone secretion in response to physiological challenges, as well as TSH blunting in response to the TRH stimulation test (Szadoczky et al., 1994).

In addition to the effects associated with the DST, it was reported (Maes et al. 1994b) that CRH administration after pretreatment with dexamethasone resulted in an increase of ACTH, β -endorphin/ β -lipotropin, and cortisol among depressive patients. Further, the enhancement of ACTH, but not that of β -endorphin/ β -lipotropin or of cortisol, was more pronounced in major depressives relative to minor depressives (dysthymics). In a subsequent

study, it was observed that relative to minor depressive patients, the levels of ACTH, cortisol, and androstenedione were elevated in major depressive-melancholic patients (Maes et al., 1995). It is not clear, however, whether these differences were a consequence of illness severity or to specific biological symptoms (or neurovegetative characteristics) that are frequent in melancholic patients.

As alluded to earlier, in considering the neuroendocrine correlates of dysthymia, it may be important to distinguish between subtypes of the illness. As will become clear, the characteristics of patients with early onset of the disorder may be distinguishable from those with late-onset of symptoms. It has indeed been proposed that early-onset dysthymics exhibit a higher frequency of personality disorders than their late-onset counterparts (Akiskal et al., 1981; see Hirschfeld, 1990; Klein et al., 1988b; Sansone et al., 1998). Moreover, there is reason to suppose that biological factors may be more closely aligned with early onset of the illness, while late onset may be associated with character spectrum disorder (Akiskal et al., 1983).

As part of a study assessing the role of psychosocial and biological variables in chronic and non-chronic major depression and dysthymia, a lower rate of DST non-suppression was observed in dysthymic patients (52% vs 8.5% non-suppression in major depression vs. dysthymia). However, the rate of DST non-suppression was higher in the early-onset than in the late-onset dysthymics (Szadoczky et al., 1994), although, when double depressives were excluded from the analysis (39 of their 75 dysthymic patients), there was no difference between early and late onset groups with respect to the rate of DST nonsuppression (9% and 8% respectively). As well, it has been reported that the blunted TSH response to TRH stimulation evident in major depression may not occur in dysthymia. Interestingly, paralleling the DST response, the blunted TSH response in dysthymic patients may be dependent upon the age of illness onset (Szadoczky et al., 1994). Among late-onset dysthymics, the blunted TSH response to TRH administration was absent. In contrast, early-onset dysthymics (who parenthetically reported more traumatic and frustrating childhood

backgrounds), had a higher rate of DST nonsuppression, and more frequently exhibited a blunted TSH response after TRH administration. In effect, these data are consistent with the notion that early-onset dysthymia may represent a biologically distinct subgroup of chronically depressed patients (Szadoczky et al., 1994). Indeed, Versiani et al. (1997) noted that early-onset dysthymics responded preferentially to moclobemide relative to imipramine, while no such distinction was found among the late-onset dysthymics. These findings suggest that the monoamine oxidase A isoenzyme might be more imbalanced among early-onset dysthymics.

Limited attention has been devoted to the analysis of monoamine abnormalities in dysthymia. It was reported that reduced levels of plasma NE coupled with elevated levels of platelet and free 5-HT were evident in dysthymia (Lechin et al., 1994). Further, it was observed that following exercise, changes of epinephrine levels were relatively modest in dysthymic patients relative to control subjects. Thus, it was posited that dysthymia may be associated with altered adrenal responsivity to environmental challenges, as well as heightened sympathetic tone as reflected by the elevated free-5-HT levels. In their assessment of platelet monoamine oxidase (MAO) activity in primary dysthymia, Ravindran et al. (1994a) observed reduced platelet MAO activity in primary, early-onset dysthymics relative to control subjects. Moreover, MAO activity prior to treatment was lower among nonresponders than among the drug responders. Along the same line, Wahlund et al (1995), who compared MAO activity in neurotic and endogenous depressives, observed that neurotic depressives tended to exhibit relatively low MAO levels. As well, MAO activity correlated positively with clinical state in the endogenous group. Thus, low MAO activity may represent a marker for vulnerability to neurotic depression. Consistent with the potential involvement of serotonergic mechanisms in dysthymia, lower urinary 5-hydroxyindoleacetic acid (5-HIAA) levels prior to treatment were observed among treatment responders relative to nonresponders, and the reduced levels in responders normalized following treatment (Ravindran et al., 1994b).

There have been few studies that assessed the psychophysiological correlates of dysthymia. However, it was reported that dysthymics exhibited subtle cognitive processing

disturbances of evoked potentials in response to task-relevant stimuli that may be related to difficulties in the processing of complex information. Moreover, hyporesponsiveness in anticipation of aversive stimuli was observed among dysthymics in skin conductance responses (Yee and Miller, 1988). These authors proceeded to study event-related brain potentials in dysthymics in an auditory augmenting-reducing paradigm, and in response to abrupt tone bursts thought to elicit an automatic P300 (a specific brain electrical response to a task-relevant stimulus, thought to be related to cognitive processing/arousal). Although the occurrence of this response in dysthymics could not be distinguished from controls, the dysthymic patients exhibited an early perceptual processing deficit relative to anhedonic subjects and controls, which paralleled results obtained from severe depressives. The investigators speculated that dysthymics may have fewer processing resources available to designate to task-relevant stimuli, skin conductance responses (Yee et al., 1992; Yee and Miller, 1988), as well as in perceptual tasks (Yee and Miller, 1994). Additionally, in a dual-task memory paradigm that assessed the subject's ability to direct resources to simultaneous tasks, a smaller P300 component of the event-related potential was evident in dysthymic patients (Yee and Miller, 1994). Given that the dysthymic subjects did not differ from controls with respect to performance accuracy, latency of the P300 response, and the P300 amplitude to the secondary-task stimuli, it was posited that dysthymia may be associated with an impoverished ability to respond appropriately to external task demands, possibly owing to inappropriate allocation of processing resources. Furthermore, it was suggested that owing to their impaired resource allocation strategies, dysthymic subjects may be more generally impaired in their ability to cope with day-to-day stressors.

Akiskal et al. (1980, 1984) have reported various alterations in the sleep architecture of dysthymia, but such effects were dependent upon the subtype of the illness. For instance, while subaffective dysthymics exhibited shortened REM latencies relative to controls, character spectrum disorder patients could not be distinguished from controls. In addition, dysthymia was associated with excessive and abnormal distribution of REM during the early

part of the night (Akiskal et al., 1984) as occurs in major depression (see Howland and Thase, 1991; Akiskal et al., 1997). However, comparisons between dysthymic and major depressive patients revealed that while the major depressive patients displayed reduced total sleep time, sleep latency, morning wake time and sleep efficiency, the sleep architecture of dysthymics in terms of stage percentages, and REM sleep features, were identical to those of major depressives. In effect, these data are consistent with the notion that the two disorders are variants of the same illness (Arriaga et al., 1995), or perhaps share common underlying mechanisms.

1.4. Pharmacological contributions to the analysis of dysthymia

The most prominent data favoring a biological substrate for dysthymia originate from studies which assessed pharmacological agents in the treatment of dysthymic illness. Admittedly, there have been relatively few studies in this respect, at least in comparison with major depression. This is not surprising given that until recently the treatment of choice for dysthymia had been one of psychotherapy.

The early pharmacological studies in dysthymia revealed that although MAO inhibitors and tricyclic antidepressants (TCAs) had superior therapeutic efficacy to placebo, their effects were not as marked as in major depression (Baldwin et al., 1995; Conte and Karasu, 1992; Howland, 1991; Lopez Ibor et al., 1994; Ravindran et al., 1997; Stewart et al., 1985). However, it has become apparent during the past few years that reliable and impressive effects of antidepressant medications can be garnered in dysthymia. In part, this has occurred owing to the development of the newer medications, such as selective serotonin reuptake inhibitors (SSRIs), characterized by fewer side effects. Moreover, increasing attention has been devoted to tailoring the drug treatment to the specific symptom profile presented (i.e., drug treatments administered primarily to patients with subaffective rather than character spectrum disorder). In fact, more recent studies which have employed rigorous diagnostic criteria and better methodology and design, established the efficacy of tricyclic agents, such as imipramine and

desipramine (Friedman et al., 1995b; Kocsis et al., 1994; Marin et al., 1994; Stewart et al., 1993; Versiani et al., 1992), MAOIs (Petursson, 1995; Stewart et al., 1993; Vallejo et al., 1987), SSRIs such as fluoxetine and sertraline (Hellerstein et al., 1993, 1996; Kocsis et al., 1994; Ravindran et al., 1994a; Thase et al., 1996; Vanelle et al., 1997), the reversible monoamine oxidase inhibitor, moclobemide (Baumhackl et al., 1989; Botte et al., 1992; Duarte and Camozzi, 1993; Lecrubier and Guelfi, 1990; Versiani, 1993; Versiani et al., 1992, 1997), as well as other agents such as the 5HT₂ antagonist, ritanserin (Bakish et al., 1993; Bersani et al., 1991) (see Table 2). As well, we have observed (Ravindran et al., 1999) in a double-blind placebo-controlled study, that sertraline was generally more effective than group cognitive behavioral therapy (CBT) in the treatment of dysthymia. However, group CBT enhanced the effects of sertraline with respect to some of the functional behaviors, which as will be discussed later, has important implications with respect to prevention of illness recurrence.

Because of dysthymia's chronicity and high rate of recidivism, several recent studies compared different antidepressant agents and varying maintenance treatment intervals. Kocsis et al. (1991) indicated that the higher relapse rate following transfer to placebo exceeded that seen in patients who continued on the maintenance desipramine treatment. Paralleling these findings, symptom improvement was sustained, and the rate of relapse reduced, among dysthymic patients who were maintained on either trazodone or fluoxetine over a 40-week interval compared to those who discontinued medication (Hellerstein et al., 1996). Commensurate with the notion that a long-standing illness, such as dysthymia, might require prolonged pharmacotherapy, Vanelle et al. (1997) observed continuous improvement among dysthymic patients treated over a 6-month period, while Kocsis et al. (1996) observed a substantially reduced rate of relapse (11%) among dysthymic patients maintained on desipramine over a two-year period, relative to the 52% relapse rate in the placebo group. Although these data do not necessarily speak to the mechanisms subserving dysthymia, the results from these controlled clinical trials are congruent with the proposition that

Table 2. Pharmacological studies of DSM-III/DSM-III-R/DSM-IV diagnosed dysthymia

Reference	Medication	Outcome
Placebo-controlled		
Stewart et al (1985)	Desipramine	Desipramine = placebo
Harrison et al (1986)	Phenelzine	Phenelzine > placebo
Kocsis et al (1988)	Imipramine	Imipramine > placebo
Tyrer et al (1988)	Doxepin	Doxepin > placebo
Stewart et al (1989)	Imipramine, phenelzine	Imipramine > placebo, phenelzine = placebo, imipramine = phenelzine
Costa-e-Silva (1990)	Amisulpride	Amisulpride > placebo
Bersani et al (1991)	Ritanserin	Ritanserin > placebo
Botte et al (1992)	Moclobemide	Moclobemide > placebo
Hellerstein et al (1993)	Fluoxetine	Fluoxetine > placebo
Stewart et al (1993)	Imipramine, phenelzine	Imipramine > placebo = phenelzine
Versiani et al (1992)	Moclobemide, imipramine	Moclobemide = imipramine > placebo
Versiani (1993)	Moclobemide, imipramine	Moclobemide > placebo, imipramine > placebo
Versiani (1997)	Moclobemide, imipramine	Moclobemide > placebo, imipramine > placebo
Bakish et al (1993)	Ritanserin, imipramine	Ritanserin = imipramine > placebo
Kocsis et al (1994)	Sertraline, imipramine	Sertraline = imipramine > placebo
Thase et al (1996)	Sertraline, imipramine	Sertraline = imipramine > placebo
Vanelle et al (1997)	Fluoxetine	Fluoxetine > placebo
Lecrubier et al (1997)	Amisulpride, imipramine	Amisulpride = imipramine > placebo
Non-placebo-controlled		
Baumhackl et al (1989)	Moclobemide, imipramine	Moclobemide = imipramine
Lecrubier and Gueffr (1990)	Moclobemide, clomipramine	Moclobemide = clomipramine
Marin et al. (1994)	Desipramine	70% response
Ravindran et al (1993)	Fluoxetine	65% response
Smeraldi (1998)	Amisulpride, fluoxetine	Amisulpride = fluoxetine

antidepressants are effective for a substantial portion of dysthymic patients (primarily the subaffective variety), and that prolonged maintenance treatment may be beneficial.

While antidepressant trials have focused on the effects of 5-HT and NE manipulations on the symptoms of dysthymia and major depression, there have been several studies implicating a role for dopamine (DA) in depressive illness. Since DA has been thought to subserve reward processes (Wise, 1985), and anhedonia is a characteristic feature of depression, the view has been taken that reduced DA activity might contribute to the depressive profile (Fibiger, 1995; Kapur and Mann, 1992; Willner, 1987). Indeed, cerebrospinal fluid (CSF) levels of the DA metabolite, HVA, were reduced in depressed patients, although this outcome may have been associated with the motor retardation that often accompanies the illness (Willner, 1987). Further, antidepressant effects have been shown using DA agonists, such as pibedil, bromocriptine, and nomifensine (Kapur and Mann, 1992; Willner, 1987), as well as bupropion, which blocks the reuptake of DA. However, it remains to be established whether the efficacy of bupropion is directly related to its DA action (Kapur and Mann, 1992). Finally, it has been reported that the administration of low doses of neuroleptics, which may act preferentially as autoreceptor antagonists, thus increasing DA synthesis, may alleviate depressive symptoms (Fibiger, 1995).

It will be recalled that in contrast to major depression, anhedonia is not one of the fundamental symptoms of dysthymia according to the DSM-IV criteria. Yet, several investigators have made the case that anhedonia is, in fact, a cardinal feature of dysthymic individuals (Ravindran et al., 1996; Ravindran et al., 1997). Relative to major depression, few studies have evaluated the effects of DA manipulations in dysthymia. Some of these investigations have involved the administration of low doses of the selective D₂ and D₃ antagonist amisulpride (Maier and Benkert, 1994; Leon et al., 1994; Lecrubier et al., 1997; Smeraldi, 1998). Although this agent is most often used as an antipsychotic when administered in high doses (400 to 1200 mg), low doses of this compound (50 mg) attenuated the dysthymic symptoms. In fact, Lecrubier et al.'s (1997) results indicated that amisulpride

was as effective as imipramine in alleviating depressive symptoms in dysthymia, and both agents were significantly better than placebo in this respect. The rationale for enhancing the dopaminergic transmission in dysthymia would be to alleviate the chronic anhedonia, lack of motivation and energy which may characterize these individuals. Unfortunately, these authors report only one parameter measuring anhedonia, that being a factor (anhedonia/asociality) from the clinician-rated Scale for the Assessment of Negative symptoms. In addition to the potential anti-anhedonic action of amisulpiride, there is reason to believe that this agent may be efficacious in the treatment of comorbid anxiety. Smeraldi (1998) compared the efficacy of amisulpiride (50 mg) with fluoxetine (20 mg) over a period of 3 months in a population of predominantly dysthymic patients. While the two medications were equally effective in reducing depression scores as measured by the Montgomery-Asberg Depression Rating Scale, amisulpiride was associated with a significantly greater reduction in anxiety scores as measured by the Hamilton Anxiety Scale.

Animal models of depression have supported the contention that DA may contribute to depressive disorders. In particular, it was reported that chronic, unpredictable mild stressor exposure engendered an anhedonia (Willner, 1995) which was antagonized by chronic antidepressant treatment. Moreover, the administration of a D_2/D_3 antagonist reversed the effects of antidepressants in this paradigm. Predictably, the stressor-provoked anhedonia was accompanied by a reduction of D_2/D_3 receptor binding in the nucleus accumbens. While highly provisional, these data lead to the suggestion that DA mechanisms play some role in depression associated with a chronic stressor. Recently, Anisman and Merali (1998) suggested that the characteristics of this paradigm, as well as the behavioral changes noted, are more closely aligned with a dysthymic-like syndrome than with major depression. Of course, it is exceedingly difficult to distinguish two such similar illnesses within an animal model. However, as will be described shortly, chronic stressor experiences may be linked to dysthymia in humans.

1.5. Genetic factors in dysthymia

There are limited data concerning the genetic factors associated with dysthymia (see review in Goodman and Barnhill, 1995). Since the prevalence rates of various affective illnesses are different in families with dysthymic and major depressive probands, it is likely that dysthymia and major depression are independent disorders (see Howland & Thase, 1991). However, dysthymia may represent a trait factor predicting increased risk for major depressive disorder. Indeed, the high genetic loading may underlie the vulnerability for the early onset of dysthymia (i.e., suggesting a highly penetrant depressive phenotype). Interestingly, while major depression and dysthymia appear to be distinct illnesses, relatives of probands with early-onset depression were at increased risk for, among other things, both major depression and dysthymia (Goldstein et al., 1994).

Further evidence for a genetic component in dysthymia is derived from the finding that dysthymics have a higher rate of family history of depressive disorders compared to nonchronic depressives. In fact, the lifetime rate for major depression was significantly higher in the relatives of dysthymic subjects (Klein et al., 1988a). In a comparative study of pure dysthymia and double depression, high rates of depressive disorders were found in both groups (including both acute and chronic depression), although double depressives had higher rates of chronic depression in their individual relatives and in their families as a whole (Kocsis and Frances, 1987).

The contribution of genetic factors to dysthymia has been gleaned from studies which examined the concordance rate of neurotic depression among monozygotic and dizygotic twins (Slater and Cowie, 1971). Subsequently, Akiskal et al. (1983) indicated a high genetic loading for one subtype of dysthymia. Specifically, as mentioned earlier, based on symptom profile coupled with treatment response to antidepressants, subjects were classified as falling into either the subaffective or character spectrum profile. The subaffective dysthymics, who had a family history of depression, were reported to respond favorably to antidepressants. In contrast, character spectrum dysthymics showed a poor antidepressant response, and were

found to have a significant family history of alcoholism / drug abuse, but not of depressive disorder. Partial support for Akiskal's classification of subaffective vs. character spectrum disorder was obtained by Anderson et al. (1996) in their assessment of dysthymic patients from both subtypes, as well as their first degree relatives. Their results indicated that, consistent with Akiskal's model, there was a higher rate of alcoholism among the relatives of the character spectrum disorder dysthymics, while the subaffective dysthymics exhibited higher rates of depressive symptoms, as well as personality and cognitive features. Unlike Akiskal's classification, however, these investigators did not observe differences between groups with respect to early home environment, family history of mood disorders, gender, or personality disorder.

Riso et al. (1996) assessed the importance of the dimension of personality disorder in dysthymia by determining the patterns of familial aggregation. These investigators assessed the first degree relatives of dysthymic patients with and without comorbid cluster B personality disorder (antisocial, narcissistic, borderline, and histrionic), and compared them to relatives of never-ill controls on the dimension of dysthymia. Whereas Klein et al. (1995) demonstrated that relatives of dysthymic patients exhibited an increased rate of personality disorder, Riso et al.'s (1996) results characterized the nature of the personality disorder which was associated with early-onset dysthymia. They focused on cluster B personality disorders due to their common co-occurrence in early-onset dysthymia (Pepper et al., 1995), and because the traits which characterize these personality disorders reflect the unstable traits by which Akiskal characterized this subset of dysthymics (Akiskal, 1983). Riso et al. (1996) found that, compared to relatives of nondepressed controls, the relatives of both subgroups of dysthymic probands exhibited significantly higher rates of dysthymia with and without cluster B personality disorder, as well as cluster B personality disorder without dysthymia. These results supported the notion that dysthymia and cluster B personality disorder share etiological factors such as genetic or familial factors, neurotransmitter dysregulation, or unhappy early home environments.

Goodman and Barnhill (1995) reported the results of a study comparing the rates of dysthymia in relatives of probands with either panic disorder, major depression, or both conditions (a subset of 33 patients were also dysthymic). Increased rates of dysthymia were observed in relatives of early-onset major depressives, and among relatives of dysthymic probands. A relationship between early-onset major depression and dysthymia was thus supported. However, it is unclear whether this concordance was limited to early-onset of major depression. Unfortunately, the small number of subjects tested makes it difficult to discern whether a still greater risk of dysthymia exists given a dysthymic proband with early- or late-onset of the disorder. The genetic epidemiological findings of Lyons et al. (1998) lend support to the importance of differentiating early- from late-onset major depression. This study involved the assessment of 3372 male twin pairs from the Vietnam Era Twin Registry. Unlike major depression, for which high concordance rates were more likely than for dysthymia (22.5% vs. 14%), monozygotic and dizygotic twins did not differ in their concordance rates for dysthymic illness (7.4% vs. 8.7% respectively), thus supporting the notion of a genetic effect in major depression, but the absence of such a relationship in dysthymia. Similarly, mild-moderate major depression was not associated with a significant MZ/DZ concordance difference, while severe depression was associated with a higher MZ twin concordance rate than that seen in DZ twins. Paralleling these findings, it was also reported that the shared or family environment may contribute more to the etiology of dysthymia than to major depression. It was argued that severity of depression and early-onset of the illness may be more aligned with a genetic association, while the milder depressive illness spectrum (including dysthymia) may be more associated with environmental factors. A curious detail of the study was the definition of early/late age of onset based on the cut-off age of 30 years, whereas DSM-IV defines late onset as 21 years or beyond (American Psychiatric Association, 1994). The authors' justification for choosing this rather advanced cut-off age was that it divided the men into two even groups.

The frequencies of different mood, anxiety, and personality disorders in the first-degree relatives of patients with early-onset dysthymia, episodic major depression, and normal controls were evaluated by Klein et al. (1995). Elevated rates of major depression in relatives of dysthymics compared to relatives of controls, confirmed a familial association between dysthymia and major depression. It was also observed that relatives of dysthymics had higher rates of chronic depression than relatives of episodic depressives. Thus, there appears to be support for familial aggregation in dysthymia, as well as for the validity of dysthymia as a distinct diagnostic category.

Donaldson et al. (1997) explored the distinctiveness of major depression and dysthymia in a family study assessing the first degree relatives of probands with pure early-onset dysthymia, major depression, or double depression. Significantly higher rates of dysthymia were evident among relatives of pure dysthymics and of double depressives, than among relatives of major depressive probands and normal controls. Furthermore, the rates of pure dysthymia did not differ between relatives of pure dysthymics and those of double depressives, nor did they differ between relatives of major depressive and normal control probands. The results lend plausibility to the notion that dysthymia is distinct from major depressive disorder, and that dysthymia and double depression may be variants of the same disorder. The conclusions are clouded, however, by the finding that there was a significantly higher rate of pure major depression among the relatives of pure major depressive probands, as well as among the relatives of double depressives, than among normal controls. These investigators indicate that the possibility cannot be dismissed that dysthymia is associated with two distinct etiological factors. It seems that increased vulnerability to depression occurs (a) in all relatives of unipolar depressive illness, irrespective of subtype, and (b) that risk for dysthymia may be particularly notable among relatives of dysthymic patients and those suffering from double depression.

On the basis of family history data, Remick et al. (1996) argued that minor depression (Research Diagnostic Criteria for minor/intermittent depression), major

depression and double depression were not distinct disorders. Specifically, these investigators compared morbidity risks for mood disorders among the first-degree relatives of patients suffering from either minor intermittent depression, major depression, or double depression. Their results indicated no differences in familial risk of mood disorder between patients with one of the three diagnoses. It remains to be determined whether these subtypes of depression can be distinguished from dysthymia based on family history or genetic factors.

1.6. Psychosocial factors and stressors in major depressive disorder and dysthymia

There is considerable evidence supporting the contention that a relationship exists between stressful events, coping deficits, and the development or exacerbation of major depressive disorder. Considerably less information, however, is available concerning the contribution of these variables to dysthymia. Accordingly, the bulk of the literature reviewed in this section deals with major depressive disorder, after which a brief discourse of the dysthymia literature is presented.

1.6.1. Major affective Disorder

Stressful events or failure experiences may provoke the formation of attributions, which give rise to negative expectancies of future performance, and may result in the development of helplessness (Abramson et al., 1978; Seligman, 1975). It is thought that among individuals vulnerable to depression, the schema (central cognitive processes) is dominated by a cognitive triad of negative attitudes about the self, the outside world and the future. These impose a negative interpretation on the perceived stressful events, hence leading to depression. These negative interpretations may be mediated by pathological processes referred to as habitual cognitive errors (e.g., arbitrary inference, over-generalization, selective abstraction, minimization, and magnification). One treatment of depression has been that of cognitive therapy (Beck, 1967, 1976), the aim of which is to

indicate to patients how cognitions influence mood, and then to teach patients alternative methods of interpreting and dealing with events. It has been reported that cognitive therapy is as effective as pharmacotherapy in the treatment of depression (Hollon et al., 1991; Leahy and Beck, 1988; Safran and Segal, 1990), and in combination with pharmacotherapy, may be superior to pharmacotherapy alone, and may also prolong the effectiveness of pharmacotherapy in preventing depressive relapse (Blackburn et al., 1986; Hollon et al., 1991; Leahy and Beck, 1988). It will be recalled, however, that in a recent study, group cognitive behavioral therapy (CBT) was inferior to pharmacotherapy in the treatment of dysthymia (Ravindran et al., 1999). However, it ought to be noted that these treatments were compared during the acute phase of illness, and it remains to be established whether CBT would be effective in preventing relapse when administered during the maintenance phase (i.e., following remission of symptoms in drug treated patients). Furthermore, it is possible that individual CBT would be more effective than group CBT in the treatment of dysthymia, and the possibility needs to be explored as to whether a CBT regimen tailored to dysthymic illness might be most efficacious in the treatment of this disorder.

As indicated earlier, it has been maintained that depressive symptoms stem from the neurochemical changes engendered by environmental insults. Stressors elicit several neurochemical alterations that may be of adaptive significance, in that they may blunt the psychological or physical impact of environmental challenge, or facilitate responses to deal with the stressor (Anisman et al., 1991), and stimulate processes which prevent excessive physiological activation (McEwen, 1994; Munck et al., 1984). However, if these systems become overly taxed or are insufficient to deal with environmental demands (or when neurochemical adaptation does not occur), vulnerability to depression is increased. In fact, studies conducted in animals indicated that stressors induce many of the central neurotransmitter alterations thought to subserve the depressive syndrome in humans. Moreover, many of the variables considered important in promoting human depression.

influence stressor-provoked amine alterations in animals (e.g., stressor controllability, predictability, chronicity) (Anisman et al., 1993; Weiss and Simson, 1985).

Support for the role of stress in major depression has been derived from both retrospective and prospective studies (Brown, 1993; Kessler et al., 1985; Monroe and Depue, 1991; Monroe and Simons, 1991). To be sure, while stressors may engender depression, they may also exacerbate an already existing depression. Affective illness itself, or the negative perceptions associated with it, may come to generate stress reactions (Price, 1991). Thus, depressed individuals, by virtue of their symptoms and behaviors, generate further interpersonal stressors, which in turn exacerbate the depressive affect (Hammen et al., 1992; Hammen, 1991; McNaughton et al., 1992; Reno and Hillaris, 1990). Moreover, the depressed individual may be particularly reactive to stress (emotionally and neurochemically) hence aggravating the disorder. It should be underscored that individuals may differ in numerous ways which may influence the affective changes elicited by aversive events. Other things being equal (a) the perception of a given stressor will vary across individuals, (b) the neurochemical response to a stressor may vary between individuals, even given comparable perception of events, (c) when stressors lead to pathologies, such as depression, the specific symptom profiles may differ across individuals, and (d) stressful events play a greater role in depression associated with one type of neurochemical disturbance than another (Swann et al., 1990). Additionally, the individual's previous stressor history (including early life trauma) may influence the subsequent response to a stressor, thereby increasing vulnerability to illness (Billings and Moos, 1982; Brown and Harris, 1978; Hammen et al., 1992; Kendler et al., 1992; O'Neil et al., 1987; Roy, 1983, 1985). Post et al. (1984) suggested that stressors, as well as episodes of depression, may influence vulnerability to later affective changes since the neurochemical substrates of the illness may evolve over time and over repeated illness episodes (sensitization effects). Thus, while a first episode of depression may be closely aligned with stressful events, subsequent episodes may evolve spontaneously because of

sensitized systems (alternatively, the apparent contribution of stressors may be less obvious or involve minor stressors that would otherwise have little impact). Indeed, it appears that stressful events are associated with first episodes of depression far more frequently than later episodes (reviewed in Post, 1992).

Not unexpectedly, some stressors are more effective than others in provoking depressive symptoms (e.g., social loss) (Brown and Harris, 1989; Monroe and Simons, 1991; Monroe et al., 1991), whereas others, are more closely aligned with anxiety disorders (e.g., threats or impending stress) (Finlay-Jones and Brown, 1981; Folkman and Lazarus, 1988; Reno and Hillaris, 1990). Although major life events often precede depression, the antecedents of affective illness may involve a series of slight stressors (day-to-day hassles). Indeed, these stressors may have particularly profound effects when applied onto a backdrop of major stressors (Dohrenwend et al., 1984; Kanner et al., 1981; Lazarus, 1990; Monroe and Simons, 1991). Of course, the potency of a stressor in promoting depression may be related to characteristics of the individual, coupled with the nature of the stressor encountered (Brown et al., 1987; Hammen et al., 1989; Overholser, 1990; Robins and Block, 1988; Segal et al., 1982; Zuroff and Mongrain, 1987). Finally, acute stressors may have very different implications than chronic predictable or chronic intermittent stressors (Monroe and Simons, 1991). Chronic intermittent stressors may not lend themselves to neurochemical adaptation and may be most likely to result in behavioral disturbances (Anisman et al., 1991; Willner, 1987).

The impact of stressors is related to the individual's ability to cope, or to perceive the stressor as being controllable (Abramson et al., 1978; Alloy and Clements, 1992; Seligman, 1975). In the face of uncontrollable stressors some individuals, particularly those with accurate and realistic judgment, would be inclined towards depression. Conversely, perceiving the stressor as being controllable, even if this perception is illusory, would reduce the likelihood of hopelessness and depression (Alloy and Clements, 1992; Needles and Abramson, 1990). In addition, major depression has been associated with

reduced social support, and dissatisfaction with the emotional component of this support, as well as the use of emotion-focused coping strategies rather than problem-focused coping (Hollohan and Moos, 1991; Lora and Fava, 1992; McNaughton et al., 1992; Roy-Burne et al., 1992). With successful pharmacological treatment of depression, several aspects of life satisfaction and psychosocial functioning were modified, suggesting that impaired psychosocial functioning is a result of the affective disorder, rather than a manifestation of characterological disturbances (Overholser et al., 1990; Perry and Shapiro, 1986).

1.6.2. Dysthymia

In view of the shared attributes between dysthymia and major depression, it is somewhat surprising that little information is available concerning the contribution of stress to the provocation of dysthymic disorder. Inasmuch as dysthymia is a chronic illness which persists for at least two years, it may be difficult to identify specific life events that precipitated illness onset. Nevertheless, it would not be unreasonable to propose that some adverse life events, particularly the inability to cope with day-to-day annoyances, or alternatively chronic stressor experiences, may precipitate or aggravate the illness (Monroe and Simons, 1991).

The frequently observed social and interpersonal impairment in dysthymia has been confirmed by systematic studies (Cassano et al., 1990; De Lisio et al., 1986; Freeman, 1994; Friedman, 1993; Friedman et al., 1995a). These reports indicate that social impairment correlates positively with increasing severity and chronicity of the illness, and worsens significantly with onset of double depression. In addition to alleviating the depressive symptoms, antidepressants can significantly improve the social impairment characteristic of dysthymic patients. Markowitz et al. (1996) demonstrated that acute treatment (10 weeks) with desipramine significantly improved interpersonal functioning in dysthymics, as measured by the Inventory of Interpersonal Problems (IIP). While improvement continued over a 16-week maintenance phase, this was not significant, and although dysthymic IIP scores approached

normative values, they did not attain this level. In addition to impaired interpersonal functioning, quality of life is substantially reduced in depressive disorders (Walker et al., 1995; Wells et al., 1989). Likewise, dysthymic patients often exhibit occupational and social impairment (Friedman, 1993; Friedman et al., 1995a), and the latter may be reduced following antidepressant treatment (Friedman et al., 1995a).

Significant improvement in quality of life was observed among dysthymic patients who were treated with either imipramine or sertraline over 12 weeks, relative to placebo-treated patients (Kocsis et al., 1997). The drug-treated patients also exhibited significant improvement in social adjustment. Interestingly, however, it was pointed out that patients' self-reported social functioning showed greater improvement than participation in leisure activity. The authors suggest that the delayed alleviation of certain aspects of psychosocial functioning may have been due to the relatively short duration of pharmacotherapy relative to the patients' often life-long impairment. In this respect, it should also be considered that the well entrenched anhedonia and impaired psychosocial functioning among dysthymics may contribute to the unremitting nature of the illness, as opposed to being a secondary factor. Contrary to this position, however, Leader and Klein (1996) observed that the strongest predictor of impaired social functioning among dysthymic patients was current depressive symptomatology and not illness duration, as the preceding notion would suggest. Surprisingly, the differential impact of life stress on social impairment was not as substantial as the detrimental effects of depressive symptoms.

It has been reported that specific aspects of life satisfaction and psychosocial functioning improved among dysthymics who responded to antidepressant treatment. Thus, it was posited that the reduced capacity to enjoy leisure time may be a state marker of chronic depression (Agosti et al., 1991; Friedman et al., 1995a,b). In fact, the response to desipramine among dysthymic patients could be predicted by extended family adjustment (Friedman et al., 1995b). Those patients who reported less difficulty with their partner or relatives, as measured by the Social Adjustment Scale at baseline, were more likely to respond

to desipramine than those who reported more family dysfunction. Similarly, baseline levels of overall social impairment were somewhat higher in nonresponders than responders.

While there is reason to suppose that stressful events contribute to the provocation of dysthymia, prospective studies have not been conducted to assess the contribution of life stressors to this illness. There have, however, been studies that assessed the potential contribution of stressors to the overlapping category of neurotic depression and non-neurotic depression. It was reported, for instance, that both illnesses were preceded, to a comparable degree, by a severe life event or major difficulty (Brown et al., 1979). Additionally, little difference was observed with respect to the perception of stressful life events between neurotic and non-neurotic depression (Benjaminsen, 1981; Katschnig and Berner, 1984; Monroe et al., 1985). However, neurotic depressives perceived significantly more life events compared to endogenous depressives (Bebbington et al., 1981; Roy et al., 1985).

It is not surprising, given the chronicity and frequent early onset of dysthymia, that prospective studies have not been conducted concerning the stressful events that may have preceded illness onset. However, it was reported that among adults who acted as care-givers for a spouse with a progressive dementia (care-giving itself is a profound stressor), a substantial proportion (30%) suffered a depressive disorder compared to 1% in a matched control sample. In fact, the incidence of major depression and dysthymia did not differ from one another (Dura et al., 1990). Thus, these data are consistent with the proposition that dysthymia, like major depression, may be provoked by chronic uncontrollable stressor conditions. It remains to be determined whether a chronic regimen of minor stressors would likewise be associated with dysthymic symptoms.

As indicated earlier, it has been suggested that depressive illness, and particularly dysthymia, may be a life-long disorder. Moreover, the high rate of recurrence of major depressive disorder may stem from under-treatment of the illness, as reflected by the persistence of residual symptoms (Markowitz, 1994; Ramana et al., 1995; Fawcett, 1994). It is similarly possible that those treatments which permit residual dysthymic features to

persist (e.g., inadequate antidepressant dosage or insufficient duration of treatment) may also favor recurrence of this illness. It is certainly conceivable that by virtue of its effects on the secondary features of dysthymia, CBT may act to limit illness recurrence (Ravindran et al., 1999). Given that quality of life changes may be a fundamental characteristic in identifying the efficacy of treatment response (Ravindran et al., 1999), such a functional measure may also be useful in distinguishing genuine drug responders from drug-treated patients actually exhibiting a placebo response. Such a differentiation may prove to be a valuable tool in predicting relapse, and may be useful in evaluating the efficacy of treatment.

1.7. Stress, depression and the immune response

In animals, acute stressors provoke suppressed natural killer (NK) cell activity, inhibit cell proliferation in response to T and B cell mitogens, and reduce the antibody forming cell response (Anisman et al., 1993; Herbert and Cohen, 1993; Keller et al., 1991). Of course, factors such as stressor severity, predictability and chronicity influence the magnitude and nature of the immunological changes (Anisman et al., 1993; Irwin and Livnat, 1987; Irwin and Hauger, 1988; Jessop et al., 1987; Zalcman et al., 1989). These outcomes may be dependent upon the immune compartment examined. While stressor effects on the splenic mitogen response may involve NE activation (and not adrenal corticoids) and is subject to an adaptation following repeated stressors, the mitogen response in blood is affected by adrenal corticoids, is not influenced by NE manipulations, and is not subject to an adaptation with repeated stressors (Cunnick et al., 1988, 1990; Rabin et al., 1989).

Human studies revealed that stressors (e.g., bereavement, divorce, loneliness, academic examination and parachute jumping) altered various aspects of immune functioning, such as reducing NK cell activity, lowering gamma interferon synthesis, suppressing cell proliferation in response to mitogens, altering expression of interleukin

(IL)-2 receptors, modifying the immunologic response to hepatitis B vaccine, and impairing immune control in reactivation of latent herpes virus (Abraham, 1991; Bartrop et al., 1977; Daruna and Morgan, 1990; Dorian and Garfinkle, 1987; Glaser et al., 1985; 1987; 1992; Irwin et al., 1987a,b; Kiecolt-Glaser et al., 1984; Schedlowski et al., 1993b; Schleifer et al., 1983; Stein et al., 1982; see Herbert and Cohen, 1993; Weisse, 1992). As well, the potential for illness may be influenced by the individual's ability to contend with the stressor behaviorally (Glaser et al., 1992). Further, it seems that the nature of the immune changes may be related to the time at which blood samples are taken relative to the occurrence of the stressor. For example, the period preceding stressor onset (e.g., anxiety associated with a parachute jump) was associated with immune activation, whereas a period of compromised NK cell activity followed stressor termination (Schedlowski et al., 1993b). It seems that although anticipatory stress mobilizes immune activity, a phase of quiescence (recuperation) follows the stress event.

As in the case of stressors, major depression has frequently been associated with compromised immunity (Calabrese et al., 1987; Evans et al., 1988; Herbert and Cohen, 1993; Kiecolt-Glaser and Glaser 1991a,b; Kiecolt-Glaser et al., 1991; O'Leary, 1990 but see Stein et al., 1991), including alterations in the numbers of leukocytes (Kronfol and House, 1984; Kruger et al., 1984; Schleifer et al., 1984, 1985; Targum et al., 1989), higher numbers and percentage of CD25+ (IL-2 receptor bearing cells) and CD4+ cells (Maes et al., 1991), a reduction of lymphocyte proliferation in response to mitogens (Calabrese et al., 1987; Cosyns et al., 1989; Darko et al., 1986; Kronfol and House, 1984; Kronfol et al., 1989, 1982, 1983; Levy et al., 1991; Schleifer et al., 1994; Syvalahti et al., 1985), as well as impaired cell-mediated immunity (Hickie et al., 1993). Such effects were particularly marked in depressed patients exhibiting melancholia (Hickie et al., 1993), and at least some of the immune changes (altered lymphocyte proliferation) were independent of age, sleep disturbances, menopausal status, prior treatment with benzodiazepines, and illness severity (Cosyns et al., 1989; Hickie et al., 1993). One of the more prominent

immune changes associated with depression is that of reduced NK cell activity (Darko et al., 1992; Evans et al., 1992; Irwin and Gillin, 1987; Irwin et al., 1987b, 1990a, 1992; Maes et al., 1992b; Mohl et al., 1987; Nerozzi et al., 1989; but see Stein et al., 1982), which may be mitigated with symptom remission (Irwin et al., 1992). Paralleling these findings, depression (with melancholia) was associated with blunted β -adrenergic responsivity of lymphocytes (Magliozzi et al., 1989; Mann et al., 1990), and this effect abated following successful treatment with electroconvulsive shock (Mann et al., 1990).

Despite the plethora of data showing compromised immunity in depressed patients, there have also been contradictory findings demonstrating that depression was neither associated with reduced NK cell activity (Miller et al., 1993; Schleifer et al., 1989), nor suppression of the mitogen response (Albrecht et al., 1985; Darko et al., 1989, 1991; Schleifer et al., 1985, 1989; Sengar, et al., 1982; Stein et al., 1982). Weisse (1992) proposed that these inconsistencies stemmed from the assessment of patients with varied depressive diagnoses (uni- and bipolar), relatively young depressive subjects, and severity of illness being in the moderate range (the suppression is most evident in melancholic depressives; Evans et al., 1992; Maes et al., 1992b). It is unlikely that the immunosuppression exhibited by depressed inpatients was due to institutionalization, since such an effect was not observed among hospitalized schizophrenic patients (Caldwell et al., 1991). Parenthetically, since immunosuppression has been associated with both depression and stressful events, it has been speculated that the altered immune functioning in depression was secondary to precipitating stressors (Price, 1991). However, several studies dissociated the two phenomena, and suggested that the immune alterations were related to the affective disorder rather than to the antecedent stressors (Irwin and Hauger, 1988; Irwin et al., 1990a, 1992; Linne et al., 1984; Locke et al., 1984; see Weisse, 1992).

In addition, to the core symptoms of depression, it is also conceivable that the patient's neurovegetative profile may come to affect immunological parameters. Specifically, while depression is often associated with neurovegetative features such as

hypophagia, insomnia, and reduced appetite, the opposite profile is often manifested. The latter depressive profile, known as “atypical” depression, is characterized by ‘reversed’ neurovegetative features including hyperphagia, hypersomnia, carbohydrate craving, and leaden paralysis. In addition to these physiological alterations, atypical depressive individuals are characterized by behavioral features such as hypersensitivity to criticism, irritability, mood reactivity and rejection sensitivity (which persists even beyond symptom remission). Inasmuch as sleep-depriving normal control subjects has immunosuppressive effects (Moldofsky, 1995), similar to those observed in certain depressive populations, the possibility ought to be considered that the other neurovegetative features may also be associated with immune alterations. Finally, although numerous studies have investigated the variations in immune functioning associated with affective disorders, there are currently no data available as to whether the immune changes are related to the chronicity of the depressive episode.

While major depressive illness may be accompanied by suppression of some aspects of immune functioning, there have also been indications of immune activation, particularly in severely depressed (melancholic) patients (Maes, 1995). There is evidence suggesting that depression may be associated with variations of either circulating cytokine levels (i.e., substances released from activated immune cells that are fundamental in cell-to-cell communication), or cytokine production from mitogen-stimulated cells. In this respect, it seems that the immune profile of severely depressive patients is not unlike an acute phase reaction (i.e., the immediate immunological and neuroendocrine response elicited by immune challenge such as infection). Among other things, depressed patients exhibit increased plasma concentrations of complement proteins, C3 and C4, and immunoglobulin (Ig) M, as well as positive acute phase proteins, haptoglobin, α 1-antitrypsin, α 1 and α 2 macroglobulin, coupled with reduced levels of negative acute phase proteins. As well, depression was associated with an increased number of activated T cells (CD25⁺ and HLA-DR⁺), secretion of neopterin, prostaglandin E2 and thromboxane, as well as increased

concentrations of soluble IL-2 receptors (sIL-2R), IL-1 β , IL-1 receptor antagonist (IL-1ra), IL-6, soluble IL-6 receptor (sIL-6R), and γ -interferon (IFN) (Connor and Leonard, 1998; Maes, 1995; Maes et al., 1991, 1992a, 1993b, 1995; Mullar and Ackenheil, 1998; Nassberger and Traskman-Bendz, 1993; Smith, 1991; Song et al., 1994). While there have been reports that the elevated levels of IL-1 β , IL-6 and α 1-acid glycoprotein normalized with antidepressant medication (Sluzewska et al., 1995), it was demonstrated that the upregulated production of sIL-2R, IL-6 and sIL-6R was not attenuated with antidepressant medication, leading to the suggestion that the latter factors may be trait markers of the illness (Maes et al., 1995).

Although severity of depressive illness is likely fundamental in determining cytokine levels (Maes, 1995), the possibility cannot be ignored that characteristics of the illness might also contribute in this respect. For instance, it is possible that the neurovegetative features of depressive illness contribute to the cytokine alterations. Likewise, it is conceivable that chronic depression (or chronic stress), may induce cytokine changes to a greater extent than those observed following acute episodes (as in the case of major depression). The limited data presently available concerning cytokine changes in depressive illness (i.e., studies showing elevations of the cytokines in severe major depression) are largely correlational. Thus, it is unclear whether the cytokine alterations seen in affective disorders are secondary to the illness (or the stress associated with the illness), or play an etiological role in the provocation of the disorder. Yet, high doses of IL-2, IFN- α and tumor necrosis factor (TNF-) α in humans undergoing immunotherapy have been shown to induce neuropsychiatric symptoms, including depression, and these effects were related to the cytokine treatment rather than to the primary illness (Meyers and Valentine, 1995). Of course, the doses administered in these studies were in the pathophysiological range, and thus their relevance to depression per se may be questioned. This notwithstanding, as IL-1 β potently stimulates the release of CRH (Dunn, 1995; Rivier, 1993), the possibility exists that this cytokine contributes to the altered HPA

functioning characteristic of major depressive illness.

1.8. Experimental Objectives

It is clear from the preceding discussion that stressful events are associated with major depressive disorder, and may also contribute to the exacerbation of dysthymic disorder. Furthermore, it seems that major depression may be associated with an immune activation, much like an acute phase reaction. Data are unavailable, however, as to whether such effects are related to the chronicity of illness. For that matter, not a single report has been published concerning the levels or production of cytokines in dysthymic patients. In view of the relationships that may exist between stressful events/coping and immune functioning in dysthymic patients, the present investigation sought to establish whether:

- (a) dysthymia, like major depression, is associated with altered stress perceptions and coping styles, and if so, whether these normalize with successful treatment of the disorder;
- (b) dysthymia is associated with alterations of circulating lymphocyte subsets, and whether changes in these subsets following stressor exposure are more readily affected in depressive groups than in controls;
- (c) alterations of circulating lymphocytes are related to the neurovegetative features associated with either major depression or dysthymia;
- (d) disturbances of circulating lymphocyte subsets normalize with successful treatment (pharmacotherapy) of depressive disorders;
- (e) depressive illness is associated with variations of cytokine (IL-1 β and IL-2) production;
- (f) cytokine variations are related to chronicity of illness and symptom characteristics (neurovegetative features).

Prior to being initiated, each of the experiments received ethics approval by the Research Ethics Committee of the Royal Ottawa Hospital. Accordingly, written informed consent was obtained from each participating subject, once the procedures had been fully

explained. In the case of patients, informed consent comprised a clinical drug trial consent as well as the laboratory procedure consent, whereas, in the case of non-depressed control subjects, only a laboratory procedure consent was required (see Appendix A).

2.1. Experiment 1: Stressful Life Events And Coping Styles in Dysthymia and Major Depressive Disorder: Effects of Pharmacotherapy

It will be recalled that life stressors may be associated with the onset of major depressive disorder (Brown and Harris, 1989; Monroe and Depue, 1991), as well as the course of the illness once it has begun (Billings and Moos, 1985; Brown and Harris, 1978; Hammen et al., 1986; Monroe et al., 1992; Monroe et al., 1983). Irrespective of the theoretical position adopted regarding the relationship between stress and depression, it is generally agreed that certain adverse experiences are more conducive than others to the provocation of depressive symptoms (Monroe and Simons, 1991). However, life events associated with affective illness are not restricted to those which are intuitively the most severe. Although major life events have been identified in the months preceding major depression (Hammen et al., 1989; Monroe and Depue, 1991; Monroe et al., 1991), it seems that day-to-day inconveniences likewise contribute to the behavioral profiles observed (Lazarus, 1993; Lazarus and Folkman, 1984).

To a great extent the impact of aversive events on major depressive disorder, as indicated earlier, may be related to the individual's ability to cope with the stressor (Alloy and Clements, 1992; Parker et al., 1986). In fact, commensurate with the view that coping factors are essential in determining the impact of stressors on affective state, relative to non-depressed individuals, major depressives have fewer social supports, and report reduced satisfaction with the emotional component of this support. Moreover, depressed patients tended to favor emotion-focused coping strategies rather than the more adaptive problem-focused type of coping style (McNaughton et al., 1992; Holohan and Moos, 1991; Roy-Byrne et al., 1992). In light of the shared attributes between major depression and dysthymia, it would not be unreasonable to expect that some forms of adverse life events, particularly the inability to cope with day-to-day annoyances (daily hassles) may serve to precipitate or exacerbate the illness (Monroe and Simons, 1991). Experiment 1 assessed (a) whether major depression and dysthymia were associated with heightened perception of

major and minor life stressors, and differences in coping styles relative to non-depressed subjects, and (b) whether treatment with SSRIs, which are effective in alleviating symptoms of major depression, would attenuate the stress/coping disturbances in major depression and dysthymia.

2.2. Method

2.2.1. Subjects

The subjects consisted of 18 primary dysthymic patients (9 male, 9 female), 17 major depressives (8 male, 9 female), and 18 normal controls (9 male, 9 female), ranging in age from 21-54. The dysthymic patients tended to be older than the major depressive patients (mean age = 41.63 ± 1.59 and 35.98 ± 2.61 , respectively), and 11 of the 18 dysthymic patients had exhibited early illness onset (i.e. < 21 years). The patients, were obtained through referrals to the Mood Disorders Clinic (outpatients) of the Royal Ottawa Hospital, satisfied DSM-III-R criteria for the respective disorders, and were free of any other Axis I disorders. Several of the dysthymic patients were self-referrals who responded to a recruitment advertisement in the Ottawa Citizen (see Appendix A). During screening, those subjects who exhibited personality difficulties sufficient to make a clinical diagnosis of a personality disorder were excluded from the study. This should not, however, be taken to imply that the patients did not exhibit traits associated with personality difficulties, but rather that these were not of sufficient magnitude to fulfill the DSM-III-R/DSM-IV criteria. Exclusion criteria also comprised viral illness during the preceding 2 weeks, any physical illness, such as severe allergies, multiple adverse drug reactions, hypertension, significant recurrent dermatitis, malignant, hematologic, endocrine, pulmonary, cardiovascular, renal, hepatic, gastrointestinal or neurologic disease, or any medical disorder that required drug treatment which may have affected immune activity. Finally, pregnant or lactating women were not included in the investigation. Comorbid personality disorders and other psychiatric disorders were excluded on the basis of clinical and DSM-III-R criteria. Major depressive patients had a primary diagnosis of mild-moderate

depression on the Clinical Global Impressions scale (CGI). The dysthymic patients had a diagnosis of primary dysthymia, and met Akiskal's criteria (1990) for subaffective subtype (i.e., early indeterminate age of onset, continuous symptomatology or fluctuating course over many years, but not free of affective manifestations for more than a few weeks at a time, presence of anhedonia, and depressive temperament such as passivity, introversion, gloominess, pessimism, self reproach, brooding, preoccupation with inadequacy, and hypercritical or complaining).

The control subjects, comprised blue- and white-collar university and hospital staff. The subjects were matched as closely as possible for age and sex to the study populations, and were screened using a clinical interview and a self-rating Beck Depression Inventory (BDI; Beck et al., 1961) to exclude any significant affective pathology.

2.2.2. Assessments

Symptoms of depression were measured using standardized instruments (see Appendix B), including the Hamilton Depression Rating Scale (HAM-D; 17-item) (Hamilton, 1967), the Montgomery-Asberg Depression Rating Scale (MADRS) (Montgomery and Asberg, 1979), the Hamilton Anxiety Scale (HAM-A) (Hamilton, 1959), and the Clinical Global Impressions Scale (CGI) of severity (Guy, 1976). The HAM-D and HAM-A total scores, at baseline and following 4 and 8 weeks of treatment, are shown in Table 3.

Patients underwent a full physical examination and clinical evaluation which included full blood count, urinalysis, and cardiogram. None of the subjects had received psychotropic medication during the preceding 6 months. After the diagnostic interview was performed, and informed consent was obtained, all patients underwent a routine one week placebo run-in phase. Upon commencement of the study, data gathered included demographic information, characteristics of the current episode (from the depressed groups), and personal and family history of psychiatric illness.

Clinical and behavioral measurements were taken prior to drug treatment and were repeated following four and eight weeks of treatment. The behavioral measurements, which were completed by all subjects, consisted of a series of questionnaires rating stressful life experiences and coping styles used (see Appendix C). These included a Life Events Scale to determine major life changes (Paykel et al., 1971), and the Daily Hassles and Uplifts Scales to determine minor life stressors and positive experiences (Kanner et al., 1981). As well, the degree of loneliness was measured using the UCLA Loneliness Scale (Russell et al., 1980), and the Coping Strategies Scale (Beckham and Adams, 1984) assessed the coping styles that subjects employed. This coping scale is subdivided into 10 subcomponents and 3 composite scores. The ten subcomponents include: blame (blaming oneself or someone else for one's problems), emotional expression (venting either anger, anxiety, despondency), emotional containment (keeping feelings of anger, depression, anxiety to oneself), social support seeking (looking to others to listen to and/or help with one's problems), religious support (using religious institutions, scriptures, or God to deal with problems), cognitive restructuring (thinking about and appraising problems in new ways rendering them less aversive or threatening), general activity (using physical or mental activities unrelated to one's problems; these methods neither solve nor avoid problems), avoidance/denial (avoiding thinking about or being reminded of one's problems), problem-solving (obtaining information about problems, evaluating possible solutions, deciding upon or carrying out a course of action), passivity (mentally accepting or dwelling on problems, without trying to solve or avoid them). The three composite scales include: individual coping (attempts to help oneself by becoming involved in a variety of activities) / cognitive restructuring, emotional containment / passivity, and finally, emotional expression / social support seeking. Some of these strategies are more oriented towards emotional coping approaches, whereas others are directed more to problem-solving styles.

2.2.3. Drug Administration

The patients from both depressive groups participated in a treatment program to assess the relative effectiveness of SSRIs. Patients were randomly assigned to treatment conditions and received either sertraline (dose range 50-200 mg), fluoxetine (20-60 mg), fluvoxamine (50-200 mg), or paroxetine (20-40 mg). The control subjects completed the behavioral measures, but did not receive any form of treatment.

2.2.4. Data Analysis

During the course of the experiment, 5 major depressive and 5 dysthymic patients were lost from the study. Two depressive patients exhibited worsening of symptoms, 2 patients did not comply with the experimental protocol with respect to medication, and the remainder failed to complete the scales appropriately. As the baseline scores of these subjects were consistent with the others in their respective groups, their data were included in the analyses of baseline scores, but were not included in the repeated measures analysis of variance to determine changes in the various behavioral measures over the course of treatment.

Inasmuch as nondepressed controls were not treated, a separate analysis of variance was conducted to compare the major depressive, dysthymic and control subjects at baseline (prior to drug treatment), while a repeated measures analysis of variance over the course of the antidepressant regimen was conducted comparing the two depressed groups to their respective baseline measures. Dunnett's tests were used to compare the means of the drug-treated major depressive and dysthymic patients to the non-drug treated control subjects. In effect, the control subjects served as a normative sample against which to compare drug efficacy. Since several patients failed to successfully complete all portions of the behavioral tests, the degrees of freedom for several of the behavioral measures differed from those of the analyses of the HAM-D and HAM-A scores.

2.3. Results

2.3.1. Clinical Measures

As shown in Table 3, the drug treatments were clearly effective in reducing the HAM-D scores in both groups. The analysis of variance of the HAM-D scores revealed a significant interaction between Affective state and Time of evaluation, $F(2, 50)=7.29$, $p < .01$. Newman-Keuls multiple comparisons ($\alpha = .05$) confirmed that prior to treatment the HAM-D scores were significantly higher in the major depressive than in the dysthymic patients. The HAM-D scores declined with antidepressant treatment, such that after 4 weeks the scores were still somewhat elevated in dysthymic patients, but after 8 weeks the HAM-D scores in the two groups, which were in the normal range, did not differ from one another. It might be noted that 10 dysthymic patients, who were maintained on an SSRI, had mean HAM-D scores of 2.5 ± 1.01 and 2.23 ± 0.91 after 9 and 12 months, respectively. Scores on the MADRS and on the CGI paralleled those of the HAM-D, and need no further elaboration. Thus, these data support our earlier contention that administration of an SSRI is as effective in the treatment of dysthymia as in the treatment of major depression, although the duration of symptoms is longer in the former condition. It ought to be underscored that although a placebo control was not employed in the present investigation, the drug treatments were effective in reducing the HAM-D scores by 50% in 14 of 17 dysthymic patients and 11 of 13 major depressive patients. These values appreciably exceed those that might be expected in the absence of therapy. Indeed, given the chronic nature of dysthymia, coupled with the relatively limited success obtained with other drug therapies (Howland, 1991), it is hardly likely that the effects of the 5-HT reuptake blockers in the present investigation represent a spurious finding.

Unlike the HAM-D scores, the analysis of variance revealed that the two patient groups did not differ from one another with respect to HAM-A scores. However, the HAM-A scores declined over the course of treatment, $F(2, 46) = 48.30$, $p < .01$. The multiple comparisons indicated that the scores on this measure declined between the initial

Table 3.

Mean ($\pm$ S.E.M.) HAM-D and HAM-A Scores For Major Depressive and Dysthymic Patients at Baseline and Following 4 and 8 Weeks of Pharmacotherapy

	<u>MAJOR DEPRESSIVE</u>			<u>DYSTHYMIC</u>		
	Baseline	Week 4	Week 8	Baseline	Week 4	Week 8
HAM-D	24.4 $\pm$.76	7.6 $\pm$ 1.94*	7.8 $\pm$ 1.49*	20.4 $\pm$.82	11.6 $\pm$ 1.42*	7.1 $\pm$ 1.11*
HAM-A	15.0 $\pm$ 1.13	5.3 $\pm$ 1.54*	6.2 $\pm$ 1.49*	11.9 $\pm$.70	6.6 $\pm$.90*	4.3 $\pm$.81*

* p<.05 relative to Baseline scores

visit and 4 weeks of treatment and remained stable thereafter.

2.3.2. Perception of Stress and Uplifts

Figure 1 shows the mean hassles, uplifts, loneliness and life events scores for each of the groups at the commencement of the study, as well as after 4 and 8 weeks of treatment. Analysis of variance revealed that although major life events encountered over the preceding 6 months were somewhat greater in depressed and dysthymic subjects than in controls, their reported occurrence was variable, and the between-groups difference did not reach statistical significance, $F(2,50)=1.71$, $p=.19$.

The frequency and severity of hassles determined at baseline were found to be similar to one another. In contrast to major life events, the frequency and the severity of Hassles varied across the groups, $F(2,51)= 8.48$ and 8.09 , $p's<.01$, respectively. Newman-Keuls multiple comparisons ($\alpha=.05$) confirmed that at baseline the frequency and severity of hassles in dysthymics exceeded those of the major depressive patients, which in turn exceeded those reported by control subjects. The repeated measures analysis of variance in the dysthymic and major depressive patients revealed a significant decline in the frequency and severity of hassles over the 8 weeks of treatment, $F's(2,44) = 8.80$ and 9.89 , $p's<.01$, respectively. It is clear from Figure 1 that despite the marked decline of hassle severity in both the depressive and dysthymic groups, the scores still exceeded those of control subjects.

The increased severity of hassles seen in the patient populations was not simply a reflection of an increase in the frequency of such events. In particular, analysis of the Hassles severity/Hassles frequency (i.e., the average intensity per hassle) revealed that the average hassle intensity varied as a function of the Affective condition $\times$ Time of evaluation interaction, $F(4, 70)=2.48$, $p=.05$. Multiple comparisons of the simple effects confirmed that at baseline the average hassle intensity in control subjects was lower (mean = $1.21 \pm .05$) than in the major depressive or dysthymic conditions (mean = $1.78 \pm .12$, $1.79 \pm .11$,

Figure 1. Mean ($\pm$ S.E.M) severity of daily Hassles, intensity of Uplifts, intensity of Loneliness, and scaled Life Event scores among controls, major depressives (MDD), and dysthymics (DYS) at baseline (admission) and following 4 and 8 weeks of treatment. Life Events were measured only at Admission and Week 8.

* $p < 0.05$ relative to control values; + $p < 0.05$ relative to admission.

respectively). Over the course of the 8 weeks of treatments the average hassle intensity declined significantly in both patient groups (mean = $1.46 \pm .11$, $1.43 \pm .07$, respectively), but still exceeded the average hassle intensity of control subjects (mean = $1.13 \pm .06$).

The frequency and the intensity of uplifts at baseline varied as a function of the Affective state, F 's (2,50) = 4.56 and 5.16, p 's <.05. Newman-Keuls multiple comparisons confirmed that the level of uplifts in control subjects exceeded that of the two depressed groups, which did not differ from one another. The repeated measures analysis in the two depressed groups revealed that neither the frequency nor the severity of uplifts varied over the course of the antidepressant treatment. Unlike the variations of hassles, which were characteristic of clinical improvement in both patient groups, the perception of increased uplifts did not accompany alleviation of depressed symptoms in either patient group.

Scores on the UCLA Loneliness scale varied as a function of Affective state, $F(2,50)=38.65$, $p<.01$. Newman-Keuls multiple comparisons indicated that both major depression and dysthymia were associated with increased feelings of loneliness relative to control subjects. In addition, in the major depressive and in the dysthymic patients a modest, but significant change of loneliness was evident over the 8 weeks of testing, $F(2, 42)=5.75$, $p<.05$. Despite the statistically significant decline of loneliness, it should be underscored that the absolute decline in these scores was small, and the degree of loneliness reported after pharmacotherapy still far exceeded control values (Figure 1).

It might have been expected that symptom severity in the major depressive and dysthymic patients would be positively correlated with hassles and inversely related with uplifts. In fact, correlations between both the HAM-D and MADRS scores with hassles, uplifts, stressful life events and loneliness were generally nonsignificant in both the major depressive and dysthymic patients. Indeed, only the frequency of hassles encountered by dysthymic patients correlated significantly with MADRS scores ($r=.43$, $p<.05$). In effect, while perception of stress may be higher among the depressed than non-depressed

subjects, within the depressed groups the severity of depression was not predictive of encountered or perceived life stressors. Indeed, even though HAM-D and MADRS scores were lower in dysthymic than in the major depressive patients, the former patients reported a higher degree of hassles encountered. Interestingly, however, in dysthymic patients the change in depression scores (HAM-D or MADRS) following pharmacotherapy was highly correlated with changes in perceived hassles, as well as feelings of loneliness (Table 4). The change in depression scores, interestingly, was not related to changes in uplifts. In contrast to these findings, among the major depressive patients, the change in depression scores was inversely correlated with uplifts and directly related to feelings of loneliness, but was not significantly related to the change in hassles encountered. In effect, it seems that while hassles were reduced with the alleviation of depression, uplifts were only marginally influenced. However, the change in uplifts was related to the alleviation of symptoms among the major depressive patients, whereas change in perceived hassles was more closely aligned with alleviation of symptoms among dysthymic patients.

2.3.3. Coping Styles

Figure 2 depicts the coping scores for each of the groups at baseline and after 8 weeks of treatment. Analysis of variance revealed that at baseline both emotional expression and emotional containment varied as a function of the Affective state, $F's (2,52) = 7.83$ and 16.61 , $p's < .01$, in that both depressive groups exhibited a higher degree of emotional expression and emotional containment than did controls. Likewise, the use of blame and avoidance/denial as coping strategies tended to be used to a greater extent in both depressive groups than in the non-depressed subjects, $F's (2,52) = 7.03$ and 8.52 , $p's < .05$. Not unexpectedly, the depressed subjects exhibited fewer coping styles involving general activity, and a greater degree of passivity than the control subjects, $F(2,52) = 4.73$ and 4.69 , $p's < .05$. Neither social support seeking, religion, cognitive restructuring nor problem solving strategies differed between groups, although the composite coping styles

Table 4.

Correlations between Changes of Depression Scores (HAM-D and MADRS at Baseline and after 8 Weeks of Pharmacotherapy) and Changes in Several Stress-Related Measures in Major Depression and Dysthymia

	MDD		Dysthymia	
	HAM-D	MADRS	HAM-D	MADRS
HASSLE FREQUENCY	-.01	.11	.68*	.75*
HASSLE SEVERITY	-.14	-.03	.60*	.57*
UPLIFT FREQUENCY	-.49*	-.50*	-.20	-.11
UPLIFT INTENSITY	-.41	-.47*	.14	.20
LIFE STRESS EVENTS	.33	.55*	.38	.40
LONELINESS	.65*	.79*	.64*	.60*

* $p < 0.05$

Figure 2. Means ($\pm$ S.E.M.) of several coping measures among controls, major depressive (MDD), and dysthymic (DYS) patients at baseline (intake) and after 8 weeks of treatment.

* $p < 0.05$ relative to controls; + $p < 0.05$ relative to intake.

of emotional expression plus social support seeking, as well as emotional containment plus passivity were more frequent in the two depressed groups, $F(2,52) = 8.51$ and 12.47 , p 's $<.01$.

Analysis of coping styles in major depressive and dysthymic patients over the 8 weeks of drug treatment confirmed that alleviation of the depressive symptoms was accompanied by alterations in some of the coping styles. In particular, analysis of variance, coupled with Newman-Keuls multiple comparisons, confirmed that the use of blame, emotional expression, emotional containment and passivity declined over the 8 weeks of pharmacotherapy, $F(1,24) = 5.88$, 6.61 and 19.73 , 5.98 , p 's $<.05$, respectively. Similarly, the alleviation of depression was accompanied by a decline in the composite coping scores of emotional expression plus social support seeking, $F(1, 24) = 15.23$, $p<.01$, and emotional containment plus passivity, $F(1,24) = 15.13$, $p<.01$.

Correlations conducted between the changes of depression scores (HAM-D and MADRS) and changes of coping styles revealed that in both major depression and dysthymia, the alleviation of depressive symptoms was highly correlated with a decline of emotional containment and the composite of emotional containment plus passivity, and an increase of general activity. In addition, in the major depressive patients, the reduction of HAM-D and MADRS scores was correlated with a reduction of blame, and avoidance/denial, and an increase of problem-solving (Table 5).

2.4. Discussion

As previously reported with other antidepressants (Corona et al., 1989; Kocsis et al., 1988; Stewart et al., 1988; Hellerstein et al., 1993; Ravindran et al., 1994c), the symptoms of dysthymia, like those of major depression, were effectively alleviated with serotonergic antidepressant medication. Indeed, it was observed in the present experiment that the effectiveness of the SSRIs in dysthymic patients was as marked as that seen in the major depressive patients, although the therapeutic effects of the drug appeared somewhat

Table 5.

Correlations between Changes of Depression Scores (HAM-D and MADRS
at Baseline and After 8 Weeks of Pharmacotherapy) and changes in Several Coping Styles
in Major Depressive and Dysthymic Patients

	MDD		Dysthymia	
	HAM-D	MADRS	HAM-D	MADRS
BLAME	.38	.53*	.09	.40
EMOTIONAL EXP	.16	.29	.19	.40
EMOTIONAL CONT	.57*	.56*	.39	.64*
SOCIAL SUPPORT	.01	0	-.35	.23
RELIGION	.12	.05	.01	.37
COG. RESTRUCT	.06	.12	-.42	-.17
GENERAL ACTIVITY	-.65*	-.68*	-.44	-.43
AVOIDANCE/DENIAL	.69*	.67*	.03	-.16
PROBLEM SOLV	-.44	-.64*	-.32	0
PASSIVITY	.04	.25	.26	.43
EMOTIONAL EXP and SOC SUPPORT SEEKING	.28	.46	-.06	.48*
EMOTIONAL CONT and PASSIVITY	.40	.59*	.43	.68*
INDIVID. COPING & COG. RESTRUCT.	-.36	-.53*	-.39	-.29

* p < 0.05

earlier in the latter condition. Of course, these data must be considered as provisional given that a placebo control was not included in this particular experiment. However, as indicated earlier, in a double-blind placebo-controlled trial, we found dysthymia to be effectively treated by sertraline (Ravindran et al., 1999).

Unlike the findings from previous reports (Brown and Harris, 1989; Kanner et al., 1981; Monroe and Depue, 1991; Grant et al., 1981), in the present experiment, major life events were not appreciably greater in the depressive patients relative to controls. However, both major depression and dysthymia were associated with heightened perception of day-to-day hassles, reduced uplifts, as well as heightened feelings of loneliness. In addition to these behavioral differences, consistent with previously reported findings in major depressive patients (Beckham and Adams, 1984; Parker and Brown, 1982; Parker et al., 1986), coping styles among major depressive and dysthymic patients could be distinguished from those of controls. For instance, relative to controls, at baseline the two depressed groups more frequently employed blame, emotional expression, emotional containment, passivity and avoidance/denial, but tended to exhibit reduced general activity. Interestingly, the patient groups did not differ from controls with respect to either problem-solving or cognitive restructuring. Thus, while it is often assumed that depressed patients do not employ effective coping strategies (particularly those involving problem-solving and cognitive restructuring), the present data suggest that rather than not using these coping strategies, depressed and dysthymic patients tend to place too great a reliance on strategies which involve emotional coping styles.

Following SSRI treatment, the reduction in depressive symptoms in both patient groups was accompanied by a diminution in the perception of day-to-day stressful encounters, although these still exceeded those of non-depressed subjects. The present results confirmed, however, that uplifting events were not appreciably altered following pharmacotherapy. It is conceivable that the reduced frequency and intensity of uplifts may be a characteristic trait marker of depressive illness. In addition to the alterations of

perceived hassles, some of the coping strategies employed by major depressive and dysthymic individuals were altered with the alleviation of depressive symptoms (e.g., social support seeking, emotional containment, passivity, blame, emotional expression, and general activity). It is unclear whether these effects reflect changes in appraisal processes and consequently altered coping styles, or simply a proportionate decline in the reliance upon emotion-focused coping. Moreover, it is not clear whether the altered coping styles contributed to the alleviation of depressive affect, or were simply secondary to the decline of depressive symptoms. Whatever the case, it is significant that the profile observed in dysthymics was remarkably similar to that seen in the major depressive patients. Interestingly, however, while change of depression in dysthymic patients was predictive of emotional containment and general activity, among major depressive patients the change of depression was also predictive of blame, avoidance/denial and problem solving.

3.1. Experiment 2: Lymphocyte Subsets Associated with Major Depression and Dysthymia: Modification by Antidepressant Treatment

As discussed earlier, major depressive disorder has been associated with a suppression of various aspects of the immune response (Calabrese et al., 1987; Evans et al., 1988; Herbert and Cohen, 1993; Kiecolt-Glaser and Glaser, 1991b; O'Leary, 1990; Stein et al., 1991), including a reduction of lymphocyte proliferation in response to mitogens (Cosyns et al., 1989; Darko et al., 1986; Kronfol and House, 1984; Kronfol et al., 1982, 1983; Levy et al., 1991; Schleifer et al., 1984; Syvalahti et al., 1985), as well as reduced NK cell activity (Darko et al., 1992; Evans et al., 1992; Irwin and Gillin, 1987; Irwin et al., 1987a,b, 1990a; 1992; Maes et al., 1992b; Mohl et al., 1987; Nerozzi et al., 1989; Stein et al., 1991). Such effects were pronounced in depressed patients exhibiting melancholia (Hickie et al., 1993; Maes et al., 1995), and at least some of the immune alterations (reduced lymphocyte proliferation) were independent of age, sleep deficits, menopausal status or prior benzodiazepine treatment (Irwin et al., 1990a). In contrast to the functional immune changes, it has generally been observed that depression was not associated with variations in T cell subsets or B cells (Albrecht et al., 1985; Calabrese et al., 1987; Evans et al., 1988; Maes et al., 1989, 1994c; Sengar et al., 1982; Syvalahti et al., 1985). Moreover, neither the absolute number nor the percentage of NK cells was found to differ between major depressive patients and control subjects (Darko et al., 1988; Maes et al., 1992a, 1994c). However, hospitalized depressives displayed fewer T and B cells (Schleifer et al., 1989) and an increase of the T helper/T suppressor ratio (Darko et al., 1988).

Despite the similarity in symptoms between major depression and dysthymia, and the high prevalence of dysthymia, little information is available concerning the immunological correlates of this illness. Experiment 2 was conducted to determine whether major depression and dysthymia were associated with alterations in the numbers of several

lymphocyte subsets, including CD3 (pan T cells), CD4 (T helper cells), CD8 (T suppressor/cytotoxic cells), CD19 (pan B cells), and CD16/56 cells (NK cells). As well, this experiment was undertaken to ascertain whether any such effects would abate with treatment of the depressive disorder using an SSRI. Inasmuch as various endocrine factors (ACTH, cortisol, norepinephrine and epinephrine) may influence immune functioning (Anisman et al., 1993; Carr, 1992), and endocrine changes may be associated with either stress and/or depression (Meltzer and Lowy, 1987; Stokes and Sikes, 1987), we assessed whether these circulating immune cells would be related to plasma ACTH, cortisol, NE, or epinephrine.

3.2. Methods

3.2.1. Subjects

The subjects consisted of 20 primary dysthymic patients (10 male, 42.30 ± 2.38 years; 10 female, 38.77 ± 2.61 years), 18 patients with major depression (9 male, 37.22 ± 3.05 years; 9 female, 36.0 ± 2.77 years), and 17 normal controls (9 male, 34.33 ± 2.56 years; 8 female, 35.0 ± 2.48 years) ranging in age from 21 and 55 years. A portion of the subjects of Experiment 2 were also represented within Experiment 1. Of the 20 dysthymic patients, 11 had been diagnosed with early onset of the illness (< 21 years). The patients were obtained and diagnosed as described in Experiment 1. The HAM-D and MADRS scores for these groups both prior to pharmacotherapy and after 8 weeks of treatment, are shown in Table 6. The control subjects satisfied the same inclusion/exclusion criteria as previously described, as did the depressive patients. The major depressive, dysthymic and control subjects reported alcohol consumption of $1.58 \pm .51$, $1.05 \pm .17$ and $2.35 \pm .69$ drinks/week and coffee consumption of $2.38 \pm .65$, $2.07 \pm .50$, $2.65 \pm .41$ cups/day, respectively. Additionally, none of the subjects reported substance abuse, and control subjects were also free of any psychotropic medication.

3.2.2. Procedures

Once informed consent was obtained, demographic information was collected, and physical and psychiatric assessments were performed as described in Experiment 1. After the diagnostic interview was performed, all patients underwent a one-week placebo run-in phase. Additionally, if subjects had been treated with psychotropic medication, the run-in phase was preceded by a wash-out period, the length of which was determined by the half-life of the medication they had received (e.g. 5-week wash-out for fluoxetine, 4-week wash-out for MAO-Is, one week for most other antidepressant agents). To preclude variations of lymphocyte subsets that might be associated with diurnal variations, all testing was conducted between 0730 and 0930 hr of the last day of the wash-out period. Subjects had a catheter inserted into the antecubital fossa of their arm and asked to relax for 30 min while watching a neutral videotape. Blood was automatically withdrawn using a Dakmed ambulatory withdrawal pump and collected in an adjacent room. Blood drawn between 30 and 35 min of catheter insertion was used for determination of lymphocyte subsets, as well as cortisol, ACTH, NE and epinephrine. This procedure was repeated again 4 and 8 weeks later for determination of lymphocyte subsets. Tubes that were used for cortisol and ACTH measurements contained EDTA and trasylol, while tubes used for amine determinations contained EDTA. Plasma samples used for endocrine determinations were quickly frozen and stored at -70°C, while immune assays were performed using fresh whole blood.

The major depressive and dysthymic patients underwent an SSRI treatment program consisting of the same drug regimens as in Experiment 1. Clinical and behavioral measurements were repeated following four and eight weeks of treatment. The control subjects did not receive any form of treatment.

3.2.3. Assay Procedures

Cortisol and ACTH: These hormones were determined using standard radioimmunoassay (RIA) kits obtained from ICN (Montreal, Que). Each of the assays were

conducted in duplicate; the interassay coefficients of variability for cortisol and ACTH have been found to be 9.9% and 9.1%, respectively, while the intra-assay coefficients were 2.69% and 7.00%. In order to eliminate inter-assay variability, the hormone assays were conducted within a single run.

NE and Epinephrine: Plasma amines were determined using a slight modification of the HPLC procedure described by Seegal et al. (1986). Essentially, whole blood treated with EDTA was spun to obtain plasma. Following protein precipitation, samples were spun in a microcentrifuge (13,000 g) for 2 min, and 100 μ L of the supernatant was injected onto a C18 reverse-phase column (Chromatography Sciences Co) in a high performance liquid chromatography system (Waters, #T12-WISP) equipped with a coulometric detector (ESA). The areas of the curve of the output signals were determined using a Hewlett-Packard (HP-3390A) plotting integrator. The mobile phase used consisted of 1.3g/L Heptanesulfonic acid, 7.3 g/L triethylamine, 0.1 g/L disodium ethylene tetraacetate, and 60ml/L acetonitrile. The pH of the mobile phase was adjusted to 2.45 using phosphoric acid, and filtered through 0.22 μ m filter paper. The flow rate of the mobile phase was 1.5ml/min.

Lymphocyte subsets: Circulating populations of T cell subsets, NK cells and B cells were determined in whole blood through flow cytometry within 3 hr of blood being collected. Specifically, 100 μ L of whole blood was mixed with 20 μ L of the monoclonal antibody reagent. After incubation for 15 min in the dark at room temperature, the red cells were lysed, vortexed and centrifuged (300 x g) for 5 min, and the supernatant removed. The cells were then washed with 2 ml PBS containing 0.1% sodium azide, and again vortexed, centrifuged (300 x g) and the supernatant removed. To the pellet, 0.5 ml of fixative (0.5% paraformaldehyde in PBS) was added, vortexed immediately and allowed to fix for 30 min at 4°C. The lymphocytes were subsequently analyzed through a FACSCAN flowcytometer by two color flow cytometry using fluorescein isothiocyanate and phycoerythrin as isotopes. Labelled monoclonal antibodies against CD3, CD3/CD4,

CD3/CD8, CD19 and CD16/CD56 were obtained from Becton-Dickinson. The absolute subset counts were determined as absolute lymphocyte counts x subset percentage (x100).

3.2.4. Statistics

A repeated measures analysis of variance was conducted to determine whether lymphocyte subsets would be increased in major depressive and dysthymic patients relative to control subjects at baseline (prior to drug treatment), and whether the lymphocyte numbers would vary over the course of the 8 weeks of treatment. Newman-Keuls post hoc comparisons ($\alpha = .05$) were conducted of the simple effects to determine between group differences at the various time points. The HAM-D scores were only collected from the major depressive and dysthymic patients, thus analysis of variance with repeated measures included only the two patient groups. Multiple a priori comparisons of treatment means were performed through Newman-Keuls multiple comparisons. Several samples used for the endocrine measures were lost, and as a result the degrees of freedom involving these analyses differed from one another and from the immune measures.

In addition to the analyses of variance, Pearson Product Moment correlations were conducted of the baseline scores to determine whether correlations existed between the endocrine and immune measures within any of the groups. Based on earlier studies which were conducted in this laboratory (Griffiths et al., 1992), we expected that plasma catecholamines would be positively related to several immune measures, particularly NK cell number. Conversely, it was expected that cortisol would be inversely related to the lymphocyte numbers. Nevertheless, owing to the large number of correlations that were computed, the α level for significance was set at .01.

During the course of the experiment, 3 major depressive and 5 dysthymic patients were lost owing to noncompliance with the experimental protocol or difficulties in obtaining blood. None of these drop-outs occurred as a result of adverse reactions to the drug treatment. As the baseline scores of these subjects were consistent with the others in

their respective groups, their data were included in the correlational analyses of baseline scores (e.g., the various immune and endocrine measures), but were not included in the repeated measures analysis of variance to determine the changes in lymphocyte subsets that may have occurred over the 8 weeks of the study.

3.3. Results

3.3.1. Clinical Measures

Commensurate with earlier results from this laboratory (Ravindran et al., 1994c), SSRI treatment reduced the symptoms of depression in both the major depressive and dysthymic patients as shown by the reductions in clinical assessment scores (see Table 6). Analysis of variance of the HAM-D scores revealed a significant Affective condition x Time interaction, $F(1, 26) = 4.35$, $p < .05$. Newman-Keuls multiple comparisons confirmed that prior to drug treatment the HAM-D scores of the major depressive patients exceeded those of dysthymic patients. A significant decline of HAM-D scores was evident after SSRI treatment in both conditions (13 of 15 major depressive and 12 of 15 dysthymic patients displayed HAM-D scores less than half of the baseline score) and the two groups did not differ from one another. MADRS scores were likewise found to decline over the 8 weeks of treatment in both the depressive and dysthymic groups, $F(1,26)=192.44$, $p < .01$, which were not found to differ from one another.

3.3.2. Lymphocyte Subsets

The analyses revealed that there was no correspondence between any of the lymphocyte subsets with either age or sex. Analyses of variance revealed that with the exception of NK cells, none of the lymphocyte subsets differed as a function of the affective illness. Inasmuch as these subsets were stable over the three sampling periods, Table 7 summarizes the lymphocyte cell numbers, collapsed over the sampling periods, for the major depressive, dysthymic and control groups. In contrast to the other subsets, NK cell number varied as a function of Affective condition x Sampling period interaction, $F(4, 88)=2.99$, $p < .05$ (see Figure 3). Newman-Keuls multiple comparisons of the simple

Table 6. Mean (±S.E.M.) HAM-D and MADRS Scores at Baseline and After 8 Weeks of Pharmacotherapy in Major Depressive and Dysthymic Patients				
	<u>HAM-D</u>		<u>MADRS</u>	
	Baseline	Week 8	Baseline	Week 8
Major Depressive	23.84 ± 0.68	7.38 ± 1.18*	28.76 ± 1.47	11.23 ± 1.86*
Dysthymia	20.06 ± 0.90	7.20 ± 1.25*	26.45 ± 1.19	9.05 ± 1.32*
* p<.05 relative to Baseline scores				

Table 7.

Lymphocyte subsets (Giga/I) among Control, Major Depressive (MDD) and Dysthymic Patients

	<u>Control</u>	<u>MDD</u>	<u>Dysthymia</u>
CD3	1.14 ± .07	1.23 ± .09	1.20 ± .06
CD4	0.76 ± .06	0.85 ± .06	0.82 ± .05
CD8	0.38 ± .03	0.42 ± .05	0.38 ± .03
CD4/CD8	2.16 ± .18	2.08 ± .13	2.38 ± .16
CD19	0.18 ± .02	0.22 ± .02	0.22 ± .02

Figure 3. Mean ($\pm$ S.E.M.) absolute number of circulating NK cells among control, major depressive, and dysthymic subjects at baseline, and after 4 and 8 weeks of antidepressant treatment in the latter two groups

effects comprising this interaction ($\alpha = .05$) confirmed that at baseline (prior to drug treatment) the NK cell number among dysthymic and depressed patients exceeded that of control subjects. The former two groups did not differ from one another. Following 4 and 8 weeks of drug treatment the number of circulating NK cells in the major depressive patients declined, such that they did not differ from control values, whereas among dysthymic patients the NK cell number following 4 weeks of treatment still exceeded that of controls. A modest decline of NK cell number in dysthymics was noted after 8 weeks of drug treatment, while NK cell number increased marginally in control subjects. As a result, the difference between control and dysthymic patients at the 8-week sampling period approached, but did not reach, statistical significance ($.05 > p < .10$).

It seems that although the antidepressant medication was equally effective in alleviating the symptoms of depression in both patient groups, the NK cells were still somewhat elevated in dysthymic patients after 8 weeks of pharmacotherapy, while in the major depressive patients NK cells could not be distinguished from control values after 4 weeks of treatment. It is tempting to speculate that the elevated NK cell numbers in dysthymic patients might be a trait marker of the illness. However, in five major depressive and 10 dysthymic patients that had been maintained on drug therapy for 6 months, the NK cell numbers were comparable to those of controls ($.113 \pm .018$ and $.134 \pm .023$, respectively). Thus, it seems that with continued antidepressant medication a progressive decline of NK cell numbers was achieved in dysthymic patients, just as such an effect occurred in the depressed patients.

3.3.3. Neuroendocrine Parameters

In contrast to reports of increased cortisol and ACTH levels in depressed patients, the results of the present investigation revealed a modest, nonsignificant elevation of plasma ACTH concentrations in the two depressed groups (40.14 ± 6.68 , 41.13 ± 5.89 , and 34.16 ± 3.56 pg/ml in major depressive, dysthymic and control subjects, respectively), while cortisol levels were somewhat lower in the two depressed groups than in control

subjects (13.24 ± 1.20 , $13.56 \pm .91$, 17.51 ± 2.20 ng/ml, respectively). Interestingly, despite the absence of increased levels of cortisol in the depressive patients, among the major depressive patients cortisol levels were correlated with scores on the HAM-D ($r=.73$, $p<.01$). A similar correlation was not evident among dysthymic patients. Epinephrine concentrations were not found to differ between the depressed, dysthymic and control subjects (91.86 ± 8.27 , 85.47 ± 5.26 , 83.97 ± 8.60 pg/ml, respectively); however, plasma NE levels were reduced significantly in the major depressive subjects (351.66 ± 22.48 pg/ml) relative to controls (433.41 ± 25.62 pg/ml), while dysthymic patients exhibited intermediate levels (377.42 ± 20.85 pg/ml), $F(2, 47)=3.18$, $p=.05$. The latter finding is an unusual one, and more recent studies conducted in this laboratory did not reveal differences of plasma NE between control and depressive patients.

Pearson Product Moment coefficients were determined between baseline plasma concentrations of the endocrine measures (NE, epinephrine, cortisol and ACTH) and each of the lymphocyte subsets in the depressed, dysthymic and control conditions (see Table 8). In control subjects NE was not correlated with any of the lymphocyte subsets, although the inverse correlation between NE and NK cell number approached significance $.01 < p < .05$, whereas none of the remaining correlations were significant. Plasma NE concentrations were unrelated to any of the lymphocyte subsets in the major depressive patients, while in dysthymics the NE levels were directly related to total lymphocytes, CD4 and NK cells (p 's $<.05$). Epinephrine concentrations were positively correlated with CD4 and the CD4/CD8 ratios in the major depressive patients, while among dysthymic patients epinephrine concentrations were positively related to CD4 cells.

Concentrations of cortisol were typically inversely related to lymphocyte subsets. However, among control and dysthymic subjects none of the correlations reached statistical significance. Among the major depressive patients the inverse relationship between cortisol levels and the CD4/CD8 ratio reached significance. In these patients there was also a trend ($p<.10$) for cortisol to be inversely related to NK, CD3 and CD4 values. Finally,

Table 8. Pearson Product Moment Correlations Between Baseline Lymphocyte Subsets and Endocrine Measures Major Depressive (MDD) and Dysthymic (Dys) Patients

	<u>Epi</u>	<u>NE</u>	<u>Cortisol</u>	<u>ACTH</u>
control	.11	.31	-.17	.43
<u>Lymph</u> MDD	.24	.25	-.37	-.57*
Dys	.34	.62*	-.24	-.24
control	-.28	-.46	.28	-.41
<u>NK</u> MDD	-.07	.02	-.36	-.10
Dys	.08	.57*	-.32	-.36
Control	.17	.36	-.06	.34
<u>CD 3</u> MDD	.32	.30	-.39	-.58*
Dys	.45	.47	-.08	-.22
Control	.21	.32	-.12	.38
<u>CD4</u> MDD	.60*	.34	-.38	-.46
Dys	.55*	.56*	-.13	-.25
Control	.04	.34	.10	.19
<u>CD8</u> MDD	.03	.36	-.20	-.35
Dys	.18	.21	0	-.05
Control	.26	-.02	-.38	.31
<u>CD4/CD8</u> MDD	.59*	-.23	-.58*	.23
Dys	.21	.14	-.35	-.24
Control	.04	.13	-.38	.31
<u>CD19</u> MDD	-.09	.01	-.04	-.51
Dys	0	-.06	-.14	-.03

*p<.01

concentrations of ACTH among the major depressive patients were inversely related to the total lymphocytes and CD3 cells, while the correlation to CD19 approached statistical significance ($p < .10$). In neither dysthymic nor control subjects was ACTH found to correlate significantly with lymphocyte subsets.

3.4. Discussion

The present investigation revealed that circulating NK cells were increased among major depressive and dysthymic patients. Other investigators, however, did not find an elevation of NK cells in major depression (see review in Maes, 1995). The source for these between-laboratory differences could not be determined from the present findings, although factors such as severity and duration of illness, diurnal variations and stressor history may be fundamental in determining NK cell numbers (see later). As indicated earlier, major depression was associated with increased total T cells (Maes et al., 1992a), and this effect was related to illness severity (Maes et al., 1992a,b). In an earlier investigation, we reported that the number of CD3, CD4, CD8 and CD19 cells were increased in major depressive patients (Griffiths et al., 1992). It seems, at least within this laboratory, that depression may be associated with increased levels of several lymphocyte subsets, but that these increases occurred less consistently than did the NK cell numbers. We have, as yet, been unable to identify the factors that contributed to these differences (Ravindran et al., 1996).

Although the magnitude of immune changes is thought to be greater with increasing illness severity (Maes et al., 1992a; Weisse, 1992), in the present study the elevation of NK cells was as great in dysthymic as in major depressive patients. As will be seen later, there is reason to believe that chronic illness may be associated with marked immune alterations, which may in fact be more pronounced than those associated with major depressive illness of moderate severity (Griffiths et al., 1996).

With alleviation of depressive symptoms, normalization of NK cell number was observed. The present data, however, revealed that although both affective disorders were successfully treated with SSRIs, NK cell numbers in the major depression group were comparable to control values within 4 weeks of treatment, whereas in dysthymic patients 8 or more weeks of treatment were required before NK cell numbers reached control levels. Interestingly, in a subset of dysthymic patients, it was observed that only after 6 months of treatment did NK cell numbers fully revert to control levels. It is possible that the sluggish return to control values in dysthymic patients may be related to the chronic nature of this condition. Thus, in assessing the relationship between depression and immune status, it may be important to recognize not only the potential contribution of illness severity, but also duration of illness, and perhaps the age of onset.

In the present investigation plasma ACTH did not correlate with any of the lymphocyte subsets in control or dysthymic patients (although in the latter condition the correlations were uniformly negative). However, in depressed patients ACTH was inversely correlated with total lymphocytes and total T cells, and the correlation between ACTH and total B cells approached significance. Moreover, plasma cortisol levels were also found to be inversely correlated with the CD4/CD8 ratio in the major depressive patients, and the correlation between cortisol and both NK cells and total T cells approached significance ($p < .10$). These correlations were less pronounced in dysthymic patients, and were entirely absent in control subjects. It must be underscored, however, that endocrine levels may fluctuate dramatically over time. In the present investigation ACTH, cortisol, NE and epinephrine were determined in blood collected over a 5 min period in subjects that had been relaxed and quiescent for more than 30 min. Yet, since only a single sample was taken, the data must be considered cautiously. As well, the role of corticoids in affecting immune activity differs as a function of the specific immune compartment examined, as well as the type of T helper cell stimulated (Th1 vs Th2) (Miller et al., 1993). In addition, changes of receptor sensitivity can be expected with sustained

stimulation, and as such it is conceivable that the relative contributions of endocrines may vary as a function of illness duration (i.e., in major depressive vs dysthymic patients). In light of these considerations, it is not surprising that the correlations between the immune measures and either cortisol or ACTH varied across the different lymphocyte subsets, and that such effects also differed between major depressive and dysthymic patients.

Although NE was previously found not to be related to NK cytotoxicity in depressed patients (Irwin et al., 1991), in the present investigation NE levels among dysthymic patients were found to be highly correlated to several lymphocyte subsets including NK and CD4 cells. In contrast, among major depressive patients and controls, NE levels were not positively correlated with these lymphocyte subsets.

4.1. Experiment 3: Variations of Lymphocyte Subsets Associated with Stress in Depressive Populations

Stressful events, whether naturalistic or applied in a laboratory, may compromise immune functioning (e.g., reduced cell proliferation in response to mitogens, reduced natural killer (NK) cell activity) (Glaser et al., 1987; Irwin et al., 1986). However, laboratory stressors have reliably been shown to increase the number of circulating NK cells (Brosschot et al., 1992; Landmann et al., 1984; Marsland et al., 1995; Mills et al., 1995; Naliboff et al., 1991) and to a lesser degree CD8 cells, but typically do not affect the number of circulating CD3 or CD4 cells (Kiecolt-Glaser et al., 1992; Marsland et al., 1995).

It seems that pronounced interindividual variations exist with respect to the stressor-provoked alterations of circulating lymphocytes. The levels of circulating NK cells may be dependent upon, among other things, reactivity of the individuals being tested and their recent stressor history (Bachen et al., 1992; Benschop et al., 1993a; Brosschot et al., 1992, 1994; Manuck et al., 1991; Naliboff et al., 1995). Likewise, the effects of stressors on CD8 cells were most apparent in subjects that displayed high cardiovascular activity and catecholamine secretion in response to the stressor (Manuck et al., 1991; Marsland et al., 1995). Inasmuch as epinephrine or NE administration also induces such alterations (Crary et al., 1983; Schedlowski et al., 1993a), it is conceivable that the effects of the stressor are subserved by catecholamine release promoting changes of cell migration.

Since dysthymia and major depression were associated with increased reports of daily hassles, as well as the adoption of inappropriate coping styles (i.e., increased use of emotion-focused coping), the possibility exists, that the altered number of circulating NK cells was secondary to the increased stressor perception associated with depressive illness (Irwin, 1995). That is, since stressors may promote elevations of circulating NK cells, the enhanced stressor perception, coupled with inadequate methods of dealing with stressors

among depressed patients, may have favored heightened NK cell numbers. Indeed, it was reported that the diminished NK cytotoxicity seen in major depressive patients was also apparent in non-depressed subjects undergoing a severe life event or major difficulty (Irwin, 1995; Irwin et al., 1990c).

Given that emotionality or reactivity of subjects might influence the immunological response to mild stressors, it is conceivable that alterations of circulating lymphocyte subsets would vary in depressed and control subjects exposed to a laboratory stressor. In the present experiment we assessed whether a stressor, comprising an arithmetic challenge, would influence lymphocyte subsets, and if so, whether such an effect would vary among major depressive, dysthymic, and control subjects. Additionally, data were collected concerning the recent stress history and coping styles of subjects, to ascertain whether these factors would be related to either the basal levels of circulating lymphocytes or to the stressor-provoked immune response. In particular, as previously indicated, depressed patients often employ emotion-focused coping strategies (e.g., emotional expression, blame, emotional containment, avoidance/denial) at the expense of more adaptive coping efforts (problem-solving, cognitive restructuring). If elevated NK cells are secondary to either enhanced stress perception or to inadequate methods of dealing with stress, then it might be expected that those patients who favor emotion-focused coping most strongly, would be most apt to exhibit the increased levels of circulating NK cells.

4.2. Method

4.2.1. Subjects

The subjects consisted of 31 primary dysthymic patients (13 male, 43.46 ± 1.34 years; 18 female, 39.61 ± 2.00 years), 18 major depressive patients (nine male, 37.88 ± 4.53 years; nine female, 37.22 ± 2.93 years), and 23 normal controls (9 male, 35.11 ± 3.00 years; 14 female, 35.14 ± 2.22 years). The patients were obtained, assessed, and diagnosed by the same means as in Experiment 1, and were recruited according to identical

inclusion/exclusion criteria. The nondepressed control group comprised not only blue- and white-collar hospital and government employees, but also university students obtained through advertisements within Carleton University. In addition to the clinical interview to exclude past or present DSM-III-R axis I disorders, control subjects had BDI scores of less than four. Subjects reported alcohol consumption of less than 3.0 drinks/week, and coffee consumption of less than 3.0 cups/day, and none of the subjects reported substance abuse.

4.2.2. Procedures

Once informed consent had been obtained, demographic and routine physical assessments were performed as described in Experiment 1. Similarly, psychiatric data were obtained from the patients by means of the HAM-D, HAM-A, MADRS, and CGI. As in Experiment 1, all subjects completed the self-rating questionnaires.

Following the diagnostic interview, all patients underwent the one-week placebo run-in phase, and if the patient had been treated with psychotropic medication, then they underwent a further wash-out period equivalent to five half-lives of the medication they had received, as described in Experiment 2. On the test day, between 0800 and 1000 hr, a catheter was inserted into the antecubital fossa of the subject's arm. Subjects sat comfortably in an easy chair while watching a neutral videotape, while blood was automatically withdrawn using a Dakmed ambulatory withdrawal pump and collected in an adjacent room. The subject was left undisturbed for a 30 min period, after which a 10 min mathematical challenge was delivered through headphones (see Appendix D). This test consisted of a series of progressively more difficult mental arithmetic tasks, including adding, subtracting, multiplying and dividing numbers, sequential subtractions, and problem solving. Blood was taken within one min of catheter insertion, 30 min after insertion (i.e., at the end of the relaxation period), and immediately after the mathematical challenge. The latter two samples were used for determination of lymphocyte subsets, as

well as cortisol, ACTH, NE, and epinephrine. Tubes that were used for endocrine and amine samples were treated as described in Experiment 2.

4.2.3 Assay Procedures

The endocrine and immune assays were carried out as described in Experiment 2.

4.2.4. Statistical Analyses

A repeated measures analysis of variance was conducted to determine whether lymphocyte subsets and endocrine measures would be influenced differentially by the stressor across groups. As well, the group differences in the various behavioral measures (e.g., hassles, uplifts and coping styles) were analyzed by analysis of variance. Newman-Keuls multiple comparisons ($\alpha = .05$) were conducted of the simple effects to determine specific between-group differences. The HAM-D, HAM-A and MADRS scores were only collected from the major depressive and dysthymic patients, thus analysis of variance of these variables included only the two patient groups. Pearson Product Moment correlations were conducted of the baseline (prestress) behavioral scores, to determine whether correlations existed between these measures and the prestress lymphocyte numbers, as well as the change in lymphocytes associated with the stressor manipulation. Owing to the large number of correlations that were computed, the α level for significance was set at .01 when a priori predictions were not made concerning these correlations.

4.3. **Results**

4.3.1 Clinical and Behavioral Measures

There were no significant differences between males and females for any of the behavioral measures, and thus the data were collapsed over this variable. Table 9 shows the HAM-D, HAM-A and MADRS scores, as well as the various behavioral stress measures for each of the groups. As seen in Table 9, the HAM-D and MADRS scores were

Table 9. Behavioral Measures ($\bar{X} \pm$ S.E.M.) Among Control, Major Depressive (MDD) and Dysthymic Subjects

	Control	MDD	Dysthymia	F
HAM-D	-----	23.61 $\pm$ 0.79	20.51 $\pm$ 0.66 ^a	(1,46) = 8.66
HAM-A	-----	14.07 $\pm$ 1.07	13.74 $\pm$ 0.66	(1,46) = 0.07
MADRS	-----	28.77 $\pm$ 1.48	25.90 $\pm$ 0.85 ^a	(1,46) = 3.37
Hassle Frequency	14.39 $\pm$ 2.10	32.50 $\pm$ 4.99 *	39.60 $\pm$ 3.70 *	(2,68) = 13.87
Hassle Severity (Total)	17.91 $\pm$ 2.82	59.93 $\pm$ 13.75 *	75.57 $\pm$ 8.62 *	(2,68) = 13.28
Average Hassle Severity	1.21 $\pm$ 0.05	1.78 $\pm$ 0.12 *	1.81 $\pm$ 0.07 *	(2,68) = 19.60
Uplift Frequency	42.89 $\pm$ 5.77	21.28 $\pm$ 4.47 *	29.00 $\pm$ 4.31 *	(2,69) = 4.46
Uplift Intensity (Total)	81.39 $\pm$ 12.77	32.89 $\pm$ 8.28 *	45.10 $\pm$ 7.10 *	(2,69) = 6.39
Average Uplift Intensity	1.79 $\pm$ 0.80	1.41 $\pm$ 0.07 *	1.46 $\pm$ 0.07 *	(2,69) = 6.90
Major Life Events	30.97 $\pm$ 6.26	47.86 $\pm$ 9.06	65.62 $\pm$ 9.83 *	(2, 69) = 4.20
Loneliness	32.61 $\pm$ 1.79	54.00 $\pm$ 1.43 *	57.66 $\pm$ 1.89 *	(2,69) = 57.18
Blame	34.38 $\pm$ 1.28	45.92 $\pm$ 2.45 *	44.99 $\pm$ 1.90 *	(2,69) = 11.14
Emotional Expression	34.39 $\pm$ 1.69	46.43 $\pm$ 2.09 *	40.80 $\pm$ 1.62 *	(2,69) = 10.13
Emotional Containment	28.68 $\pm$ 2.92	52.05 $\pm$ 2.05 *	50.09 $\pm$ 1.53 *	(2,69) = 34.09
Social Support Seeking	41.00 $\pm$ 1.55	46.71 $\pm$ 2.17	41.86 $\pm$ 1.68	(2,69) = 2.54
Religion	40.49 $\pm$ 1.62	42.17 $\pm$ 1.98	42.73 $\pm$ 1.38	(2,69) = 0.55
Cognitive Restructuring	42.77 $\pm$ 1.68	47.78 $\pm$ 2.06	42.52 $\pm$ 1.82	(2,69) = 2.19
General Activity	54.19 $\pm$ 1.46	44.89 $\pm$ 2.70 *	47.90 $\pm$ 1.57	(2,69) = 6.03
Avoidance/Denial	31.96 $\pm$ 1.65	45.76 $\pm$ 2.13 *	41.80 $\pm$ 1.93 *	(2,69) = 12.52
Problem Solving	40.06 $\pm$ 2.68	43.50 $\pm$ 2.15	41.81 $\pm$ 2.12	(2,69) = 0.47
Passivity	31.58 $\pm$ 1.66	40.93 $\pm$ 2.65 *	39.30 $\pm$ 1.75 *	(2,69) = 6.12

^a p < .05 relative to MDD

* p < .05 relative to controls

significantly greater in major depressive than in dysthymic patients, while the HAM-A scores did not differ between the two groups.

With respect to the various measures of negative and positive events, the major depressive and dysthymic groups could be distinguished from control subjects, but not from one another (see Table 9). In particular, the frequency and intensity of uplifts, as well as the average intensity of each uplift were reduced in both the major depressive and dysthymic patients. Furthermore, the frequency of hassles, the total hassle severity, the average severity of each hassle, as well as feelings of loneliness were greater in the major depressive and dysthymic subjects than in controls. Finally, although we previously found that major life events did not vary as a function of the affective condition (Experiment 1), in the present investigation reports of major life events in dysthymic patients significantly exceeded those of controls, whereas in major depressives major life events did not differ significantly from non-depressed subjects.

As previously observed, the use of emotion-focused coping strategies (i.e., blame, emotional expression, emotional containment, general activity, avoidance/denial, passivity) in both major depression and dysthymia exceeded that seen in control subjects. In contrast, the groups did not differ with respect to the absolute amount of cognitive restructuring, problem solving and social support seeking. However, the proportionate efforts expended in cognitive restructuring and social support seeking (i.e., determined by calculating the individual cognitive restructuring or social support seeking scores divided by the total of all coping styles) was significantly reduced in both depressed groups relative to controls, F 's (2, 69) = 8.95 and 7.44, p 's < .05, respectively. In effect, the major depressive and dysthymic patients tended to employ emotion-focused coping efforts to a greater extent than did control subjects. These included not only efforts in which emotions were contained, but also those meant to attribute blame, and those involving venting. While depressive subjects might not have made fewer efforts at cognitive restructuring and social support seeking, the proportionate efforts in this direction were reduced in these groups.

4.3.2. Lymphocyte Subsets

Table 10 shows the baseline number for each of the circulating lymphocytes in each of the depressive groups. Analyses of the circulating lymphocytes indicated that NK cells in major depressive and dysthymic patients significantly exceeded those of control subjects, $F(2,64)=5.45$, $p < .01$. The CD8 and CD19 levels in major depressive patients were somewhat higher than in controls, but neither of these effects reached an acceptable level of significance ($.05 < p < .10$). The CD3 and CD4 levels were comparable across groups.

Irrespective of the affective condition, the number of NK cells increased appreciably following the stressor manipulation, $F(1, 64) = 42.74$, $p < .01$ (see Figure 4). The stressor produced a small, but significant increase in the number of CD8 cells, $F(1, 64) = 15.83$, $p < .01$, and reduced the CD4/CD8 ratio, $F(1, 64) 21.76$, $p < .01$, while the numbers of CD3, CD4 and CD19 cells were unaffected by the stress manipulation. In additional studies conducted in our laboratory we have consistently observed that in subjects who were not exposed to a laboratory stressor, the various lymphocyte subsets were remarkably stable over the testing session (see also Experiment 4). Thus, the changes of NK and CD8 cells were attributable to the stress manipulation rather than to the stress associated with having the catheter in place over an extended period of time. The NK differences were independent of plasma ACTH, cortisol and NE, as these endocrine measures neither varied between depressive groups, nor as a function of the stressor manipulation (see Table 11). In contrast, a small but significant increase in plasma epinephrine levels was noted following stressor exposure, $F(1, 57) = 4.43$, $p < .05$, although the interaction between Affective condition x Stressor exposure was not significant.

The finding that the NK changes following stressor exposure were comparable, irrespective of the affective condition, could not be attributed to the relatively brief duration of the stressor employed. In particular, in a follow-up study in which a lengthier and more difficult mathematical challenge was used (i.e., a 14 min challenge involving a series of

Table 10.

Absolute Number (Giga/l $\pm$ S.E.M.) of Circulating Lymphocyte Subsets
in Controls, Major Depressive (MDD) and Dysthymic Patients

	Control	MDD	Dysthymia
Lymphocyte			
NK	.083 $\pm$.008	.164 $\pm$.012*	.148 $\pm$.016*
CD3	1.071 $\pm$.063	1.302 $\pm$.092	1.116 $\pm$.039
CD4	.712 $\pm$.048	.836 $\pm$.055	.765 $\pm$.031
CD8	.365 $\pm$.023	.454 $\pm$.047	.377 $\pm$.118
CD19	.180 $\pm$.016	.241 $\pm$.023	.195 $\pm$.009

* p <.05 relative to Controls

Figure 4. Mean ($\pm$ S.E.M.) absolute number of circulating NK cells before and after a mathematical challenge in control, major depressive (MDD) and dysthymic subjects

Table 11. Levels Of Plasma NE, Epi, ACTH and Cortisol Before and After Stress
in Controls, Major Depressive (MDD) and Dysthymic Patients

	Control		MDD		Dysthymia	
	Pre stress	Post stress	Pre stress	Post stress	Pre stress	Post stress
NE (pg/ml)	431.67 ± 19.19	398.75 ± 19.70	381.67 ± 22.49	393.55 ± 24.66	415.72 ± 19.47	449.85 ± 20.64
EPI (pg/ml)	91.08 ± 7.58	95.11 ± 9.72	82.24 ± 7.91	95.27 ± 7.21	86.61 ± 3.80	90.70 ± 5.87
ACTH (pg/ml)	32.36 ± 2.95	34.82 ± 3.83	38.87 ± 6.28	38.35 ± 5.85	37.60 ± 3.69	38.05 ± 2.38
cortisol (ug/dl)	15.42 ± 1.68	14.86 ± 1.62	13.24 ± 1.22	13.22 ± 1.35	12.87 ± 0.09	13.89 ± 1.16

more difficult problems), the results observed were essentially identical to those obtained using the relatively brief stressor. Specifically, among those depressed and control subjects who had been exposed to the stressor ($n = 14$ and 5 , respectively), circulating NK cells were higher (means = $.186 \pm .02$ and $.156 \pm .05$, respectively) than among depressed and control subjects ($n = 8$ and 5) who had not been exposed to the stressor (means = $.11 \pm .02$ and $.074 \pm .01$, respectively), $F(1, 28) = 6.63$, $p < .05$.

Basal levels of circulating NK cells correlated with several behavioral characteristics. Among control subjects the basal (prestress) number of circulating NK cells was inversely related to hassle severity, $r(21) = -.51$, emotional containment, $r(21) = -.46$ and avoidance/denial, $r(21) = -.41$. The correlations among the depressed groups were clearly distinguishable from those seen in control subjects. Specifically, among the dysthymic subjects NK cells did not correlate with either hassles or any of the emotion-focused coping scores, while problem-focused coping was inversely related to NK cell number, $r(28) = -.40$. In the major depressives, NK cells were likewise inversely related to problem-focused coping, $r(16) = -.574$, while the correlations between NK cells and emotional containment and blame, were positive, $r(16) = .41$ and $.42$, respectively.

The hassle scores were not significantly related to the stressor-provoked change of NK cells in control subjects, $r(21) = -.22$. However, among depressed patients the stressor-induced change of NK cells was significantly correlated with the HAM-D scores, $r(16) = .46$, as well as several coping strategies including cognitive restructuring, $r(16) = .78$, general activity, $r(16) = .50$, problem-solving, $r(16) = .58$, and avoidance/denial, $r(16) = .55$. In effect, even though severity of depression and coping styles were not predictive of basal NK levels, depression severity was associated with the NK change induced by the laboratory stressor. Moreover, among depressed subjects who favored the use of these particular coping strategies on a day-to-day basis, the laboratory stressor tended to produce greater variations of NK cell numbers.

4.4. Discussion

Consistent with findings from Experiment 1, both major depression and dysthymia were associated with elevated levels of perceived hassles, reduced uplifts, and increased use of emotion-focused coping styles. Likewise, as was reported previously (Seidel et al., 1996), NK cells were elevated in both major depressive and dysthymic patients. Inasmuch as stressors increase circulating NK cell levels, it is conceivable that the elevated NK cells among depressive patients were related to their heightened stress perception and their reliance on emotion-focused coping. Indeed, the results of the present investigation indicated that a laboratory stressor may influence circulating NK cells, but had little effect on other lymphocyte subsets. However, it appeared that in major depressive patients the NK cell number was highest among those patients that reported the greatest use of emotion-focused coping strategies (e.g., emotional containment and blame). Conversely, the use of problem-focused coping, which likely reflects a more effective coping strategy, was inversely related to circulating NK cells in both the major depressive and dysthymic patients. Thus, it appears that although depressive illness is accompanied by increased stress perception and reliance on emotion-focused coping, only the latter is predictive of NK cell number.

It has been reported that the increase of circulating NK cells induced by mental stress in healthy subjects was antagonized by the β -adrenergic antagonist, propranolol, (Benschop et al., 1994). Since epinephrine reduces the adhesion of NK cells to endothelium, it was suggested that the increase of circulating NK cells following stress or β -adrenergic activation may involve catecholamine-induced recruitment of immunocompetent cells from storage sites (Benschop et al., 1993b). Given that stressors have frequently been shown to influence plasma catecholamines, it is tempting to speculate that the NK changes in the present experiment might involve such a mechanism. However, the stressor procedures employed in the present study produced only a modest change of plasma epinephrine, and did not affect NE levels. Parenthetically, although stressors

typically increase plasma catecholamines and cortisol, these effects tend to be less pronounced during the a.m. than during the p.m. (Kirschbaum et al., 1995). Thus the lack of a stressor-provoked a.m. endocrine change in the present investigation was not particularly unusual. Moreover, the fact that the NK alterations occurred in the absence of cortisol or ACTH variations, provisionally suggests that the stressor-induced variations of circulating NK cells were probably independent of these endocrine changes.

It might have been expected that relative to controls, depressed individuals would be more reactive to stressors and hence would have shown a greater increase of NK cells following the stress manipulation. Using two levels of stress, however, we observed that the NK cell increase in depressed patients was comparable to that seen in non-depressed subjects. Inasmuch as a more potent stressor was associated with more pronounced NK alterations, as well as variations of other lymphocyte subsets in healthy volunteers (see Experiment 4), the failure to detect greater stressor related NK changes in the depressed than in non-depressed subjects could not be attributed to ceiling effects. Of course, this does not imply that depressed and non-depressed subjects were equally reactive with respect to other aspects of the immune response, nor do these data suggest that these groups are equally reactive to more intense life stressors or to minor day-to-day inconveniences. Moreover, it is possible that the nature of the stressor used in the present experiment was simply inappropriate to detect differential responses to environmental insults. In particular, more meaningful stressors, such as those involving day-to-day challenges, may have led to more pronounced changes of NK cells in the depressed subjects.

5.1. Experiment 4: Neuroendocrine Measures and Lymphocyte Subsets in Depressive Illness: Influence of a Clinical Interview Concerning Life Experiences

The variations of circulating lymphocytes associated with a laboratory challenge may be dependent on the background conditions upon which the stressor is superimposed. For instance the alteration of NK cell numbers was most profound among subjects that had recently experienced low levels of day-to-day hassles. Among these subjects the laboratory stressor also increased CD3 and HLA-DR+ cells (Benschop et al., 1993; Brosschot et al., 1992, 1994). Furthermore, as discussed earlier, the effects of a stressor on T suppressor cells and on mitogen-induced T cell proliferation were found to be particularly pronounced in subjects that exhibited the greatest catecholamine secretion in response to the stressor (Manuck et al., 1991; Marsland et al., 1995).

Like the effects of stressors, the induction of mood states may influence immune parameters (Futterman et al., 1994; Naliboff et al., 1995). It was demonstrated that recall of either disturbing or pleasurable emotional experiences influenced circulating NK cells in a comparable fashion, suggesting that either arousal or the provocation of mood changes, rather than the polarity of the mood per se, was responsible for the immune changes (Knapp et al., 1992). In contrast to the effects on circulating lymphocytes, the induction of positive or negative mood states differentially influenced mitogen-induced cell proliferation (Futterman et al., 1994).

As indicated earlier, inasmuch as depressive illness is associated with increased stressor perception, coupled with the use of inadequate coping styles (i.e., increased use of emotion-focused coping) (Experiment 1), the altered NK cell numbers observed in Experiment 2 may have been secondary to heightened stressor perception characteristic of the illness. However, since comparable increases of NK cells were evident in depressed and nondepressed subjects exposed to a mental arithmetic challenge (Experiment 3), the

basal differences in circulating NK cells in these subjects could not be attributed simply to differences in physiological reactivity. Yet, it is conceivable that while depressed patients may not be generally more reactive, the biological response to personally significant stimuli may be more intense than in non-depressed subjects. Accordingly, in Experiment 4 the effects of a mood induction procedure (comprising completion of a daily hassles or uplifts questionnaire, followed by a brief interview concerning the indicated experiences), on circulating lymphocytes and several endocrine measures, were assessed in depressed and non-depressed individuals. Additionally, in order to determine whether alterations of these measures were sensitive to graded stressor severities, a subset of non-depressed university students was evaluated following a relatively strong stressor (anticipation of an academic examination).

5.2. Method

5.2.1. Subjects

The subject population comprised 12 male (23.72 ± 1.25 years) and 18 female (27.41 ± 2.31 years of age) non-depressed controls, 20 male and 22 female depressive patients. Of the depressed subjects, 11 male and 13 female patients met the criteria for dysthymia (38.94 ± 1.30 and 41.27 ± 1.44 years of age), and 9 male and 9 female patients were diagnosed as major depressive (39.21 ± 1.38 and 38.80 ± 1.28 years of age). Patients were amassed and assessed in the same manner as described in Experiment 1. One minor alteration regarding diagnosis was the use of the DSM-IV criteria. The fourth edition of the DSM did not change the diagnostic criteria for dysthymic disorder or for major depression. In all other regards, dysthymic and major depressive patients were subject to the same inclusion/exclusion and diagnostic criteria as described in the aforementioned experiments. The control subjects were screened using a clinical interview (M.I.N.I.: Mini-International Neuropsychiatric Interview, clinician-rated; see Appendix B) (Sheehan et al . 1992) to exclude past or present DSM-III-R/DSM-IV axis I disorders, and had BDI scores

of less than 4 with a score of 0 on the first item ("I do not feel sad") (Beck et al., 1961). Caffeine and alcohol intake was identical to that described in Experiment 3, as was the exclusion of substance abuse.

5.2.2. Procedures

Once informed consent had been obtained, the information gathered was consistent with that described in Experiment 1. Approximately 95% of both the major depressive and dysthymic patients were free of psychotropic medication upon screening. The small percentage of subjects who were being treated with medication at that time were equally distributed between groups. The run-in phase and washout regimen were carried out as described in Experiment 1.

On the test day, between 0800 and 1000 hr, subjects had a catheter inserted into the antecubital fossa of their nondominant arm. Subjects sat comfortably in an easy chair while watching a neutral videotape, while blood was automatically withdrawn using a Dakmed ambulatory withdrawal pump and collected in an adjacent room. After 30 min the videotape was terminated, subjects in the control and depressive conditions were randomly assigned to one of three experimental conditions. Subjects received either no treatment, or were required to complete either the Daily Hassles or the Daily Uplifts Scale (see Appendix C). A supplementary section was added to each scale to elicit a description of the subject's mood in response to their reported experiences: "Of the items you've listed as irritating or stressful (that make you feel good) choose the most troubling (uplifting), and describe it in more detail, by including (a) the people, places, events involved, and (b) the feelings which were evoked within you, such as sadness, anger, loneliness, rejection, anxiety (happiness, comfort, acceptance, excitement)". After 12-15 min, the time to complete the scale in the latter two groups, the interviewer entered the room and conducted a 7-min semi-structured interview concerning the responses they provided on the Hassles or Uplifts questionnaire (e.g., "Of the various experiences you have listed, which did you find the most stressful

(pleasant)?”, “Tell me about these other stresses (or uplifts) that you have reported.”). The nontreated subjects neither completed the inventory nor were interviewed, but instead continued to watch the neutral videotape. During the course of the experiment, blood samples were collected immediately following catheter insertion, 30 min after catheterization (i.e., while the subject continued to watch the video), 12 min after the subject started the inventory, and immediately after the interview. Blood was taken at equivalent time points from the no-stress subjects.

It was of interest to determine whether the profile of circulating lymphocytes associated with a laboratory challenge would be comparable to that seen following a naturalistic stressor, particularly one considered to be more severe than the simple laboratory challenge. Thus, in the present experiment blood samples were also collected from a subset of non-depressed university students prior to an academic examination. The seven control subjects obtained from Carleton University (5 female and 2 male; mean age 20.28 ± 0.28) consented to donate blood prior to their final examination in Introductory Psychology. This exam, which was the last scheduled during the final exam period, was written at 0900 hr and subjects had a single blood sample drawn between 0815 - 0830 hr.

Samples used for endocrine and lymphocyte subset determinations were processed and subsequently assayed as described in Experiment 2.

5.2.3. Statistical Analyses

Analyses of variance with repeated measures were performed to assess the effects of the stressor manipulation on the various lymphocyte subsets. Comparisons between individual means were conducted by Newman-Keuls multiple comparisons ($\alpha = .05$). Since the number of subjects in each of the depressive conditions was relatively small, the depressive subgroups were pooled to assess the effects of the stressor manipulation. As will be seen shortly, the effects of the stressor were comparable in depressive and non-

depressive subjects, and inspection of the data indicated that the depressive subgroups could not be distinguished from one another.

Endocrine levels and lymphocyte subsets were determined for the seven nondepressed subjects who provided blood samples prior to their final examination. Because these samples were taken without a 30 minute relaxation period following needle insertion (as in the case of the laboratory stressor), comparisons of the endocrine and immune measures were made relative to the laboratory blood samples taken immediately following catheter insertion. Repeated measures analyses of variance were performed to compare the endocrine measures and lymphocyte subsets between the relaxation period prior to the inventory completion, following the laboratory interview, and immediately prior to the academic examination.

5.3. Results

5.3.1. Clinical and Behavioral Measures

As shown in Table 12, the HAM-D scores (17-item) among the major depressive patients significantly exceeded those of the dysthymic patients $F(1, 41) = 5.66, p < .05$. It ought to be noted that the relatively small difference of HAM-D scores between the major depressive and dysthymic patients likely stemmed from the particular procedures employed. Specifically, only those dysthymic patients were included who had a score of 15 or higher on the 17-item HAM-D. This rigorous criterion ensured the exclusion of mild or borderline cases of dysthymia. In addition, the major depressive group consisted of outpatients, whose depression was of moderate severity. Consistent with our previously observed results, relative to control subjects both depressive groups reported greater hassles, greater use of emotion-focused coping strategies, and reduced perception of uplifts. However, the major depressive and dysthymic patients could not be distinguished from one another on any of these dimensions.

Table 12.
Mean HAM-D (17-item) Scores ($\pm$ S.E.M.) Among Major Depressive (MDD) and Dysthymic SUBJECTS

	MDD	Dysthymia	F
HAM-D	21.26 $\pm$ 0.97	18.00 $\pm$.94 ^a	(1,41) = 5.66

^a p<.05 relative to MDD

5.3.2. Lymphocyte Subsets and Neuroendocrine Measures

Table 13 shows the circulating lymphocyte subsets at baseline (relaxation) for each of the groups. While monocytes and granulocytes were not found to differ across groups, the total number of circulating lymphocytes varied as a function of the affective condition, $F(2, 69) = 3.33$, $p < .05$. Multiple comparisons indicated that the lymphocytes among dysthymic patients were reduced relative to control subjects, but a similar reduction was not evident in the major depressive patients. Likewise, CD8 cells were reduced as a function of Affective condition, $F(2, 69) = 4.34$, $p < .05$. The multiple comparisons indicated that this effect was attributable to lower CD8 cells in dysthymic than control subjects, while in major depressive patients CD8 cells were comparable to those of controls. The analyses of variance also confirmed the previous finding that NK cells varied as a function of the depressive subtype, $F(2, 69) = 3.54$, $p < .05$, and as a function of the interaction between Affective condition and Sex, $F(2, 66) = 3.39$, $p < .05$. The multiple comparisons indicated that among the two depressive groups the NK cell number was greater among male than female patients. A comparable sex difference was not evident among the non-depressed subjects. The multiple comparisons also revealed that NK cells were elevated in major depressive patients relative to controls, while the NK cells in dysthymic patients were intermediate between the major depressive and control subjects, differing significantly from neither group.

Owing to the small number of major depressive and dysthymic patients, coupled with the fact that the stressor manipulation provoked comparable immune changes in the depressive subtypes (among major depressives NK cells increased by 0.87 giga/l from baseline to the post-interview period, while among dysthymics the increase was 0.88 giga/l), the data were pooled to assess the effects of the stressor manipulations. The analyses of variance indicated that the interview did not affect either CD3, CD4, CD8 or CD19 cells. In contrast, NK cells varied as a function of the interaction between the Interview condition x Time of Blood sampling, $F(4, 130) = 5.24$, $p < .05$. As seen in

Table 13. Mean Absolute Number (Giga/L $\pm$ S.E.M.) of Circulating Lymphocyte Subtypes in Controls, Major Depressive (MDD) and Dysthymic Patients

	CONTROL	MDD	DYSTHYMIA
Lymphocyte	1.516 $\pm$.065	1.665 $\pm$.108	1.328 $\pm$.075*
Monocyte	0.361 $\pm$.020	0.348 $\pm$.022	0.357 $\pm$.027
Granulocyte	3.152 $\pm$.189	3.465 $\pm$.239	3.482 $\pm$.246
NK	0.081 $\pm$.007	0.122 $\pm$.017*	0.096 $\pm$.010
CD3	1.173 $\pm$.054	1.210 $\pm$.099	0.982 $\pm$.055
CD4	0.755 $\pm$.037	0.816 $\pm$.067	0.677 $\pm$.045
CD8	0.411 $\pm$.024	0.396 $\pm$.037	0.305 $\pm$.027
CD19	0.207 $\pm$.015	0.236 $\pm$.015	0.191 $\pm$.023

* p <.05 relative to Controls

Figure 5 and confirmed by Newman-Keuls multiple comparisons, in the absence of the experimental manipulation the number of NK cells remained relatively stable over the 3 sampling periods in both control subjects and depressed patients. Among control subjects, completing the questionnaire did not significantly affect circulating NK cells. Likewise, among the depressed subjects the Hassles questionnaire was without effect, but completing the Uplifts questionnaire resulted in a significant elevation of the number of circulating NK cells. Interestingly, in both the control and depressed subjects the number of NK cells increased significantly following the interview, irrespective of its content (i.e., whether Hassles or Uplifts were the focus). Although NK cell number was found to differ between male and female depressed subjects, the stress interview provoked comparable increases of NK cells in both conditions (male and female depressed subjects showed NK cell increases of $.079 \pm .026$ and $.087 \pm .012$, respectively). In contrast to the circulating NK cells, the interview did not significantly influence either plasma cortisol, ACTH or NE. In fact, the variations associated with the stressor were less than 15% in each instance. Thus, it is likely that the NK cell changes were independent of these endocrine factors.

Table 14 shows the lymphocyte subsets and the endocrine measures for the seven nondepressed subjects from whom blood was taken during a quiescent period (baseline), following the stress interview, and prior to the academic examination. With the exception of circulating NK cells, in these subjects the interview neither affected circulating lymphocytes nor the endocrine measures. In contrast, anticipation of the academic examination was associated with an increase of cortisol, ACTH and NE, as well as each of the lymphocyte subsets.

5.4. Discussion

As previously observed in studies that evaluated the immunological consequences of laboratory stressors (Bachen et al., 1992; Benschop et al., 1993a, 1994; Brosschot et al., 1992, 1994; Kiecolt-Glaser et al., 1992; Landmann et al., 1984; Manuck et al., 1991;

Figure 5. Mean ($\pm$ S.E.M.) absolute circulating NK cells in control (left-hand panel) and depressive subjects (right-hand panel) who completed a questionnaire concerning recently experienced hassles or uplifts and were then interviewed by a psychiatrist concerning these experiences. Blood samples were taken prior to any treatment, following the questionnaire being completed, and immediately following the interview. 'No-treatment' subjects had blood taken at equivalent time points, but did not receive the questionnaire or undergo the interview.

Table 14.

Mean Concentrations of Circulating Lymphocyte Subsets (Giga/l), Plasma Cortisol (ng/ml), ACTH (pg/ml), and Norepinephrine (NE; pg/ml) at Baseline, Following the Stress Interview, and Immediately Prior to an Academic Examination

	BASELINE	INTERVIEW	EXAMINATION	F(2, 12)
NK	0.084 ± .016	0.126 ± .034	0.246 ± .067	6.09*
CD3	1.250 ± .074	1.191 ± .034	1.799 ± .161	14.06*
CD4	0.767 ± .052	0.739 ± .038	1.036 ± .113	9.38*
CD8	0.433 ± .024	0.430 ± .027	0.637 ± .077	7.73*
CD19	0.273 ± .041	0.244 ± .036	0.343 ± .049	7.64*
CORTISOL	19.23 ± 1.20	20.22 ± 2.54	25.22 ± 3.61	6.12*
ACTH	45.14 ± 5.88	41.65 ± 3.02	72.58 ± 11.89	6.41*
NE	389.80 ± 21.20	387.91 ± 46.29	476.27 ± 23.01	3.89*

*p <.05

Mills et al., 1995; Naliboff et al., 1991), as well as studies that assessed the impact of a mood induction procedure (Futterman et al., 1992, 1994; Knapp et al., 1992; Naliboff et al., 1995; Strauman et al., 1993), in the present investigation the interview procedure was found to increase circulating NK cells, and to a lesser extent influenced the number of CD8 cells. In contrast, neither CD3, CD4 nor CD19 cells were influenced by the experimental manipulation.

In contrast to the interview procedure, which did not influence plasma endocrines, the stress associated with anticipation of an academic examination provoked an increase of plasma cortisol, ACTH and NE levels in nondepressed subjects. These findings are consistent with the supposition that anticipation of the examination may have been a more potent stressor than the interview. Indeed, the extent of the NK cell change prior to the examination was more pronounced than that observed following the interview. Moreover, unlike the interview, the more severe stressor was also associated with increases of the other lymphocyte subsets. These findings support the contention that circulating lymphocytes are sensitive to graded differences in stressor severity. As such, if the interview regarding life experiences had been more stressful for depressed than for control subjects, then the NK cell variations would have been more pronounced among the depressed patients, and increases of other lymphocyte subsets might have been expected as well. However, the effects provoked by the interview were limited to alterations of NK cells, and these were comparable between the depressed and control subjects. These data also suggest that the absence of more pronounced NK changes in depressed subjects could not be attributed to ceiling effects precluding such an outcome. After all, the more intense stress of the academic examination yielded an increase of NK cells among controls which exceeded that produced by the interview. These data, coupled with the earlier finding that a mathematical challenge provoked comparable NK increases in depressed and non-depressed subjects (Experiment 3), suggest that the elevation of NK cells characteristic of depressed patients, is not due to greater stress reactivity. Of course, this should not be

taken to suggest that depressed and non-depressed subjects are equally reactive with respect to other aspects of the immune response, nor should they be taken to imply that these groups are equally reactive to more intense life stressors. Certainly it is conceivable that the more profound and protracted stress ordinarily associated with depression (irrespective of whether these stresses are antecedents or consequences of the illness) was responsible for the elevations of basal circulating NK cells.

As indicated in the preceding experiments, it was reported that NE or epinephrine administration effectively increased circulating NK cell levels (Schedlowski et al., 1993a; Crary et al., 1983). Since stressors also provoke increased plasma catecholamine levels, it was hypothesized that these amines might be responsible for the altered NK cells associated with a stressor (Kiecolt-Glaser et al., 1992; Schedlowski et al., 1993b). In accordance with this position, it was demonstrated that the increase of circulating NK cells induced by a laboratory stressor in healthy subjects was attenuated by pretreatment with the β -adrenergic antagonist, propranolol, (Benschop et al., 1993b, 1994). The variations of NK cells associated with the interview procedure in the present experiment were not accompanied by alterations of plasma NE levels. Thus, it is unlikely that this amine was solely responsible for the lymphocyte changes. Likewise, although changes in endocrine levels have been associated with immune alterations, as in Experiment 3, the laboratory stressors in the present experiment did not induce variations of either cortisol or ACTH. Accordingly, the contribution of these hormones to the altered NK cells is questionable. Although stressors are typically associated with increased plasma cortisol and ACTH, the lack of such an effect was not particularly surprising. As will be discussed later, in part this may stem from the nature of the procedures employed (i.e., blood being sampled in a somewhat stressful, yet contrived, laboratory setting, and involving blood taken at only a single time point), and alternative methods, including salivary sampling may have provided a more realistic reflection of the stressor effects. Parenthetically, in addition to the circadian variations with respect to stressor-induced cortisol levels, it has also been reported that the effectiveness of

academic examinations in affecting cortisol and ACTH levels varied with circadian and circannual factors (Malarkey et al., 1995). Yet, the finding that the more profound stress of the academic examination yielded marked increases of plasma NE, cortisol and ACTH, and also provoked a greater rise of circulating NK cells, is consistent with a role for one or more of these endocrine factors in promoting the NK alterations. Of course, an academic examination may be associated with a variety of other endocrine and behavioral alterations that may have influenced circulating lymphocytes.

One final issue warrants some discussion. Specifically, the diagnostic interview is the basic technique employed in clinical work. In psychiatry, it also plays an equally important therapeutic role in that the patient's unburdening of concerns often contributes to the relief experienced. However, it is recognized that the psychiatric interview may be perceived as being stressful, likely owing to the recall of painful events. Despite its value as a diagnostic and therapeutic tool, there are few systematic investigations concerning the effects of the interview on emotions experienced by the subject. The studies which have addressed this issue have focused primarily on patient satisfaction and compliance (Comstock et al., 1982; Cox et al., 1981, 1988; Inui et al., 1982), although it has been reported that active fact-oriented interviews (particularly with request for self-disclosure) elicited emotions comparable to active feeling-oriented techniques, including psychotherapy (Cox et al., 1981, 1988). In the main, the emotions engendered by a psychiatric interview were based on subjective reports and non-verbal behavioral cues, and no attention was devoted to the physiological correlates of emotional arousal (Inui et al., 1982). The clinical interview used in the present investigation was a relatively brief one, and certainly did not encroach on personal feelings and experiences to the extent that a more elaborate psychiatric interview would have. The stressfulness of the interview was evidently limited, having no effect on cortisol, ACTH or NE, although increasing circulating NK cells. Given that these effects were comparable in depressed and control subjects, it seems that physiologically.

the effects of the clinical interview were no more profound in the depressed than in the non-depressed subjects.

6.1. Experiment 5: Behavioral Correlates of Major Depression and Dysthymia With Typical or Atypical Features

Although stressful experiences and/or perceptions and the use of inappropriate coping strategies have been associated with major depressive disorder (Brown and Harris, 1989; Monroe and Depue, 1991), far less information exists with respect to dysthymia or to depressive illness involving atypical features. The purpose of this experiment was to explore several stressor-related behavioral characteristics (e.g., perception of major and minor stress events, coping styles, as well as uplifts and quality of life) of atypical and typical subgroups of major depressive and dysthymic patients.

It will be recalled that the atypical depressive profile comprises mood reactivity, coupled with reversed neurovegetative symptoms (hyperphagia, significant weight gain, hypersomnia, extreme fatigue), and persistent rejection sensitivity which is not restricted to episodes of depression (American Psychiatric Association, 1994; Quitkin et al., 1988, 1991). Relative to major depressive disorder, atypical depression tends to occur at a younger age (see Beckham et al., 1995), may persist for longer periods, and has a greater ratio of females to males (Asnis et al., 1995). Atypical depression also precludes melancholia (American Psychiatric Association, 1994), and tends to be less severe than typical depression, although severity among inpatients was reported to be comparable in atypical and typical depressives (Derecho et al., 1996). Like major depressive disorder, dysthymia can also be categorized into typical and atypical subgroups. In view of the distinctive characteristics of atypical depressives, the possibility exists that their stressor perception and coping styles may differ from those of typical major depressive patients. Likewise, data have not been reported concerning uplifting events in atypical patients, even though mood reactivity (mood enhancement in response to actual or potential positive events) is a fundamental feature of the illnesses. Accordingly, the present experiment also

assessed the perception of uplifts and quality of life in atypical and typical major depressive patients.

6.2. Method

6.2.1. Subjects

The number of male and female subjects in each group, as well as the age (in parentheses), HAM-D, HAM-A, MADRS and BDI scores are shown in Table 15. The experimental subjects represented consecutive outpatients obtained through referrals to the Mood Disorder Clinic (outpatients) of the Royal Ottawa Hospital, and met the inclusion/exclusion criteria. Patients in both of the major depressive groups were of moderate severities for the disorders as measured by the Clinical Global Impressions scale (CGI). Patients were assessed using the 29-item HAM-D (see Appendix B); the first 17 items were used as a measure of illness severity, while items 23-28 were used to detect atypical features. Patients diagnosed with atypical depression scored ≥ 6 on items 23-28 of the 29-item HAM-D and fulfilled the Columbia criteria for atypical depression with a score of 4 on the Atypical Depression Diagnostic Scale (ADDS) (see Appendix B). The dysthymic patients had a diagnosis of primary dysthymia, and met Akiskal's (1990) criteria for subaffective subtype. These patients were also classified as exhibiting either typical or atypical features based on items 23-28 of the 29-item HAM-D scale, and none of these patients met the criteria for concurrent major depression. Additionally, subjects with a previously diagnosed major depression or undiagnosed patients with symptoms sufficient to make a retrospective diagnosis of major depression were excluded from a diagnosis of dysthymia. During screening, subjects who showed personality difficulties sufficient to make a clinical diagnosis of a personality disorder were excluded from the study. This is not to say that these patients did not exhibit traits associated with personality difficulties, but rather that these were not of sufficient magnitude to fulfill DSM-III-R/DSM-IV criteria.

The non-depressed control subjects, were recruited and screened for psychiatric illness as described in Experiment 4.

6.2.2. Procedures

Informed consent was obtained for participation in the experiment from those subjects who met the inclusion and exclusion criteria. During the screen visit, demographic and diagnostic data gathered were the same as those described in Experiment 1. After the screen visit all patients underwent a placebo run-in period as in the preceding experiments. Following the wash-out period subjects were asked to complete several self-rating scales regarding their stress experiences and the coping styles which they employed. These included the stress and coping questionnaires described earlier, as well as an additional measure of stress perception (Perceived Stress Scale: Cohen et al., 1983), and the Battelle Quality of Life scale (Ravicki et al., 1992) (See Appendix C). The latter scale is divided into several dimensions including health perception, bed disability days, energy/vitality, cognitive functioning, alertness, work behavior, home management, social interaction and life satisfaction.

6.2.3. Statistical Analyses

Analyses of variance were performed to determine behavioral differences as a function of the affective disorder. Comparisons between individual group means were conducted by Newman-Keuls multiple comparisons ($\alpha = .05$), or by orthogonal contrasts when two groups were simultaneously compared to two other groups (e.g., comparisons of the typical vs. atypical groups, or major depressive vs. dysthymic groups). Unless otherwise indicated the behavioral scores were not found to vary as a function of either Sex or the interaction between Sex and Diagnostic category. During the course of the experiment some subjects may have failed to correctly fill out a scale. Accordingly, the degrees of freedom for the analyses varied across the different dependent measures.

6.3. Results

6.3.1. Clinical Measures

Table 15 shows the HAM-D and MADRS scores for each of the depressive subtypes, as well as the BDI scores for the control and depressive groups. The HAM-D and MADRS scores differed as a function of the Depressive subtype, $F(3, 127) = 11.79, 10.94, p's < .01$, and the BDI scores were also found to differ as a function of Diagnostic category, $F(4, 140) = 64.00, p < .01$. The Newman-Keuls multiple comparisons revealed that the scores of the major depressive patients significantly exceeded those of the three remaining depressive groups, which did not differ from one another. In contrast, HAM-A scores did not differ as a function of the diagnostic category.

6.3.2. Stress and Uplift perceptions

The scores for the stress and uplift measures are shown in Table 16. The analysis of variance indicated that major life events varied across the Affective conditions, $F(4, 187) = 3.05, p < .01$. Multiple comparisons indicated that relative to controls, neither typical depression nor typical dysthymia was associated with elevated major life stresses. In contrast, increased stressful life events were reported by both atypical groups.

The frequency and the intensity of hassles, as well as the perceived stress scores varied with Affective condition, $F(4, 183) = 14.44, 16.76, 51.77, p < .01$. Newman-Keuls multiple comparisons confirmed that these scores were elevated in each of the depressive groups relative to controls, but the extent of the perceived minor stresses did not differ as a function of the affective subtype. Likewise, the average intensity of each hassle (i.e., the summed severity of hassles divided by the total number of reported hassles) varied as a function of the illness, $F(4, 183) = 20.85, p < .01$. In addition to varying with the depressive condition, the frequency and intensity of hassles were greater in females than in males, $F(1, 183) = 4.68, 7.80, p's < .05$.

Table 15.

Mean ($\pm$ S.E.M) Age, HAM-D (17-item), MADRS, HAM-A and BDI scores in control subjects and in depressive subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
AGE M	32.29 $\pm$ 1.59 (22)	39.60 $\pm$ 3.72 (18)	39.08 $\pm$ 3.46 (11)	45.47 $\pm$ 2.94 (18)	41.30 $\pm$ 2.40 (12)
F	36.36 $\pm$ 1.58 (31)	38.70 $\pm$ 2.39 (20)	39.26 $\pm$ 2.66 (26)	39.19 $\pm$ 1.76 (17)	42.87 $\pm$ 1.75 (22)
HAMD	-----	23.97 $\pm$.66*	19.89 $\pm$.73	19.35 $\pm$.96	18.43 $\pm$.53
MADRS	-----	31.03 $\pm$ 1.32*	24.78 $\pm$ 1.99	24.41 $\pm$.90	23.5 $\pm$.72
HAMA	-----	16.85 $\pm$ 1.18	15.61 $\pm$ 1.62	14.91 $\pm$ 1.03	15.94 $\pm$.70
BDI	1.31 $\pm$.26	17.21 $\pm$ 1.49#	14.94 $\pm$.87#	13.38 $\pm$.89#	14.96 $\pm$ 1.08#

* p <.05 relative to other depressive groups

p <.05 relative to nondepressed control subjects

Table 16.

Mean ($\pm$ S.E.M.) Major Life Events, Perceived Stress, Hassle Severity, Uplift Intensity, UCLA Loneliness, and Coping Scores In Control Subjects and Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
Life Events	44.50 $\pm$ 5.33	50.33 $\pm$ 6.57	58.13 $\pm$ 7.48*	41.26 $\pm$ 7.21	66.69 $\pm$ 8.16*
Perceived Stress	18.72 $\pm$.91	35.26 $\pm$ 1.32*	36.94 $\pm$.854*	36.55 $\pm$ 1.54*	34.21 $\pm$.52*
Hassle Severity	22.06 $\pm$ 2.25	68.42 $\pm$ 8.15*	74.85 $\pm$ 5.83*	66.71 $\pm$ 6.74*	65.00 $\pm$ 6.80*
Uplift Intensity	90.38 $\pm$ 8.40	35.53 $\pm$ 5.92*	47.97 $\pm$ 6.26*	39.38 $\pm$ 5.64*	49.97 $\pm$ 6.32*
Loneliness	33.11 $\pm$ 1.18	54.54 $\pm$ 1.32*	56.38 $\pm$ 1.93*	54.72 $\pm$ 2.12*	58.39 $\pm$ 1.93*

* p<.05 relative to control subjects

Feelings of loneliness varied over the Affective conditions, $F(4, 183)=49.01$, $p < .01$, with each of the depressive groups reporting greater feelings of loneliness than controls. The four depressive conditions were not found to differ from one another. The analysis also indicated that males reported greater feelings of loneliness than did females, $F(1, 183) = 12.24$, $p < .01$.

Not surprisingly, depressed and control subjects were distinguishable on the basis of Uplift frequency and intensity, $F(4, 187) = 7.63, 11.62$, p 's $< .01$, as well as the average intensity of each uplift, $F(1,187)= 7.38$, $p < .01$. Multiple comparisons indicated that the depressed groups reported fewer uplifts and reduced uplift intensity relative to control subjects. Interestingly, both atypical subgroups showed a tendency for greater uplift intensity compared to the typical groups, but this effect did not reach statistical significance ($p = .06$). The analysis of the uplifts also revealed that females perceived a greater number of positive events than males, $F(1, 187) = 4.90$, $p < .05$, and a greater total intensity of uplifts, $F(1, 187) = 6.90$, $p < .01$.

6.3.3. Coping Styles

The coping styles of depressed patients differed from those of controls. As seen in Table 17, depressed subjects relied on emotion-focused coping strategies to a greater extent than did control subjects. Moreover, comparable coping profiles were evident across the depressive subtypes. The depressed groups reported greater use of blame, emotional expression, emotional containment, avoidance/denial, and passivity than did controls, and engaged in less general activity, F 's $(4, 188) = 11.17, 7.70, 28.81, 7.50, 6.55, 8.19$, p 's $< .01$. As well, they made fewer attempts at cognitive restructuring than did controls, $F(4, 188) = 3.38$, $p < .01$, but did not differ with respect to problem-solving, social support seeking or religious coping. Finally, analyses of the composite coping scores indicated that the depressed subjects engaged in more Emotional expression & social support seeking, Emotional containment & passivity, and less Individual coping & cognitive restructuring,

Table 17.

Mean ($\pm$ S.E.M.) coping scores in control subjects and depressive subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
Blame	35.40 $\pm$.823	45.23 $\pm$ 1.57*	45.50 $\pm$ 1.54*	45.08 $\pm$ 1.64*	44.12 $\pm$ 1.93*
Emotional Expression	35.85 $\pm$ 1.05	44.37 $\pm$ 1.45*	43.51 $\pm$ 1.44*	41.30 $\pm$ 1.99*	37.23 $\pm$ 1.40*
Emotional Containment	29.63 $\pm$ 1.80	52.00 $\pm$ 1.32*	48.02 $\pm$ 1.79*	45.47 $\pm$ 2.19*	48.14 $\pm$ 1.80*
Social Support Seeking	43.46 $\pm$ 1.17	44.62 $\pm$ 1.59	43.82 $\pm$ 1.26	43.15 $\pm$ 1.79	41.81 $\pm$ 1.54
Religion	41.34 $\pm$ 1.01	41.48 $\pm$ 1.21	41.42 $\pm$ 1.02	42.81 $\pm$ 1.54	40.67 $\pm$ 1.19
Cognitive Restructuring	47.97 $\pm$ 1.44	44.88 $\pm$ 1.52*	40.98 $\pm$ 1.64*	42.95 $\pm$ 1.94*	41.89 $\pm$ 1.63*
General Activity	54.43 $\pm$ 1.19	44.95 $\pm$ 1.62*	46.69 $\pm$ 1.67*	45.85 $\pm$ 1.63*	47.22 $\pm$ 1.30*
Avoidance/Denial	34.79 $\pm$ 1.29	44.69 $\pm$ 1.54*	41.98 $\pm$ 1.61*	43.28 $\pm$ 1.81*	43.12 $\pm$ 1.90*
Problem-Solving	43.14 $\pm$ 1.62	41.02 $\pm$ 1.74	42.66 $\pm$ 2.32	42.87 $\pm$ 1.85	40.34 $\pm$ 2.14
Passivity	33.01 $\pm$ 1.25	42.46 $\pm$ 1.71*	37.59 $\pm$ 1.41*	40.76 $\pm$ 1.87*	40.39 $\pm$ 1.81*
Emo Exp/Soc Supp	36.01 $\pm$.97	45.11 $\pm$ 1.42*	44.63 $\pm$ 1.29*	43.13 $\pm$ 1.53*	41.06 $\pm$ 1.34*
Emo Con/Passivity	32.55 $\pm$ 1.32	47.44 $\pm$ 1.24*	42.82 $\pm$ 1.57*	43.74 $\pm$ 1.56*	44.54 $\pm$ 1.71*
Ind Coping/Cog Restr	50.43 $\pm$ 1.22	43.89 $\pm$ 1.41*	44.97 $\pm$ 1.77*	44.69 $\pm$ 1.59*	44.10 $\pm$ 1.17*

*p < .05 relative to control subjects

$F(4, 188) = 10.11, 18.41, 4.75, p's < .01$. In addition, males used emotional expression and social support seeking less than did females, $F(1, 180) = 18.45, 5.36, p < .01$, and used emotional containment to a greater extent, $F(1, 180) = 4.73, p < .05$.

6.3.4. Quality of Life

In most respects the quality of life of the depressed groups was appreciably lower than that of control subjects (see Table 18). In particular, the self-reported Health of subjects varied as a function of Affective state, $F(4, 142) = 10.87, p < .01$. Each of the depressed groups perceived their health as being poorer than did controls, and the self-reported health of the typical major depressives was lower than that of the remaining depressive groups. The number of Sick Days also varied as a function of the Affective State, $F(4, 142) = 3.34, p < .05$. Orthogonal contrasts confirmed that Sick days among both major depressive groups exceeded those of the controls and of the dysthymic patients. The latter groups did not differ from one another in this respect.

Self-reported Energy varied across groups, $F(4, 142) = 56.81, p < .01$. Each of the depressed groups reported having less energy than control subjects, and the typical major depressives differed significantly from each of the remaining depressed groups. Concentration, Alertness, Social interaction, and Life Satisfaction were all found to vary across the Affective conditions, $F(4, 142) = 22.63, 20.88, 2.91, 46.34, p's < .05$. In each instance the depressive groups differed from controls, but not from one another. Finally, behaviors related to Work outside the home did not differ between the conditions, whereas Domestic work varied across the groups, $F(4, 142) = 3.52$. The multiple comparisons revealed that relative to controls and to dysthymic patients, housework was perceived less favorably among the two major depressive groups.

Table 18.

Mean ($\pm$ S.E.M.) Quality of Life Scores in Control Subjects and Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
Health (1)	4.31 $\pm$.15	2.69 $\pm$.28*#	3.29 $\pm$.12*	3.60 $\pm$.14*	3.18 $\pm$.17*
Health (2)	84.95 $\pm$ 2.89	49.46 $\pm$ 5.49*#	62.47 $\pm$ 3.10*	64.82 $\pm$ 2.75*	61.49 $\pm$ 3.79*
Sick Days	0.10 $\pm$.07	2.69 $\pm$ 1.18*	2.02 $\pm$ 1.41*	0.20 $\pm$.10	0.85 $\pm$.37
Energy	71.27 $\pm$ 2.21	20.00 $\pm$ 3.41*#	28.10 $\pm$ 2.19*	36.00 $\pm$ 2.90*	27.63 $\pm$ 2.18*
Concentration	85.86 $\pm$ 1.87	45.03 $\pm$ 7.03*	45.27 $\pm$ 2.75*	46.14 $\pm$ 3.47*	50.42 $\pm$ 4.49*
Alertness	2.96 $\pm$ 1.19	54.11 $\pm$ 9.24*	52.51 $\pm$ 3.71*	44.39 $\pm$ 3.96*	41.77 $\pm$ 5.44*
Work Behavior	27.41 $\pm$ 6.34	36.76 $\pm$ 5.80	39.53 $\pm$ 3.00	32.50 $\pm$ 4.26	30.84 $\pm$ 4.84
Domestic Work	19.13 $\pm$ 6.04	37.76 $\pm$ 4.65*	36.91 $\pm$ 2.53*	23.37 $\pm$ 3.95	30.16 $\pm$ 3.60*
Social interaction	66.03 $\pm$ 6.47	49.38 $\pm$ 4.12*	49.55 $\pm$ 3.02*	57.83 $\pm$ 3.27*	49.80 $\pm$ 3.64*
Life Satisfaction	4.49 $\pm$.21	2.39 $\pm$.29*	2.62 $\pm$.16*	2.83 $\pm$.16*	2.67 $\pm$.21*

*p < .05 relative to controls

#p < .05 relative to other depressive groups

6.4. Discussion

Despite the marked differences in the symptoms characterizing typical and atypical patients, and the fact that the present investigation involved both acute and chronic depressive illness, in most respects the stress-related behavioral correlates of the depressive subtypes could not be distinguished from one another. Relative to controls, each of the depressed groups exhibited increased perception of day-to-day stressors, reduced perception of uplifts, and greater use of emotion-focused coping styles at the expense of cognitive reappraisal/individual coping. While the diminished uplift perception was marked in each of the depressive conditions, this effect was more pronounced in the typical than in the atypical groups. Furthermore, while it is generally accepted that depressed patients report increased life stressors (Brown and Harris, 1989; Monroe and Depue, 1991), it was observed in our previous studies (e.g., Experiments 1 and 3; Ravindran et al., 1996) that the occurrence of major life events was only moderately increased in major depressive and dysthymic illness. In the present experiment, major life events were again found not to be elevated among patients presenting with typical symptoms. In contrast, the reports of major life stresses among the atypical major depressive and atypical dysthymic patients significantly exceeded those of control subjects.

Finally, it ought to be noted that while the clinical profiles of the male and female depressive patients were generally comparable, distinct gender differences were observed with respect to various behavioral measures. Females tended to have less of a pessimistic bias, as revealed by their uplift scores, and had coping styles that were characterized by greater use of both emotional expression and social support seeking, coupled with diminished emotional containment. As will be discussed in ensuing sections, it is conceivable that these male-female differences may have therapeutic implications.

7.1. Experiment 6: Circulating Lymphocyte Subsets In Major Depression And Dysthymia With Typical Or Atypical Features

As described earlier, there are limited data concerning the potential contribution of symptom profile and illness chronicity to immune status. It is certainly conceivable that specific symptoms of depression, particularly the neurovegetative features of the illness (e.g., altered sleep, appetite, food consumption, and weight), rather than dysphoric mood per se, might be responsible for the altered immune functioning. In view of the potential relationship between neurovegetative symptoms and immune status in depressed patients, in the present experiment we assessed circulating lymphocyte subsets in major depressive and dysthymic patients who exhibited either typical or atypical features. If the alterations of circulating lymphocytes associated with depression were related to neurovegetative disturbances rather than dysphoric mood, then it would be expected that variations of circulating lymphocytes would be evident in depressive and dysthymic patients presenting with typical but not with atypical features. In effect, rather than simply assessing the relationship between depressive illness and immune characteristics, it may be more appropriate to consider specific aspects of the illness, including the neurovegetative symptoms, and chronicity of illness. Thus, it may be possible to identify the specific features of the illness which might be responsible for immune alterations, just as such features may also be related to specific endocrine factors associated with the depressive profile (Ravindran et al., 1997).

7.2. Method

7.2.1. Subjects

The number of subjects, age, sex, as well as the HAM-D (17-item), HAM-A and BDI total scores for each of the groups are shown in Table 19. The patients were referrals to the Mood Disorders Clinic (outpatients) of the Royal Ottawa Hospital, who satisfied DSM-III-R/DSM-IV criteria for the respective disorders, and were free of any other current

Table 19. Mean ($\pm$ S.E.M.) Age, HAM-D (17-item), HAM-A and BDI Scores in Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
AGE M	34.37 $\pm$ 1.69 (17)	31.41 $\pm$ 3.58 (18)	40.54 $\pm$ 3.43 (11)	44.42 $\pm$ 2.27 (19)	41.35 $\pm$ 1.50 (17)
F	37.61 $\pm$ 1.60 (27)	39.00 $\pm$ 2.61 (19)	39.66 $\pm$ 2.64 (21)	40.72 $\pm$ 2.89 (27)	40.23 $\pm$ 1.82 (21)
HAM-D M	-----	23.25 $\pm$ 1.09*	20.45 $\pm$ 1.21	17.85 $\pm$ 0.85	18.64 $\pm$ 0.70
F	-----	25.05 $\pm$ 0.79*	19.81 $\pm$ 0.84	19.94 $\pm$ 1.08	18.38 $\pm$ 0.63
HAM-A M	-----	15.81 $\pm$ 1.80	14.50 $\pm$ 1.04	13.74 $\pm$ 1.07	15.11 $\pm$ 0.79
F	-----	17.92 $\pm$ 1.74	17.39 $\pm$ 1.10	16.52 $\pm$ 1.20	17.85 $\pm$ 1.18
BDI M	1.72 $\pm$ 0.57	14.75 $\pm$ 1.26#	17.20 $\pm$ 2.34#	14.28 $\pm$ 1.08#	14.85 $\pm$ 1.07#
F	1.27 $\pm$ 0.35	20.66 $\pm$ 2.40#*	14.05 $\pm$ 0.85#	13.65 $\pm$ 1.13#	14.50 $\pm$ 1.52#

* p <.05 relative to other depressive groups

p<.05 relative to nondepressed control subjects

Axis I disorders. The characteristics of the depressive subtypes, the diagnostic instruments, as well as the inclusion/exclusion criteria, were the same as those of Experiment 5. The characteristics of the non-depressed control subjects, as well as the exclusion/inclusion criteria, were the same as in the preceding experiment.

7.2.2. Procedures

Full and informed consent was obtained for participation in the experiment. During the screen visit, data gathered were the same as in Experiment 5. After the screening diagnostic interview those patients taking any psychotropic medications (amounting to fewer than 3% of patients) underwent a washout period as previously described. For all patients, irrespective of their prior psychotropic history, this was followed by a one week placebo run-in period.

While subjects sat in a comfortable easy chair, a catheter was inserted into the antecubital fossa of their arm, after which they relaxed for approximately 20 min while watching a neutral videotape. Blood was then withdrawn automatically using a Dakmed ambulatory pump over a 10 min period. In order to assess whether variations of lymphocytes associated with depression would be modified with the alleviation of symptoms, blood samples were also obtained from a portion of the patients who were enrolled in one of several drug trials. These patients received antidepressant medication for a 12-week period, followed by a 1-week washout period (as described earlier), after which morning blood samples were obtained as described. As the medications differed across subjects, depending on the drug trial (e.g., sertraline, moclobemide, nefazodone, paroxetine, venlafaxine), in the present investigation subjects were subdivided into those that exhibited a positive drug response and those that did not. A post-treatment 17-item HAM-D score of less than 10, with more than a 50% drop over 12 weeks, was considered a positive response.

7.2.3. Assay Procedures

The assay procedures were the same as in Experiment 2.

7.2.4. Statistical Analyses

Analyses of variance were performed to determine differences between the depressive groups with respect to depression scores. Although age was not found to vary significantly as a function of Affective condition, the control subjects were somewhat younger than the depressed patients. Accordingly, analyses of covariance, with age as the covariate, were conducted to assess whether the lymphocyte subsets varied across the Affective conditions. Comparisons between individual means were conducted by Newman-Keuls multiple comparisons ($\alpha=.05$) or by orthogonal contrasts when two groups were simultaneously compared to two other groups (e.g., comparisons of the typical vs. the atypical groups, or major depressive vs. dysthymic groups).

7.3. **Results**

7.3.1. Clinical Measures

The analysis of variance indicated that the HAM-D scores differed as a function of the Depressive subtype, $F(3, 145) = 17.74, p < .01$. Subsequent Newman-Keuls multiple comparisons revealed that, as in the preceding experiment, the HAM-D scores of the major depressive patients significantly exceeded those of the three remaining depressive groups, which did not differ from one another. HAM-A scores did not vary across the depressive groups; however, the anxiety scores of females significantly exceeded those of males, $F(3, 145) = 7.50, p < .01$. The analysis of variance also revealed that BDI scores varied across groups, $F(4, 187) = 50.30, p < .01$, and the interaction between Depressive Illness $\times$ Sex was also significant, $F(1, 187) = 3.15, p < .01$. The multiple comparisons of the simple effects of this interaction indicated that BDI scores among depressed patients exceeded those of the control subjects. As well, among females, the BDI scores in the major

depressive subjects exceeded those of the remaining depressive groups, while no such effect was evident among males.

7.3.2. Lymphocyte subsets

Analyses of covariance, with age as the covariate, indicated that there were no differences among the depressive groups or as a function of sex, for either CD3, CD4, CD8, CD19 cells, or for the CD4/CD8 ratio (see Table 20). While we previously observed that CD8 cells were reduced in dysthymic patients (Ravindran et al., 1996), this was not the case in the present experiment. In contrast to these cell subsets, the number of NK cells varied as a function of the Affective illness, $F(4, 187) = 4.63$, $p < .01$, while Age had no influence on the NK values. Newman-Keuls multiple comparisons revealed that the number of circulating NK cells in the typical major depressives, exceeded that of the remaining groups, while NK cells of the atypical major depressive and typical dysthymic patients exceeded those of the control subjects. In addition, the analysis of variance revealed that circulating NK cells in males exceeded those in females, $F(1, 187) = 4.73$, $p < .05$. Although the interaction between Affective Illness and Sex was not statistically significant, Newman-Keuls multiple comparisons of the simple effects comprising this interaction were conducted since a priori predictions had been made in this respect. In particular, we previously observed that circulating NK cells in major depressive and dysthymic males exceeded those of females (Ravindran et al., 1996), thus the possibility existed that the elevated NK cells associated with illness in the present investigation would be most pronounced among males. In fact, the multiple comparisons confirmed that among typical major depressive patients, NK cells were higher in males than in females (see Table 21). Among the females, circulating NK cells were elevated only in major depressive patients relative to control subjects. Among the males, however, circulating NK cells were elevated in each of the depressive groups relative to controls. Additionally, orthogonal contrasts indicated that NK cells in the two typical groups exceeded those of the two

Table 20.Mean Absolute ($\pm$ S.E.M.) Levels of Circulating Lymphocyte Subsets (Giga/L) in Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
Total lymphocytes	1.522 $\pm$.058	1.714 $\pm$.090	1.638 $\pm$.092	1.475 $\pm$.052	1.571 $\pm$.068
NK	0.092 $\pm$.008	0.150 $\pm$.011*	0.113 $\pm$.014*	0.113 $\pm$.009*	0.109 $\pm$.010
CD3	1.175 $\pm$.057	1.268 $\pm$.073	1.231 $\pm$.080	1.084 $\pm$.044	1.169 $\pm$.058
CD4	0.796 $\pm$.036	0.822 $\pm$.045	0.803 $\pm$.053	0.749 $\pm$.033	0.789 $\pm$.044
CD8	0.389 $\pm$.020	0.437 $\pm$.035	0.431 $\pm$.035	0.338 $\pm$.021	0.391 $\pm$.027
CD4/CD8	2.242 $\pm$.132	2.078 $\pm$.115	2.060 $\pm$.148	2.695 $\pm$.263	2.234 $\pm$.159
CD19	0.194 $\pm$.014	0.237 $\pm$.020	0.230 $\pm$.017	0.197 $\pm$.015	0.221 $\pm$.015

p < .05 relative to controls

Table 21.

Mean Absolute ($\pm$ S.E.M.) Circulating NK Cells (Giga/L) in Male and Female Control Subjects and Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
Male	.083 $\pm$.013	.169 $\pm$.017*	.144 $\pm$.027*	.124 $\pm$.012*	.114 $\pm$.012*
Female	.097 $\pm$.010	.132 $\pm$.015*	.097 $\pm$.014	.099 $\pm$.014	.105 $\pm$.015

* p <.05 relative to controls

atypical groups. Moreover, the circulating NK cells in the typical and atypical major depressives exceeded those of the two dysthymic groups.

The possibility was considered that severity of illness might be related to NK cell levels. Thus, Pearson Product Moment correlations were conducted to evaluate the relationship between the 17-item HAM-D scores and circulating NK cells. Regardless of whether these correlations were conducted using the entire pool of depressed subjects, or whether independent correlations were conducted for each of the depressive subtypes, the correlations were uniformly low. Likewise, correlations between the HAM-A scores and NK cell numbers were low and nonsignificant. Furthermore, since the analyses suggested that the number of circulating NK cells might vary as a function of atypical vs. typical features, Pearson Product Moment correlations were conducted among depressed patients to determine whether a relationship existed between NK values and the specific symptoms differentiating the typical and the atypical depressive conditions (i.e., items 23-28 on the 29 item HAM-D scale: changes in weight, appetite, eating, carbohydrate craving, hypersomnia, and fatigability). While NK scores were not correlated with either hypersomnia or fatigability ($r = .12$ and $.04$), NK scores were inversely related to eating ($r = -.21$, $p < .05$), while the correlation between NK cells and weight gain approached significance ($r = .18$, $p = .10$). Additional correlational analyses conducted only among atypical major depressive and atypical dysthymic patients confirmed that NK cells were inversely related to eating ($r = -.26$, $p < .05$). Thus, these data suggest that the specific depressive characteristic associated with the elevated NK cells was, in fact, related to the decreased eating characteristic of some depressive patients, rather than the sleep disturbance or fatigability.

As indicated earlier, in a portion of the patients an analysis was conducted to determine whether pharmacotherapy influenced circulating NK cells. The patients were subdivided into those that exhibited a positive drug response and those that did not. As several different pharmacological treatments were used, and these were not equally

represented in each of the conditions, no attempt was made to compare treatment efficacy across depressive subtypes, and are provided simply for descriptive purposes. Thus, a positive treatment response was observed in 20/24 major depressive, 20/30 atypical major depressive, 13/19 typical dysthymic and 6/10 atypical dysthymic patients.

Analyses of variance of the NK scores for each of the depressive groups both prior to treatment and after 12 weeks of antidepressant medication is shown in Table 22. In general, the NK values of these patients were lower than those of the entire set of patients (contrast Table 21 and Table 22). Analysis of variance revealed that the number of circulating NK cells declined significantly over the 12-week period among subjects that showed a positive drug response, $F(1, 55) = 6.09$, $p < .01$. Among the drug responders the Affective condition $\times$ Time of Treatment interaction (before vs. after treatment) was not significant. Since a priori predictions had been made concerning this interaction, Newman-Keuls multiple comparisons were conducted to assess the variations of NK cells over the treatment period in each of the depressive groups. These comparisons confirmed that NK cells declined among the typical major depressive and the typical dysthymic patients, but not among any of the remaining groups. Finally, a significant decline of NK cells was not evident among nontreated control subjects nor among drug nonresponders. The number of nonresponders was low, and thus it is not surprising that a statistically significant effect was not obtained. However, among the nonresponders the change of NK cells appeared to be modest and more variable relative to that seen in drug responders.

7.4. Discussion

It will be recalled that while some investigators reported reduced circulating NK cells in depressive patients (Andreoli et al., 1993; Evans et al., 1992), others reported either no effect (Darko et al., 1992) or an increase of this lymphocyte subset (Seidel et al., 1996; see also Experiment 2, 3 and 4). While severity of illness may account for some of the disparate findings, it seems likely that the immune variations may be related to either the

Table 22.

Mean NK Cells (Giga/L $\pm$ S.E.M.) Prior to and Following Antidepressant Treatment in Subtypes of

Depressive Illness among both Drug Responders and Nonresponders

	Responders		Nonresponders	
	Baseline	Post-treatment	Baseline	Post-treatment
Typical MDD	.138 $\pm$.014	.107 $\pm$.013*	.132 $\pm$.032	.121 $\pm$.037
Atypical MDD	.103 $\pm$.015	.096 $\pm$.013	.108 $\pm$.034	.098 $\pm$.015
Typical dysthymic	.099 $\pm$.021	.085 $\pm$.017*	.134 $\pm$.038	.125 $\pm$.040
Atypical dysthymic	.080 $\pm$.007	.073 $\pm$.010	.084 $\pm$.029	.075 $\pm$.029
Control	.092 $\pm$.008	.096 $\pm$.009		

* p < .05 relative to baseline

chronicity of the illness or the specific symptom profiles presented. Commensurate with Experiments 2, 3 and 4, in Experiment 6 circulating NK cells were elevated in depressive patients. However, the latter effect was dependent on the characteristic features of the illness, as well as the sex of the subject. In particular, as previously reported (Maes et al., 1994a; Evans et al., 1992), circulating NK cells among male depressive patients, irrespective of the depressive subtype, were higher than in females. Among male subjects the highest NK cell levels were seen in major depressive patients, but relative to control subjects, the circulating NK cells were also elevated in each of the remaining depressive groups. It is significant, however, that in the two typical groups circulating NK cells exceeded those seen in the atypical patients. As well, NK cells in the major depressive groups were greater than in the two dysthymic conditions. In contrast to the effects seen in male depressive patients, among females the circulating NK cells were elevated in the typical major depressive patients, but not in any of the remaining conditions. The finding that the elevated circulating NK cells seen in major depressive patients were less profound in the presence of reversed neurovegetative symptoms (i.e., in atypical patients) is consistent with the proposition that the immune changes may be secondary to neurovegetative characteristics (such as those associated with reductions of food intake).

In accordance with the proposition that the elevated NK levels were related to depressive illness or symptoms, it was observed that in those subgroups in which NK cells were elevated, successful antidepressant treatment resulted in normalization of this parameter. These data suggest that the number of NK cells may represent a state, rather than a trait marker of depression. Of course, the possibility cannot be dismissed that the peripheral effects of antidepressants may have promoted redistribution of lymphocytes, but such an outcome should have provoked alterations of NK values in both drug responders and nonresponders.

8.1. Experiment 7: Endocrine And Cytokine Correlates Of Major Depression And Dysthymia With Typical Or Atypical Features

Limited data are available concerning the neuroendocrine correlates of atypical depression, although it was suggested that hypersecretion of CRH was less prominent in atypical than in typical major depression (Nemeroff, 1996). As well, desipramine-elicited plasma cortisol secretion was greater in atypical patients, suggesting a less dysfunctional noradrenergic system in this subgroup (Asnis et al., 1995; McGinn et al., 1996). Finally, in illnesses involving atypical features (e.g., bulimia, seasonal affective disorder, and chronic fatigue syndrome) variations of HPA functioning were observed, including elevated ACTH, reduced cortisol, a blunted ACTH response to CRH challenge, and absence of CRH hypersecretion characteristic of typical depression (Demitrack et al., 1991; Gold et al., 1995; Joseph-Vanderpool et al., 1991; Levitan et al., 1997).

Although major depression was accompanied by suppression of some aspects of immune functioning, it will be recalled that there were also indications of immune activation, including increased concentrations of soluble IL-2, IL-6 and transferrin receptors, as well as enhanced production of IL-1 and IL-6 from mitogen-stimulated cells (Connors and Leonard, 1998; Maes et al., 1995). It was further suggested that the immune dysfunction associated with depression may be secondary to feedback processes generated during immune activation, which served to preclude excessive immune activity (Maes et al., 1995). Inasmuch as IL-1 is a potent stimulator of CRH release (Dunn, 1990; Rivier, 1993), the possibility exists that this cytokine contributes to the HPA alterations characteristic of major depressive illness.

While not denying that severity of depression is an important element in determining some immune alterations, as discussed earlier, such effects may also be related to either illness chronicity or to the neurovegetative profile exhibited (e.g., alterations in sleep, appetite, food consumption) rather than to the dysphoric mood per se. In view of the

varied symptom profiles, and the different neuroendocrine correlates associated with subtypes of depression, it was of interest to assess circulating hormone levels (ACTH and cortisol), and mitogen-stimulated cytokine production (IL-1 β and IL-2) in major depression and dysthymia characterized by either typical or atypical features. It was previously demonstrated that heightened IL-1 β production was evident in melancholic depression (Maes, 1995), a diagnosis which precludes atypical features (American Psychiatric Association, 1994). Accordingly, the present experiment was undertaken to establish whether the cytokine variations associated with major depression of moderate severity would be more pronounced in patients exhibiting typical features than in those with atypical features. Furthermore, although it has been reported that patients with minor depression did not exhibit cytokine alterations (Maes, 1995), we included dysthymic patients in our comparison in order to determine whether typical and atypical subtypes could be differentiated on the basis of mitogen-stimulated cytokine production.

8.2. Method

8.2.1. Subjects

The age of the subjects, number of males and females (in parentheses), HAM-D, MADRS and BDI scores are shown in Table 23. A proportion of the patients were the same as those of the preceding experiment. The patients were obtained as described in Experiment 1. As described in Experiment 4, patients in the depressed groups fulfilled the DSM-III-R/DSM-IV criteria for the respective disorders, and were currently free of any other Axis I or of Axis II disorders. The diagnostic instruments were identical to those described in the preceding experiments.

The non-depressed control subjects, were amassed as previously described, and were screened, as previously described, to exclude past or present DSM-III-R/DSM-IV axis I disorders. Exclusion criteria for all subjects remained consistent with those described in Experiment 1. All subjects reported fewer than 5.0 alcoholic drinks/week, 3.0 cups of

Table 23.

Mean ($\pm$ SD) Age, HAM-D (17-item), MADRS, HAM-A and BDI Scores in Control Subjects and in Depressive Subtypes

	CONTROL	MDD	ATYP-MDD	DYSTHYMIA	ATYP-DYSTHYMIA
AGE M	34.20 $\pm$ 1.79 (11)	46.00 $\pm$ 4.08 (5)	41.10 $\pm$ 3.44 (10)	43.62 $\pm$ 4.35 (6)	38.78 $\pm$ 2.13 (6)
F	36.93 $\pm$ 2.38 (16)	38.89 $\pm$ 4.54 (9)	39.28 $\pm$ 2.60 (21)	40.50 $\pm$ 3.52 (8)	39.67 $\pm$ 3.52 (9)
HAM-D	-----	25.14 $\pm$ 1.12*	20.16 $\pm$ 0.62	18.14 $\pm$ 1.32	18.20 $\pm$ 0.73
MADRS	-----	31.12 $\pm$ 2.65*	25.62 $\pm$ 2.04	22.92 $\pm$ 1.36	23.00 $\pm$ 1.23
BDI	1.31 $\pm$ 1.89	18.30 $\pm$ 1.97	14.76 $\pm$ 1.32	14.21 $\pm$ 1.19	15.20 $\pm$ 1.33

*p <.05 relative to other depressive groups

coffee/tea or less per day, and had not used any elicit drugs during the preceding 6 months. Once the study had been fully explained to the subject, written informed consent was obtained.

8.2.2. Procedures

Symptoms of depression were measured using the standardized clinician-rated instruments previously described, as well as the patient-rated BDI. The run-in and washout periods were consistent with those of the preceding studies. Blood collection followed the protocol described in Experiment 5. Aliquots of blood pooled over a 10 min period were used as single samples for assay of the various hormones and for mitogen-stimulated cytokines. Tubes that were used for cortisol, ACTH and plasma amine determinations contained EDTA, while samples used for IL-1 β and IL-2 determinations contained sodium heparin. Following centrifugation, plasma samples used for endocrine analyses were quickly frozen and stored at -70° C.

8.2.3. Assay Procedures

Cortisol and ACTH were determined as described in Experiment 2.

To determine mitogen-stimulated IL-1 β and IL-2 production, mononuclear cells were isolated by layering whole blood onto Histopaque-1077 (Sigma) in a 1:1 dilution in a conical tube and centrifuged (400 x g) for 30 min at room temperature. The mononuclear fraction was washed in PBS containing streptomycin (x 3), and centrifuged at 400 x g for 10 min at 4° C. The supernatant was discarded, and the cells resuspended in 5.0 ml of complete RPMI (containing 10% fetal calf serum). Mononuclear cells were counted and adjusted to 1×10^6 /ml in complete RPMI. PHA was added (0, 1, 3 or 5 μ g in a volume of 100 μ l) to 1×10^6 cells in RPMI containing 10% fetal calf serum, penicillin-streptomycin (10,000 IU/l), streptomycin (10,000 μ g/l) and sodium pyruvate (0.5 ml/l). The plates were incubated for 72 hr in 5% CO₂ at 37° C, centrifuged, and the supernatant was removed and

stored at -80°C . Concentrations of IL-1 β and IL-2 were determined by ELISA, in duplicate, in 96-well flat-bottomed titer plates, using kits obtained from R & D systems.

8.2.4. Statistical Analyses

Analyses of variance were performed to determine cytokine and endocrine differences as a function of the affective disorder. Comparisons between individual group means were conducted by Newman-Keuls multiple comparisons ($\alpha = .05$), or by orthogonal contrasts when two groups were simultaneously compared to two other groups (e.g., comparisons of the typical vs. the atypical groups, or major depressive vs. dysthymic groups). Unless otherwise indicated, the scores were not found to vary as a function of either Sex, or the interaction between Sex and Diagnosis. Over the course of the experiment samples were occasionally lost due to contamination or equipment failure and as a result the degrees of freedom for the immune and endocrine analyses varied across the different dependent measures.

8.3. **Results**

8.3.1. Clinical Measures

Table 23 shows the Hamilton Depression Rating Scores (HAM-D: 17-item) and the Montgomery-Asberg Depression Rating Scale (MADRS) scores for each of the depressed groups, as well as the Beck Depression Inventory (BDI) scores for the control and depressive groups. The HAM-D and MADRS scores differed as a function of the Depressive subtype, F 's (3, 70) = 10.48, 11.63, p 's $<.01$, stemming from the scores of the major depressive patients significantly exceeding those of the remaining depressive groups. The HAM-D scores of dysthymic patients were relatively high, owing in part to the frequent presence of anxiety and sleep disturbances, and the high HAM-D weightings of these symptoms. The difference between the typical major depressive and dysthymic patients was better reflected in the lower MADRS scores of the dysthymic patients.

8.3.2. Plasma Neuroendocrine Parameters

While plasma NE and epinephrine levels did not vary as a function of the Affective Illness, levels of ACTH differed across groups, $F(4, 96) = 4.12$, $p < .01$. Post hoc tests indicated that, relative to controls, ACTH levels were elevated in atypical major depressives, while in the typical major depressives a modest, nonsignificant increase of ACTH was apparent (see Table 24). In neither of the dysthymic groups was ACTH found to differ from controls. The levels of ACTH were also found to be higher in males, $F(1, 91) = 4.75$, $p < .05$, but this effect was only marked in the atypical major depressive patients. Further, plasma cortisol levels were lower in the two atypical than in the typical depressive conditions, $F(1, 91) = 4.12$, $p < .05$.

8.3.3. Mitogen-Stimulated Cytokine Production

The concentrations of IL-1 β in the supernatants of mitogen-stimulated cells are shown in Figure 6 as a function of the affective condition. The concentrations of IL-1 β increased with the dosage of PHA, $F(2, 192) = 59.36$, $p < .01$, and varied as a function of the Affective illness, $F(4, 96) = 5.02$, $p < .01$. Newman-Keuls multiple comparisons confirmed that relative to control subjects the concentrations of IL-1 β were not affected in either the major depressive or the atypical depressive patients. In contrast, IL-1 β was significantly elevated among the dysthymic patients, irrespective of whether they exhibited typical or atypical features.

Production of IL-2 varied as a function of Affective condition x Mitogen dosage interaction, $F(4, 180) = 2.33$, $p < .05$. At the lowest mitogen dose none of the groups differed from one another; however, at the remaining two mitogen doses, IL-2 production was significantly reduced in the two dysthymic groups and in the atypical major depressive patients relative to nondepressed controls (see Figure 6). The reductions of mitogen-stimulated IL-2 were less profound in the typical major depressive patients, although a significant IL-2 reduction was observed at the highest PHA concentration.

Table 24.

Mean Absolute ($\pm$ SD) Levels of Plasma Cortisol, ACTH, Norepinephrine (NE) and Epinephrine (Epi)
in Control Subjects and Depressive Subtypes

	CONTROL	DEP	ATYP-DEP	DYSTHYMIA	ATYP-DYSTHYMIA
Cortisol (μ g/dl)	16.18 $\pm$ 5.45	16.00 $\pm$ 8.02	11.09 $\pm$ 4.88*	15.59 $\pm$ 5.30	13.05 $\pm$ 5.30
ACTH (pg/ml)	36.23 $\pm$ 15.72	42.62 $\pm$ 19.88	47.50 $\pm$ 25.17*	28.61 $\pm$ 9.06	38.10 $\pm$ 10.94
NE (pg/ml)	435.07 $\pm$ 92.30	461.52 $\pm$ 181.61	465.98 $\pm$ 85.94	387.31 $\pm$ 94.44	494.45 $\pm$ 142.29
Epi (pg/ml)	223.26 $\pm$ 109.35	190.58 $\pm$ 108.28	212.51 $\pm$ 105.11	154.39 $\pm$ 78.86	176.43 $\pm$ 50.89

* p < .05 relative to control subjects

Figure 6. Mean ($\pm$ S.E.M.) concentrations of IL-1 β (left-hand panel) and IL-2 (right-hand panel) in supernatants of lymphocytes stimulated with various doses of PHA, among control subjects and several depressive subtypes: typical major depression (Typ-MDD), atypical major depression (Atyp-MDD), typical dysthymia (Typ-Dys) and atypical dysthymia (Atyp-Dys).

Inasmuch as dysthymia is a chronic illness, often associated with early age of onset, either of these variables may have contributed to the altered IL-1 β production. Accordingly, Pearson Product Moment correlations were conducted between IL-1 β levels (at each mitogen dosage) and the age of onset and chronicity of dysthymia. In fact, both of these variables were highly correlated with the IL-1 β concentrations. Specifically, the correlations between Age of onset and IL-1 β (at the 1.0, 3.0 and 5.0 μ g doses of PHA) were -.58, -.44 and -.50, $p < .01$, respectively, while the correlations between duration of illness and IL-1 were .60, .46 and .43, p 's $< .01$, respectively. In contrast, neither age of onset nor duration of illness were significantly correlated with IL-2 levels. Furthermore, levels of IL-1 and IL-2 were unrelated to age of onset or duration of illness in major depressive patients. Likewise, in none of the groups was age itself found to be correlated with the cytokine levels.

Since cortisol has been reported to inhibit cytokine production (Maes et al., 1994d), while NE stimulates lymphocyte activity (Benschop et al., 1994), correlations were conducted between production of IL-1 β and IL-2 and plasma neuroendocrine concentrations. These analyses indicated that the correlations between IL-1 production (after 1.0, 3.0 or 5.0 μ g of PHA) and levels of cortisol, ACTH and NE were nonsignificant among both major depressive and dysthymic patients. Likewise, plasma cortisol levels were not correlated with IL-2 production. However, the production of IL-2 was highly correlated with NE concentrations in both of these patient groups. In particular, among dysthymic patients the correlations between NE levels and IL-2 production (following 1.0, 3.0 and 5.0 μ g of PHA) were .45, .57 and .54, respectively, while in the major depressive patients these correlations were .52, .43 and .50 (p 's $< .05$). Among control subjects the correlations between IL-2 production and NE were low (.03, .08 and .14).

8.4. Discussion

Summarizing briefly, it appeared that elevations of plasma ACTH were most pronounced in atypical major depression, while cortisol levels were reduced. Of course, HPA disturbances associated with depressive disorders may occur at any number of different levels, and may involve more than a single dysfunctional mechanism (Holsboer, 1995; Akil and Morano, 1995). Thus, it is premature to assume that HPA disturbances in atypical depression involve the same mechanisms as those subserving other illnesses with atypical features. As well, only morning blood samples were collected over only a few minutes, and as will be discussed, it would be advantageous to measure neuroendocrine profiles over a wider range of times and using less invasive means (e.g., salivary sampling).

Major depressive and dysthymic patients (both typical and atypical) were distinguishable from one another on the basis of their cytokine profiles. Whereas IL-1 β production was elevated in dysthymia relative to control subjects, this was not the case in the major depressive patients. Moreover, it did not appear that this effect was related to the neurovegetative characteristics of the illnesses, in that comparable effects were seen irrespective of typicality. Interestingly, the finding that the elevated levels of IL-1 β among dysthymic patients were correlated with the age of onset and illness duration, suggests that illness chronicity, rather than simply severity, may be fundamental in determining whether a relationship between cytokine production and dysthymia will be evident. Of course, it is unclear whether the IL-1 β levels were causally related or secondary to the illness. Parenthetically, although catheter insertion itself may promote local inflammation and hence increased cytokine production, the short duration of catheterization (i.e., 30 min) likely was not sufficient to elicit cytokine variations. Moreover, since the elevated IL-1 β production was only evident in dysthymic patients, it is unlikely that local inflammation contributed to the observed outcome.

In contrast to IL-1 β production in dysthymic patients, it appeared that mitogen-stimulated IL-2 was reduced in both major depressive and dysthymic patients, regardless of the presence of typical or atypical features. Interestingly, IL-2 production was highly correlated with plasma NE levels in both depressive groups, possibly reflecting characteristics of NE receptors on lymphocytes of depressive subjects. The reduced IL-2 production may be taken to suggest that this aspect of immune functioning, insofar as it may reflect T cell activation, is impaired in depression. However, as will be discussed later, IL-2 levels in the absence of measurement of sIL-2R, may not provide an adequate gauge of IL-2 functioning. Thus, it is premature at this juncture to draw a conclusion concerning the functional T cell response associated with depressive illness. In fact, as discussed earlier, the bulk of the current evidence, at least within melancholic patients, is that the illness is associated with increased IL-2 functioning (Maes, 1995).

9. GENERAL DISCUSSION

9.1. Pharmacotherapy in Dysthymia

Dysthymia has been shown to be effectively treated with tricyclic antidepressants, such as imipramine, desmethylinipramine, amitriptyline and doxepin (Corona et al., 1989; Kocsis et al., 1988; Stewart et al., 1988), although the success rate was reported to be lower than generally seen in patients with major depressive disorder (Howland, 1991). Several studies (Hellerstein et al., 1993; Ravindran et al., 1994c) have reported that SSRIs (e.g., fluoxetine) alleviated depressive symptoms in dysthymic patients. Indeed, it was observed in the present investigation that the effectiveness of the SSRIs in dysthymic patients was as marked as that seen in major depressives, although the therapeutic effects of the drug appeared somewhat earlier in the latter condition. Although Experiments 1 and 5 were conducted as open-label trials without the inclusion of a placebo group, the magnitude of the observed effects were too large to simply reflect a placebo response. Moreover, we recently observed that in a double-blind, placebo controlled trial of sertraline efficacy, symptoms of dysthymia were reduced to a significantly greater extent among drug-treated than placebo patients. Additionally, it was noted in this particular study that the drug treatment was more effective than 12 weeks of group cognitive behavioral therapy (Ravindran et al., 1999).

9.2. Stress and Coping

Although major depressive and dysthymic patients reported a somewhat greater intensity of major life events than did controls, these data were highly variable and did not consistently reach statistical significance. While it is generally accepted that depressed patients report increased life stressors (Brown and Harris, 1989; Monroe and Depue, 1991), we have noted previously that the occurrence of major life events is only moderately increased in major depressive and dysthymic illness (Ravindran et al., 1996). Interestingly, in Experiment 5 the reports of major life stresses among the atypical major depressive and

atypical dysthymic patients significantly exceeded those of control subjects. Thus, it appears that typical and atypical types of depression can be distinguished, at least to some extent, on the basis of major life stressors reported. Of course, this should not be taken to imply that such events were precursors of atypical, but not typical depression, since the relatively greater stress perception may simply have reflected the increased mood reactivity characteristic of the atypical patients.

Commensurate with earlier reports, major depressive disorder was associated with increased occurrence of day-to-day hassles (Brown and Harris, 1989; Kanner et al., 1981; Monroe and Depue, 1991; Grant et al., 1981), reduced uplifts, as well as heightened feelings of loneliness (Eisemann, 1984). The perception of uplifts in dysthymic patients was also lower than it was among control subjects. Alloy and Abramson (1988) suggested that depression may be associated not only with increased perception of stressors, but also with a decline in the perception of positive events. The present results were consistent with this view although it is unclear whether the stress profiles among the depressed patients were secondary to (symptomatic of) these affective states, or in some way etiologically related to them.

Despite the marked differences in the symptom profiles among typical and atypical patients, and the fact that Experiment 5 involved both acute and chronic depressive illness, in most respects the stress-related behavioral correlates of the depressive subtypes could not be distinguished from one another. Relative to controls, each of the depressed groups exhibited increased perception of day-to-day stressors and reduced perception of uplifts. While the diminished uplift perception was marked in each of the depressive conditions, this effect was more pronounced in the typical than in the atypical groups.

In addition to the differences in perception of stressful events (Parker and Brown, 1982; Parker et al., 1986), as previously reported in major depressive patients (Beckham and Adams, 1984), coping styles among both depressed and dysthymic patients were found to differ from those of controls. For instance, relative to controls, at baseline the two

depressed groups more frequently employed blame, emotional expression, emotional containment, passivity and avoidance/denial, but tended to exhibit reduced general activity. Interestingly, the patient groups did not differ from controls with respect to either problem-solving or cognitive restructuring. Thus, while it is often assumed that depressed patients do not employ effective coping strategies (particularly those involving problem-solving and cognitive restructuring), the present data suggest that rather than not using these coping strategies, depressed and dysthymic patients tend to place too great a reliance on strategies which involved emotional coping styles. These effects were particularly notable in typical as opposed to atypical depressive groups. It should be emphasized that the suggestion is not being offered that depressed patients were using effective cognitive styles in the same fashion as control subjects. They may well make attempts to employ these styles, but may use them ineffectively.

9.3. Effects of Antidepressant Therapy

Following treatment with the SSRI, the reductions in depressive symptoms in both major depression and dysthymia were accompanied by a diminution in the perception of day-to-day stressful encounters, although these still exceeded those of nondepressed subjects. As indicated earlier, it was reported that depressed patients do not display the optimistic biases that may be characteristic of non-depressed individuals (Alloy and Abramson, 1988; Taylor and Brown, 1988). The present results confirmed this finding in major depressive and dysthymic patients, indicating that with the alleviation of depressive symptoms, a concurrent increase in perceived uplifts was not evident. It is conceivable that the reduced frequency and intensity of uplifts may be a characteristic trait marker of depressive illness. It might be noted that cognitive therapy has been reported to be as effective as pharmacotherapy in the treatment of depression (Hollon et al., 1991; Leahy and Beck, 1988; Safran and Segal, 1990), and in combination with pharmacotherapy may be superior to pharmacotherapy alone. Furthermore, it seems that cognitive therapy may also

prolong the effectiveness of pharmacotherapy in preventing depressive relapse (Blackburn et al., 1986; Leahy and Beck, 1988). The possibility exists that while pharmacotherapy may be an effective treatment for the symptoms of depression, cognitive therapy may be required to alter perception and appraisal of positive day-to-day events. Yet, recent experiments conducted in our laboratory have indicated that unlike the effects seen in major depressive patients, cognitive therapy alone was not particularly helpful in the majority of dysthymic patients, and also was found not to enhance the effects of pharmacotherapy (Ravindran et al., 1999). Of course, these data need to be considered as highly provisional, since the trial was limited to 12 weeks, and the psychotherapy consisted of group rather than individual cognitive behavioral therapy. Further, given that anhedonia is a characteristic and persistent feature of dysthymia, it may have been beneficial to employ cognitive techniques which focused specifically on the inability of patients to experience or perceive positive events. Finally, it might be noted that the aforementioned study only assessed the acute therapeutic efficacy of the different treatment strategies. Given the high rate of depressive relapse/recurrence following cessation of treatment, it will be interesting to establish whether the combination therapy minimizes recurrence of illness relative to that seen among patients that had received only pharmacotherapy.

It is interesting that despite the absence of a significant increase of perceived uplifts, among major depressive patients the alleviation of symptoms associated with the drug treatment was correlated with an increase of uplifts, whereas a similar correlation was not apparent in dysthymic patients. Conversely, among dysthymic patients the alleviation of depressive symptoms was highly correlated with a reduction of perceived hassles, while a similar correlation was not evident in major depressive patients. That the alleviation of depression was not predictive of uplifts in dysthymic patients was surprising given that anhedonia is characteristic of this disorder. It is, however, possible that because dysthymia is a sustained disorder, in which anhedonia is deeply ingrained, these patients while being able to recognize the diminished occurrence of aversive events (hassles) may be less

attuned to variations of hedonic stimuli or events. Consequently, treatment response would not be correlated with a change in uplifts. The fact that such a correlation was apparent in major depressives suggests that in a subset of these patients the decline of depressive symptoms was accompanied by an increase of uplifts, even though the number of uplifts reported did not differ following antidepressant medication. Indeed, in approximately half the subjects (those showing the largest decline of HAM-D scores) perceived uplifts increased, while in the remaining subjects uplifts declined.

In addition to the alterations of perceived hassles, some of the coping strategies employed by depressed and dysthymic individuals were altered with the alleviation of depressive symptoms (e.g., social support seeking, emotional containment, passivity, blame and emotional expression, in conjunction with an increase in becoming involved in activities unrelated to their problems). It is unclear whether these effects reflect changes in appraisal processes and consequently altered coping styles, or simply a proportionate decline in the reliance upon emotion-focused coping. Moreover, it is not clear whether the altered coping styles gave rise to the alleviation of depressed affect or were simply a consequence of the waning of the depressed symptoms. Whatever the case, it is significant that the profile observed in dysthymic patients was remarkably similar to that seen in major depressive patients. Interestingly, however, while change of depression in dysthymic patients was related to emotional containment and general activity, among major depressive patients the change of depression was also related to blame, avoidance/denial and problem-solving.

The relationship between major depression and dysthymia is complex, but it appears that both illnesses share characteristics, such as the response to antidepressant drugs, altered perception of life stressors, and the use of inappropriate coping strategies. Despite the finding that the behavioral profiles of major depression and dysthymia can be differentiated on the basis of the frequency and/or severity of hassles, the data are commensurate with the views expressed by Akiskal (1990) that dysthymia is a subtype of

depression. Additionally, the possibility ought to be considered that the subtle, but significant differences in the perception of day-to-day experiences between major depressives and dysthymics might be useful in distinguishing between these illnesses. Finally, with the alleviation of depressive symptoms following pharmacotherapy the perception of hassles is reduced and coping styles shift away from emotion-focused styles. Interestingly, uplift perception is not enhanced. It is possible that the antidepressant medication may influence the patient's ability to cope with stressors, without influencing the cognitive appraisal associated with the anhedonic or pessimistic demeanor. According to this position, if the depression was primarily stress-related, then the drug treatment would be optimally effective. Conversely, pharmacotherapy would be less effective in alleviating depression associated with a profound anhedonia. In fact, Ravindran et al. (1997) have argued that the high rate of recurrence of depression may relate to the presence of residual symptoms, which include anhedonia.

The behavioral profiles of the male and female depressive patients were generally comparable. However, the females tended to have less of a pessimistic bias as revealed by their uplift scores, and had coping styles that were characterized by greater use of both emotional expression and social support seeking, coupled with diminished emotional containment. These differences between male and female patients, albeit subtle, may have important therapeutic implications. In particular, as previously discussed, among major depressive and dysthymic patients successfully treated with antidepressant agents (in this instance 8 weeks of SSRI treatment), the uplift perception did not increase significantly. Given that males and females exhibit different profiles with respect to the use of emotion-focused coping strategies and with respect to perceived uplifts, the possibility exists that they might also be differentially responsive to antidepressant therapy, as in the case of atypical depression (Davidson and Pelton, 1986). This gender difference may also have implications for the type and forms of psychotherapeutic interventions used in these patients. Specifically, in the development of cognitive therapies, increasing attention has

been devoted to establishing structured approaches for specific types of disorders. In a similar fashion, it might be advantageous to have therapeutic interventions tailored for varying degrees of anhedonia.

9.4. Cell subsets in depressive illnesses

As indicated earlier, inconsistent results have been reported concerning the relationship between depression and immune functioning. A meta-analysis, however, suggested that major depressive disorder is associated with reductions of immune activity, including proliferative responses to mitogens, NK cell activity, as well as numbers of lymphocyte subsets (Herbert and Cohen, 1993). Yet, it will be recalled that the reduction of NK cytotoxicity or number associated with affective disorders may be restricted to older patients or to the relatively severe melancholic form of the disorder (Weisse, 1992). The finding that NK cell numbers were increased in the present experiments was inconsistent with several earlier reports (see reviews in Herbert and Cohen, 1993; Maes, 1995). The source for the between-laboratory differences are unclear, although factors such as severity and duration of illness, diurnal variations and stressor history may be fundamental in determining NK cell numbers. It is unlikely that the findings of the present studies were spurious, however, given the persistence of the elevated NK cell number in dysthymic patients.

It is interesting that the elevation of circulating NK cells was as great in dysthymic as in major depressive patients. As indicated earlier, it has been argued that some immune alterations associated with depression (e.g., reduced NK cytotoxicity and proliferation in response to mitogens) may be related to the severity of the illness (Weisse, 1992; Maes et al., 1992a). Inasmuch as dysthymia is considered to be less severe than major depression, the present data suggest that the absolute increase of NK cell number is unrelated to depression severity, at least not in the mild to moderate range of the illness. However, it does appear that the effectiveness of antidepressant medication in promoting a return of NK

cell number to control levels may be related to the chronicity of the depressive disorder. The present data revealed that although both major depression and dysthymia were successfully treated with SSRIs, different durations of treatment were required for NK cell numbers to return to control values. In particular, NK cell numbers in the major depressive group were comparable to control values within 4 weeks of treatment, whereas in dysthymic patients 8 or more weeks of treatment were required before NK cell numbers reached control levels. In fact, only after 6 months of treatment did NK cell numbers of dysthymic patients fully recover to control levels. The differential rates at which NK cell levels returned to control values in the two depressed populations suggest that the drug treatments did not act directly on lymphocytes. Rather, it can be argued that the decline of NK cell numbers was in some fashion related to the alleviation of symptoms of depression (i.e., change in cognitions). It is possible that the sluggish return to control values in dysthymic patients may be related to the chronic nature of this condition. Thus, in assessing the relationship between depression and immune status, it may be important to recognize not only the potential contribution of illness severity, but also duration of the illness and perhaps the age of onset.

It will be recalled that depression may be associated with HPA dysfunction, and there is reason to believe that a relationship exists between immune functioning and HPA activity (Miller et al., 1993). In Experiment 2, plasma ACTH did not correlate with any of the lymphocyte subsets in control or dysthymic patients (although in the latter the correlations were uniformly negative). However, in major depressive patients ACTH was inversely correlated with total lymphocytes and pan T cells and the correlation between ACTH and pan B cells approached significance. Moreover, plasma cortisol levels were also found to be inversely correlated with the CD4/CD8 ratio in major depressives, and the correlation between cortisol and both NK cells and pan T cells approached significance ($p < .10$). These correlations were less pronounced in dysthymic patients, and were entirely absent in control subjects. It must be underscored, however, that endocrine levels may

fluctuate dramatically over time. In the present experiments ACTH, cortisol, NE and epinephrine were determined in blood collected over a 5-minute period in subjects that had been relaxed and quiescent for more than 30 minutes. Yet, since only a single sample was taken, the data must be considered cautiously. As well, the role of glucocorticoids in affecting immune activity may differ as a function of the specific immune compartment examined, as well as the type of T helper cell stimulated (Th1 vs Th2) (Miller et al., 1993). In addition, changes of receptor sensitivity can be expected with sustained stimulation, and as such it is conceivable that the relative contributions of endocrine factors may vary as a function of illness duration (i.e., in major depressive vs. dysthymic patients). In light of these considerations, it is not surprising that the correlations between the immune measures and either cortisol or ACTH varied across the different lymphocyte subsets, and that such effects also differed between major depressive and dysthymic patients.

Although NE was previously found not to be related to NK cytotoxicity in depressed patients (Irwin et al., 1991), in Experiment 2 NE levels among dysthymic patients were found to be highly correlated with several lymphocyte subsets including NK and CD4 cells. In contrast, among major depressive and control subjects, NE levels were not correlated with these lymphocyte subsets. It has been reported previously that mild to moderate stressors, including anticipation of a stressor (Bachen et al., 1992; Landmann et al., 1984; Manuck et al., 1991; Schedlowski et al., 1993b), or administration of sympathomimetics (Crary et al., 1983; Schedlowski et al., 1993a), will increase lymphocyte subsets, particularly NK cells, possibly reflecting a change of cell trafficking pattern (Ottaway and Husband, 1992). Further to the same point, Manuck et al. (1991) reported that the stress-induced increase of CD8 cells was apparent primarily in those subjects that were high sympathetic reactors. It is tempting to speculate that the increased number of lymphocytes in dysthymic and depressed patients likewise represent a change in cell trafficking patterns stemming from the increased perception (or reactivity) of stressors.

9.5. Variations of lymphocyte subsets in response to stressors

As previously observed (Bachen et al., 1992; Benschop et al., 1993a, 1994; Brosschot et al., 1992, 1994; Kiecolt-Glaser et al., 1992; Landmann et al., 1984; Manuck et al., 1991; Mills et al., 1995; Naliboff et al., 1991), in Experiment 3 exposure to a laboratory stressor in the form of a cognitive challenge (mental arithmetic) increased circulating NK cells, and to some extent influenced the number of CD8 cells. In a like fashion, several investigators (Futterman et al., 1992, 1994; Knapp et al., 1992; Naliboff et al., 1995; Strauman et al., 1993) demonstrated that elevated levels of NK cells were not only induced by cognitive stressors, but were also elicited by mood-induction procedures. In these studies, circulating NK cells were increased when subjects were asked to recall (or to simulate) positive or negative emotional experiences. In Experiment 4 the interview procedure increased circulating NK cells, and to some extent influenced the number of CD8 cells, without modifying either CD3, CD4 or CD19 cells. The finding that an interview regarding either recently experienced positive or negative events led to comparable increases of circulating NK cells, is congruent with earlier reports (Futterman et al., 1992, 1994; Knapp et al., 1992). Indeed, simply completing the questionnaires was sufficient to provoke an increase of NK cells, albeit to a lesser extent than that seen after the interview. Thus, these data suggest that either arousal associated with the questionnaire or interview, induction of mood changes, or the cognitive appraisal associated with positive or negative events was sufficient to promote the alterations of circulating NK cells. Curiously, in the depressive subjects an increase of NK cells was evident immediately after completion of the uplifts questionnaire, but not after completion of the questionnaire concerning day-to-day hassles. The source for such an outcome is not clear; however, it is conceivable that the elevated circulating NK cells were related to depressed patients becoming acutely aware of the few uplifts in their lives. Alternatively, searching for uplifts may have been stressful, or may have required more of a cognitive effort given the incongruency between their affective

state and being asked to consider positive events. Whatever the case, it is clear that variations of circulating NK cells are exquisitely sensitive to alterations of arousal/mood.

It has been reported that norepinephrine or epinephrine administration effectively increase circulating NK cell levels (Schedlowski et al., 1993a; Crary et al., 1983). Since stressors also provoke increased plasma catecholamine levels, it was hypothesized that these amines might be responsible for the altered NK cells associated with a stressor (Kiecolt-Glaser et al., 1992; Schedlowski et al., 1993b). In accordance with this position, it was demonstrated that the increase of circulating NK cells induced by a laboratory stressor in healthy subjects was attenuated by pretreatment with the β -adrenergic antagonist, propranolol, (Benschop et al., 1993b, 1994). Inasmuch as the adhesion of NK cells to endothelium is reduced by epinephrine, the increase of circulating NK cells following stress or β -adrenergic activation was attributed to catecholamine-induced recruitment of immunocompetent cells from storage sites (Benschop et al., 1994). The variations of NK cells associated with the interview procedure in Experiment 4 were not accompanied by alterations of plasma NE levels. Thus, it is unlikely that this amine was solely responsible for the lymphocyte changes. Likewise, although endocrine alterations have been associated with immune variations, the laboratory stressors in this experiment did not induce changes of either cortisol or ACTH. Accordingly, the contribution of these hormones to the altered NK cells is questionable. Yet, the findings that the more profound stress of the academic examination yielded marked increases of plasma NE, cortisol and ACTH, and also provoked a greater rise of circulating NK cells, are consistent with a role for one or more of these endocrine factors in promoting the NK alterations. Of course, an academic examination may be associated with a variety of other endocrine or behavioral variations that may have influenced circulating lymphocytes.

In contrast to the mental arithmetic and interview procedures, which did not influence plasma endocrine factors, the stress associated with anticipation of an academic examination provoked an increase of plasma cortisol, ACTH and norepinephrine levels in

nondepressed subjects. These findings are consistent with the supposition that anticipation of the academic exam may have been a more potent stressor than the psychiatric interview. Indeed, the extent of the NK cell change in anticipation of the academic examination was more pronounced than that produced by the interview. Moreover, unlike the interview, the more severe stressor was also associated with increases of the other lymphocyte subsets. These findings support the contention that circulating lymphocytes are sensitive to graded differences in stressor severity. As such, if the interview regarding life experiences had been more stressful for depressed than for control subjects, then the variations of NK cells should have been more pronounced among the depressed patients, and increases of other lymphocyte subsets might also have been expected. However, the effects provoked by the interview were limited to alterations of NK cells, and these were comparable in the depressive and control subjects. These data also suggest that the absence of more pronounced NK changes in the depressed subjects could not be attributed to ceiling effects precluding such an outcome. After all, the more intense stress of the academic examination yielded an increase of NK cells among controls which exceeded that produced by the interview. These data suggest that the elevated NK cells characteristic of depressed patients, are not due to greater stress reactivity. Of course, this should not be taken to suggest that depressed and non-depressed subjects were equally reactive with respect to other aspects of the immune response, nor should they be taken to imply that these groups are equally reactive to more intense life stressors. Certainly it is conceivable that the more profound and protracted stress ordinarily associated with depression (irrespective of whether these stresses are antecedents or consequences of the affective illness) was responsible for the elevations of basal circulating NK cells. Moreover, it is possible that the nature of the stressors used in the present experiments were simply inappropriate to detect differential responses to environmental insults. To be sure, an attempt was made to include relatively meaningful stressors, such as those involving recollection and discussion of day-

to-day adversities, but as will be discussed later, the efficacy of this procedure may have been compromised by the contrived laboratory setting involved in testing subjects.

Inasmuch as stressors increase circulating NK cells, it is conceivable that the elevated NK cells seen among depressive patients were related to their heightened stress perception and reliance on emotion-focused coping. Indeed, in their investigation of the contribution of stress and depression to immune measures, Irwin et al. (1990c) demonstrated that NK cytotoxicity was reduced in major depressive patients and in control subjects that were experiencing particularly severe stressors. In contrast, the numbers of circulating lymphocytes were elevated in those depressive patients that had been experiencing a relatively high degree of stress. In Experiment 3 stressful events and/or coping styles were related to circulating NK cells, but the specific correlates of circulating NK cells varied with affective state. In particular, in major depressive patients the NK cell number was highest among those patients that reported the greatest use of emotion-focused coping strategies (e.g., emotional containment and blame). Conversely, the use of problem-focused coping, which likely reflects a more effective coping strategy, was inversely related to circulating NK cells in both the major depressive and dysthymic patients. Curiously, in control subjects the number of circulating NK cells was inversely related to perceived stress and emotion-focused coping strategies (e.g., emotional containment and avoidance denial). This finding was not a spurious one, in that we have observed a similar relationship in a recently conducted study involving a larger number of control and dysthymic patients (Ravindran et al., 1996). It appears that although depressive illness is accompanied by increased stress perception and reliance on emotion-focused coping, only the latter is predictive of the NK cell number. It may simply be that increased stress perception is a fundamental characteristic of virtually all depressed patients and hence may not be effective as a discriminating factor for variations of circulating lymphocytes.

The change of NK cell numbers associated with the laboratory stressor in depressed subjects was directly related to the severity of depression. That is, those patients that

exhibited the most severe depression scores also tended to exhibit the greatest rise of NK cells in response to the laboratory stressor. As well, it appeared that the stressor-provoked variations of NK cells were also dependent on the coping styles that patients favored. In particular, the effects of the laboratory stressor were most pronounced in those patients who used individualistic coping strategies (e.g., cognitive restructuring, problem solving, avoidance denial, general activity), whereas as coping efforts involving external resources (e.g., social support, blame, emotional expression) were unrelated to the effects of the stressor on circulating NK cells. In effect, it seemed that while basal NK cell numbers were most closely aligned with the use of emotional focused strategies, the response to the stressor was associated with individualistic coping efforts. Results commensurate with these have recently been observed in our laboratory when cell proliferation stimulated by Con A was assessed in normal subjects (Zaharia et al., unpublished report). The finding that such an outcome occurred with respect to circulating NK cells as well as cell proliferation makes it likely that the outcome was not spurious.

Although stressors are often associated with increased plasma cortisol and ACTH, the lack of such effects in experiments 3 and 4 was not particularly surprising. As previously indicated, other investigators have noted that laboratory stressor-induced cortisol changes were more pronounced during the p.m., than when morning samples were taken (Kirschbaum et al., 1995). Additionally, since cortisol release may be pulsatile, blood samples collected over an extended period of time may have provided a better index of the dynamic variations of cortisol engendered by stressors. However, an indwelling catheter is fairly invasive, and in addition to itself provoking a stress response, may engender local inflammation which could affect cytokine, immune and neuroendocrine parameters. It has been suggested that cortisol measurement in saliva may provide an accurate index of circulating cortisol by non-invasive means (Kirschbaum and Hellhammer, 1994; Pruessner et al., 1997), and thus might have proven to be a more effective tool in the present investigations. As well, salivary cortisol measurements permit determination of

sequential cortisol values over extended periods, and hence might have been useful in determining not only cortisol variations attributable to the stressor, but also anticipatory variations that may have preceded the stressor. In this respect, Lupien et al. (1997) indicated that anticipatory cortisol responses peak at different times across individuals, and that in most cases, these actually occur prior to the laboratory challenge. Along the same line, Hellhammer and his associates (Pruessner et al., 1997; Smyth et al., 1997), indicated that cortisol variations in the first half hour following awakening, may reflect life stresses specific to the individual. Thus a series of salivary cortisol measures taken by the subject prior to coming into the laboratory, might better have reflected the differential HPA fluctuations associated with major depression and dysthymia.

9.6. Cell subsets in typical vs. atypical

As indicated earlier, inconsistent data have been reported concerning the immune correlates of depressive disorder. In contrast to the reduced circulating NK cells reported by some investigators (Andreoli et al., 1993; Evans et al., 1992), others reported either no effect (Darko et al., 1988) or an increase of this lymphocyte subset (Seidel et al., 1996; see also Experiments 2, 3, 4). While varied severity of illness may account for some of the disparate findings, the possibility exists that the immune changes may be related to either the chronicity of the illness or to the specific symptom profiles exhibited. In an effort to assess the contribution of these variables on circulating lymphocyte subsets, in Experiment 6 we assessed subtypes of depressed patients. These subgroups varied in terms of the chronicity of the illness (major depression vs. dysthymia) and the presence of either typical or atypical (reversed neurovegetative) symptoms.

Commensurate with the findings of Experiments 2, 3, and 4, it was observed that the T and B cell subpopulations did not vary as a function of depressive illness, whereas circulating NK cells were elevated in depressive patients. However, the latter effect was dependent upon the characteristic features of the illness, as well as the sex of the subject. In particular, as previously reported (Evans et al., 1992; Maes et al., 1994a), circulating NK

cells among male depressive patients, irrespective of the subtype of the illness, were higher than in females. Among male subjects the highest NK cell levels were seen in major depressive patients, but relative to control subjects, the circulating NK cells were also elevated in each of the remaining depressive groups. It is significant, however, that in the two typical groups circulating NK cells exceeded those seen in the atypical patients. As well, NK cells in the major depressive groups were greater than in the two dysthymic conditions. In contrast to the effects seen in male depressive patients, among females the circulating NK cells were elevated in the typical major depressive patients, but not in any of the remaining conditions. As indicated by Maes et al. (1994a), some of the inconsistent results that have previously been reported may stem from the lack of specificity regarding severity of illness, patients' previous drug history, as well as their gender and age. Inasmuch as the NK cell elevations in the present investigation were greater in major depressive than in dysthymic patients, these data suggest that severity of illness was fundamental in promoting the elevated NK cells. In addition, the results indicated that the presence of typical vs. atypical features, as well as the sex of the patient, may be particularly pertinent in determining circulating NK cell levels.

In accordance with the proposition that the elevated NK cell levels were related to depressive illness or symptoms, it was observed that in those subgroups in which NK cells were elevated, successful antidepressant treatment resulted in normalization of the NK cells, suggesting that this immune parameter was directly related to the depressive illness. Moreover, these data would suggest that the number of NK cells was a state, rather than a trait marker of depression. Of course, the possibility cannot be dismissed that the peripheral effects of antidepressants may have promoted redistribution of lymphocytes, but such an outcome should have provoked alterations of NK scores in both drug responders and nonresponders. Nevertheless, as part of a more detailed analysis of this and related questions, we are currently comparing the potential effects of cognitive therapy and drug therapy in the alleviation of dysthymic symptoms and on endocrine and immune factors.

Given that depressive illness may be preceded by stressful experiences, and depression itself is associated with increased stress perception coupled with the use of ineffective coping styles (Billings and Moos, 1985; Brown and Harris, 1989; Lazarus, 1993), the view might be taken that the elevated number of NK cells in depressives may be secondary to their stressor experiences and perceptions. However, we observed that relative to control subjects, the increased perception of day-to-day stress and reliance on emotion-focused coping strategies were comparable among depressive patients, irrespective of whether the mood disorder involved major depression or dysthymia, as well as typical or atypical features.

As indicated earlier, since immune alterations similar to those seen in depression, including the reduced NK cytotoxicity, can be induced in non-depressed subjects by sleep deprivation (Irwin et al., 1994; Moldofsky, 1995), the possibility exists that immune alterations in depressed patients may be secondary to neurovegetative symptoms. The finding that the elevated circulating NK cells seen in major depressive patients were less profound in the presence of reversed neurovegetative symptoms (i.e., in atypical patients) is consistent with this proposition. Although objective indices of sleep were not obtained in the present investigation, correlations between the patients' sleep disturbances and fatigability, as determined from the HAM-D scale, revealed that these variables were unrelated to NK cell levels. In contrast, eating was found to be inversely related to circulating NK cells, raising the possibility that this feature of depression was particularly pertinent with respect to levels of this lymphocyte subset. It must be underscored, however, that the number of NK cells was increased in male patients regardless of whether they exhibited typical or atypical features (although this effect was more pronounced in the typical patients). Thus, these data suggest that while neurovegetative symptoms may contribute to the immune alterations among the male patients, other factors, such as dysphoric mood, are responsible for the elevated circulating NK cells. It ought to be considered, as well, that in view of the altered neurovegetative features of atypical

depression, particularly with respect to increased sleep, differences of diurnal rhythms may exist regarding endocrine factors and circulating lymphocytes in subtypes of affective illness. Although blood was only sampled during the morning hours in the present investigation, it is conceivable that the differences in circulating NK cells between typical and atypical depressive patients may be secondary to shifts of diurnal rhythm.

The finding that the NK elevation was less marked in dysthymic than in major depressive patients suggests that the NK levels may also be related to severity and/or chronicity of depression. In particular, the severity of depression in dysthymic patients is less profound than in major depressive patients, and it is certainly possible that illness severity and NK cell levels are directly related to one another. Alternatively, it is possible that the elevations of NK cells associated with mood disorders are subject to adaptation with continued depression. After all, it is conceivable that in chronic depression compensatory changes occur wherein immune alterations, that might otherwise be associated with affective illness, are diminished. In the present experiments the major depressive patients did not have a prior history of depressive illness. It remains to be determined whether superimposition of a major depressive episode on a dysthymic backdrop (i.e., double depression) would be associated with alterations of circulating NK cells.

Not surprisingly, most studies of immune functioning in depression have focused on functional, rather than enumerative measures of immunity (e.g., cell proliferation or cytotoxicity). In fact, the meaningfulness of studies assessing enumerative immune measures has been questioned. Yet, it seems that one of the most consistent findings in the psychoneuroimmunological literature is that stressors are associated with elevated levels of circulating NK cells (Kiecolt-Glaser et al., 1992). In fact, as the results in Experiments 3 and 4 would suggest, circulating NK changes may not only be sensitive to graded stressor severities, but may also be more sensitive to stressor characteristics than traditional endocrine measures, such as plasma cortisol concentrations. As indicated earlier, it has

been suggested (Dhabhar et al., 1995) that the distribution of immune cells between different tissues may be a fundamental feature of the immune system's ability to deal with challenges. As well, Maes (1995) has proposed that the distribution of circulating lymphocyte subsets may be indicative of an immune activation associated with affective illness. The findings of the present investigation, while not necessarily inconsistent with this proposition, suggest that neurovegetative characteristics of the illness, as well as the sex of the subject, need to be considered in the analysis of immune-depression relationships. In fact, although functional immune measures were not determined in the present studies, when considered in conjunction with reports that alterations of sleep may affect NK cytotoxicity and cytokine activity (Irwin et al., 1994; Moldofsky, 1995), it seems clear that analysis of the depression-immune profile ought to consider the specific symptoms and chronicity of the illness. Further to this same point, inasmuch as recurrence of depressive illness, or protracted exposure to stressful experiences, could ostensibly be related to variations of endocrine and neurotransmitter functioning (Post and Weiss, 1995; Tilders et al., 1993), the possibility needs to be considered that the immune correlates of depression might also vary across episodes of illness.

Summarizing, it is clear that depressive illness may be associated with immune alterations, such as elevated circulating NK cells. The data of the present investigations make it clear that an accounting of the factors responsible for the diverse outcomes seen across studies requires that greater attention be devoted to the characteristics of the illness. This may include documentation not only of demographic factors (e.g., sex and age), but also of the characteristic features comprising the illness. In addition to severity of the depression, it may be essential to determine the chronicity of the illness, as well as the typical vs. atypical symptoms. Furthermore, since endocrine variations associated with dysthymia and major depression may be dependent on whether the illness involved early vs. late age of onset, as well as the episode chronology (initial episode vs. recurrence)

(Ravindran et al., 1997), it ought to be considered that such factors might also contribute to the diverse immune effects reported across laboratories.

9.7. Cytokine variations with typical / atypical features

Depressive disorders, it will be recalled, may be associated with several neuroendocrine variations. However, profound interindividual differences exist in the symptoms subserving the illness, the response to pharmacotherapy, as well as the neuroendocrine correlates of the disorder. Further, subtypes of depression may differ in terms of their response to specific pharmacological treatments, and with respect to their neuroendocrine factors. For instance, major depression is characterized by HPA alterations, including variations of plasma ACTH and cortisol levels, nonsuppression of cortisol release following dexamethasone challenge, and a blunted ACTH response to CRH challenge (Gold et al., 1995; Nemeroff, 1996). While limited data are available, it appears that in illnesses involving atypical features (e.g., bulimia, seasonal affective disorder, and chronic fatigue syndrome) the elevated ACTH levels are accompanied by reduced cortisol, a blunted ACTH response to CRH challenge (Demitrack et al., 1991; Gold et al., 1995; Nemeroff, 1996), and absence of the CRH hypersecretion characteristic of typical depression (Nemeroff, 1996). Among dysthymic patients the profile is different yet again; the HPA alterations are generally not evident, and there is reason to believe that cortisol levels may actually be reduced (Ravindran et al., 1997).

It appeared in Experiment 7 that although affective illness may be associated with HPA dysfunction, elevations of ACTH were most pronounced in atypical major depression, while plasma cortisol levels were reduced. These data clearly parallel the aforementioned neuroendocrine characteristics observed in disorders involving atypical depressive features. Further, these findings are consistent with the possibility that the elevated ACTH and reduced cortisol, may reflect hypofunction of CRH neurons or adrenal insensitivity, hence resulting in less feedback inhibition (Demitrack et al., 1991; Gold et

al., 1995; Levitan et al., 1997; Joseph-Vanderpool et al., 1991; Nemeroff, 1996). Of course, HPA disturbances associated with depressive disorders may occur at any number of different levels, and may involve more than a single dysfunctional mechanism (Holsboer, 1995; Akil and Morano, 1995). Thus, it is premature to assume that HPA disturbances in atypical depression involve the same mechanisms as those subserving other illnesses with atypical features. Furthermore, in the present investigation morning blood samples were collected over only a few minutes, and it remains to be determined whether a similar outcome would be evident with more protracted blood sampling over other phases of the diurnal cycle, particularly since cortisol and ACTH levels were not elevated in the typical major depressives.

Consistent with reports in melancholic patients (Maes, 1995), levels of mitogen stimulated IL-1 β production were enhanced in depressive illness. However, the finding in Experiment 7 that this occurred among dysthymic patients, and not in either the typical or atypical major depressives, was unexpected. It is, of course, possible that alternate types of assays for cytokine production may have yielded different results. For instance, the use of PHA combined with LPS to stimulate cells in whole blood may have provided a better indication of cytokine production from macrophages and T cells than that observed in assays involving mitogen-stimulated mononuclear cells (De Groote et al., 1993).

The view was offered, it will be recalled, that depression may be associated with the enhancement of some components of the immune response (much like an acute phase reaction), which may ultimately promote suppression of other aspects of immune functioning (Maes, 1995). Among other things, mitogen-induced IL-1 β and IL-6 production was increased in blood mononuclear cells of severely depressed patients (Maes, 1995). While not excluding the possibility that illness severity may be a pertinent feature in promoting the enhanced IL-1 β production (after all, melancholic patients were not assessed in the present study), it seems likely that illness chronicity or age of onset may also be important in this respect. Dysthymia is a chronic illness and is associated with increased

stress perception and inadequate coping styles (Experiments 1, 5), and as such, some of the neuroendocrine characteristics of a chronic stressor may be engendered in these patients. For instance, chronic stress may be associated with phenotypic variations of CRH neurons within the paraventricular nucleus, such that they coexpress arginine vasopressin (AVP) and CRH (Tilders et al., 1993). The fact that dysthymic patients do not exhibit increased ACTH and cortisol, and even display reduced cortisol levels (Ravindran et al., 1997), necessitates evaluation of the effects of various challenges in dysthymic patients (e.g., ACTH, CRH, AVP, as well as serotonergic acting agents) in order to identify the nature of the HPA dysfunction (Holsboer, 1995; Gold et al., 1984). At this juncture it is not clear whether the cytokine alterations in dysthymia are related to HPA functioning, but given the well documented interactions between cytokines and endocrine systems, such a possibility needs to be considered (Munck and Guyre, 1991), despite the fact that plasma cortisol levels in the present investigation were unrelated to IL-1 β production in either dysthymic or major depressive patients. Further to this point, as already indicated, plasma cortisol was assessed at only a single time for each subject. Yet, the release of cortisol is pulsatile and hence plasma cortisol levels may not be stable. Moreover, when evaluating cell proliferation or cytokine production in proliferating cells, it should be considered that these represent both active and sustained processes. Thus, it should not be surprising to find that measurements of static cortisol may not be closely aligned with the dynamic processes associated with lymphocyte functioning. Thus, while cortisol may indeed have immunosuppressive effects in depressed populations, identifying the contribution of hormonal changes to immune functioning may be exceedingly difficult unless temporal characteristics of both measures are considered.

The increased IL-1 β production is one of few physiological characteristics that distinguish dysthymic patients from both major depressive and control populations. It will be recalled that the HPA disturbances which characterize major depression (as evidenced following dexamethasone and/or CRH challenge) are not observed in dysthymic patients

(Ravindran et al., 1997; Thase and Howland, 1995; Maes et al., 1994b). Similarly, growth hormone secretion in response to physiological challenges, as well as thyroid stimulating hormone blunting in response to TRH stimulation, are less likely to occur in dysthymia than in major depression (Ravindran et al., 1997; Thase and Howland, 1995). While neuroendocrine disturbances may occur primarily among early illness onset dysthymics (Szadoczky et al., 1994), there are few neuroendocrine studies that distinguished between dysthymic and non-depressed subjects. In part, this may stem from the subtle pathophysiological disturbances in dysthymia, and the use of neuroendocrine analyses that tap circulating hormonal levels rather than the temporal patterns of hormone release (Akiskal et al., 1996). Yet, as IL-1 β is a potent stimulator of CRH release, the production of this cytokine may be an essential component of the neuroendocrine cascade characterizing dysthymia. As discussed earlier, it was shown that with the passage of time following stressor or IL-1 exposure (or following chronic stressor experiences), phenotypic variations of CRH neurons may occur, such that they come to coexpress CRH and AVP in the external zone of the median eminence (Tilders et al., 1993). As CRH and AVP may synergistically stimulate HPA activity, one can imagine that immune activation and the cytokine variations may elicit persistent HPA alterations. As these phenotypic alterations are thought to represent permanent (or at least very long lasting) variations of the HPA state, it is thought that the resulting neuroendocrine activation may favor the depressive symptomatology, and may play an important role with respect to the high rate of illness recurrence and relapse.

In contrast to the increased IL-1 β in supernatants of mitogen-stimulated lymphocytes, we have observed that circulating serum IL-1 β was not increased in either typical major depressive or in dysthymic patients. In contrast, however, among atypical major depressive patients, circulating IL-1 β levels were greatly increased, and normalized with treatment response (Griffiths et al., 1996). It could be assumed that the elevated levels of serum IL-1 β seen in atypical depression were secondary to the neurovegetative features

of this depressive subtype. However, animal studies have shown that IL-1 β provokes illness behaviors (including increased sleep and fatigue) (Kent et al., 1992), and thus it is just as likely that elevated circulating IL-1 β contributes to the neurovegetative features of atypical depression. The finding that illnesses involving atypical depressive features (e.g., chronic fatigue syndrome) may be associated with HPA disturbances (e.g., reduced plasma cortisol, increased ACTH, and reduced ACTH release following oCRH challenge) (Demitrack et al., 1991; Gold et al., 1995), coupled with the fact that IL-1 β is a potent stimulator of CRH release (Sluzewska et al., 1995), raise the possibility that elevated circulating IL-1 β levels contribute to the pathophysiology of atypical depression.

In contrast to IL-1 β , mitogen-stimulated IL-2 levels were appreciably reduced irrespective of whether patients presented with typical or atypical features, or whether patients fit the dysthymic or major depressive profile. The reduced IL-2 production was highly correlated with plasma NE levels in both depressive groups, possibly reflecting characteristics of NE receptors on lymphocytes of depressive subjects. It would seem that the reduced IL-2 production might suggest that this aspect of immune functioning, insofar as it may reflect T cell activation, is impaired in depression. However, the net action of IL-2 is dependent upon the levels of soluble IL-2 receptors, and measurement of IL-2 *in vitro* is hampered by the binding of the cytokine to its receptor, a problem which can be overcome by adding anti-Tac (an IL-2 receptor blocking antibody) to the culture (Baroja and Cueppens, 1987). Since anti-Tac was not used in the present investigation, it cannot be said with any certainty whether depressive illness was associated with reduced IL-2 or altered binding with IL-2 receptors. However, we recently observed cell proliferation stimulated by the T-cell mitogens, PHA and Con-A, to be profoundly reduced in dysthymic patients, but only modest reductions were seen in typical and atypical major depressive patients (Griffiths et al., 1996). Thus, depressive illness may indeed be associated with impairment of some aspects of immune functioning, and once again illness chronicity or age of onset may be determining factors in this respect.

Summarizing, although it had been expected that IL-1 β and IL-2 production would differ with typical and atypical depressive profiles, such an outcome was not observed. While neurovegetative features may affect some aspects of immune functioning (Irwin et al., 1994; Moldofsky 1995; Experiment 6), these factors did not account for the IL-1 β variations observed in depressive illness. However, major depressive and dysthymic patients (both typical and atypical) could be distinguished from one another on the basis of their cytokine profiles. Specifically, IL-1 β production was elevated primarily in dysthymia, and the extent of the increase was correlated with the age of onset and the chronicity of the illness, rather than simply reflecting the presence of a mood disorder.

9.8. A provisional model of chronic depressive illness

It is clear from the preceding sections that limited data are available concerning the mechanisms underlying dysthymia. Because of dysthymia's chronic, low-grade nature, animal models of the illness have yet to be developed. Nevertheless, data derived from animal studies offer some clues as to the potential persistent effects of stressor experiences that may be relevant to dysthymia. In particular, it seems that in addition to any immediate consequences, stressors may also proactively influence the neurochemical response to subsequently encountered aversive stimuli, hence favoring long-term behavioral repercussions. Post (1995) indicated that although the variations of certain peptides (as gauged by activation of immediate early genes) persist for relatively brief periods following a single stressor session, with repeated challenges the variations of some peptides will be more readily induced and will be more persistent (e.g., CRH, and to a greater extent TRH). It was suggested that depressive illness may initially stem from the neuroendocrine alterations provoked by a stressor. However, with each subsequent stress experience, or with each episode of depression, the sensitization becomes more pronounced, such that progressively smaller degrees of psychosocial stress are required to provoke the onset of a depressive episode. Ultimately, episodes of depression may occur in the absence of obvious stress triggers. In fact, it was reported that unlike the initial episode, recurrent

episodes of depression were less likely to be preceded by antecedent stressors (Cassano et al., 1990; Perris, 1984) and even occurred spontaneously (i.e., without any obvious precipitant stressor) (see Thase and Sullivan, 1995; see Post, 1995). Of course, it is often difficult to identify significant or meaningful stressors that may be pertinent to a given individual, thus conclusions concerning the presence or absence of stressful precipitants of depression may be difficult to validate. In this respect, it is conceivable that in addition to stressors of a psychological nature, the individual may be challenged by a physiological stressor, such as a virus. Such an insult may be interpreted by the CNS in the same way as a psychosocial stressor, and hence may trigger the cascade of events resulting in neuroendocrine, cytokine, and mood alterations (Anisman and Merali, 1998).

In support of the notion that the first episode of depression and recurrent depression are biologically distinguishable from one another (possibly reflecting a sensitization effect as proposed by Post), EEG sleep profiles were more severely disrupted among individuals suffering from recurrent depressive episodes than among individuals suffering from a single episode (Thase et al., 1995). However, it has been reported that the degree of HPA dysregulation did not vary with recurrent episodes of depression (Meador-Woodruff et al., 1987). Specifically, neither post-dexamethasone cortisol levels nor suppression status varied as a function of prior number of depressive episodes or duration since the initial episode. However, HPA dysregulation may occur at any number of different levels, and conclusions limited to cortisol functioning may not provide an unambiguous portrayal of peptide variations relevant to depressive disorders. This notwithstanding, Post and Weiss (1995) indicated that following remission of symptoms, patients who show persistent neurobiological characteristics (e.g., early escape from dexamethasone suppression of cortisol release; blunted TSH response to TRH stimulation; high CSF levels of CRH) are those who are most vulnerable to illness recurrence (Arana et al., 1985; Banki et al., 1988, 1992; Nemeroff and Evans, 1984).

While the model developed by Post and Weiss (1995) was meant to accommodate recurrent depression, it may also be applicable to the analysis of dysthymia. In this respect, however, it is important to underscore that sensitization effects are not limited to the neuroendocrine factors discussed by Post, nor is such a sensitization limited to antecedent stressors. In fact, it has been demonstrated that several neurochemical variations associated with stressors, psychostimulant use (amphetamine and cocaine), and electrical stimulation of the amygdala or the piriform cortex, may permanently enhance the neuronal response to subsequent manipulations (sensitization) (Kalivas and Stewart, 1991). As will be seen shortly, this applies to the effects of cytokine treatments as well. Thus, while the long-term behavioral impairments associated with stressors may stem from a sensitization (Anisman and Sklar, 1979; Anisman et al., 1991), such deficits may also be provoked by other extrinsic challenges.

As already indicated, the sensitization model is amenable to the recurrence of depression, and it also appears to be relevant to bipolar illness involving slow cycling, and even the chaotic profile associated with ultra rapid cycling. There is reason to suppose that it may also be pertinent to the analysis of dysthymia. Indeed, dysthymia is associated with increased stress perception, the use of inappropriate coping styles, and has been shown to develop with chronic stressors (such as care-giving; Dura et al., 1990). Moreover, since the symptoms are most often mild-moderate, tend to wax and wane, it ought to be considered that dysthymia may, like recurrent depression, be related to chronic, mild/moderate stressors. The illness may be exacerbated through sensitization processes, possibly culminating in double depression. Of course, this does not suggest that such processes are equally applicable to the various dysthymic subtypes. It may be important to distinguish not only between character spectrum and subaffective disorders, but also between early- and late-onset, as well as typical and atypical subtypes of the disorder.

In modeling dysthymic disorders, several features of this illness need to be considered. Among other things, it needs to be understood why (a) this disorder is not

associated with cortisol abnormalities like those seen in major depressive illness, (b) following treatment for double depression, patients return to their dysthymic states rather than a full abatement of symptoms (euthymic state), and (c) the illness is characterized by a chronic (albeit fluctuating) course. The sensitization model described earlier may be pertinent in this respect, but relatively recent findings concerning sensitization effects introduced still another facet of stressor actions that may be relevant. Specifically, these findings derive from the observation that the variations of ACTH ordinarily associated with acute stressors do not decline following repeated insults, and may even increase upon subsequent presentation of a novel stimulus. The absence of an adaptation to chronic stressor effects was thought to reflect the conjoint action initiated by increased CRH and AVP (Dallman, 1993), which acted synergistically to provoke pituitary ACTH release. Tilders et al. (1993) indicated that in addition to the functional amine and endocrine variations associated with stressor reexposure, repeated stressor experiences may come to promote phenotypic variations of neurons that are ordinarily responsive to stressors, particularly CRH containing neurons originating in the PVN and having terminals in the external zone of the median eminence. In particular, following chronic stressor exposure the increased expression of AVP appeared in the CRH containing neurons. As the co-release of these peptides act synergistically to promote ACTH secretion, the chronic stressor regimen increased the potential for elevated HPA functioning (Bartanusz et al., 1993; Tilders et al., 1993).

It is of interest that in the studies conducted by Tilders and his associates, the chronic stressor-induced phenotypic variations were absent immediately following an acute stressor. Similarly, it has been reported that the sensitization of neurotransmitter changes (e.g., DA in mesolimbic sites) exerted by either stressors or psychostimulants take some period of time to develop (approximately 1- 2 weeks), and may not be evident at relatively brief intervals following the initial treatment (Antelman, 1988; Kalivas and Stewart, 1991). In a like fashion, Tilders et al (1993) indicated that two weeks following exposure to an

acute stressor, increased CRH and AVP coexpression (like that seen after a chronic stressor) occurred within the external zone of the median eminence. Thus, the effect may have been related to the passage of time following the initial stressor experience, rather than simply to the chronicity of the stressor per se.

In addition to their effects on endocrine factors, stressors are also associated with central neurotransmitter alterations that may, of course, be fundamental to the induction of dysthymic illness. Indeed, variables thought to be pertinent to depression have been shown to influence the amine alterations associated with stressors (Anisman et al., 1991). Specifically, acute stressors increase monoamine turnover and levels, with the magnitude of the effects being directly related to the severity of the stressor, and varying with organismic variables, such as the strain and age of the animals (Anisman et al., 1991; Weiss and Simson, 1985, 1989). Following mild-moderate stressors, 5-HT, NE, and mesocorticolimbic DA utilization and synthesis increase. When the stressor is uncontrollable, the burden of coping rests more heavily on endogenous mechanisms, and as a result, amine utilization may exceed synthesis, culminating in a net amine decline. Typically, the amine alterations engendered by an acute stressor are fairly transient, persisting for a matter of hours. However, it is of particular interest that when the stressor is presented at some later time, the amine variations are more readily induced (Abercrombie et al., 1989; Deutch and Roth, 1990; Deutch et al., 1985; Kalivas and Stewart, 1991). In addition to such a sensitization effect, the increased utilization may occur even when the subsequent session involves a novel stressor (cross-sensitization) (Kalivas and Stewart, 1991; Nisenbaum et al., 1991, 1992). The view was expressed that such conditions render the organism less well prepared to deal with psychological insults, hence favoring behavioral disturbances (Anisman et al., 1993; Deutch and Roth, 1990; Weiss and Simson, 1985). Of course, the possibility should not be dismissed that sensitization of stressor-induced HPA alterations, and the variations of central glucocorticoid receptors, may

promote monoamine variations which feature in depression (DeKloet and Reul, 1987; Dinan, 1994).

Analyses of the stressor-depression relationship ought to focus to a greater extent on the contribution of repeated insults. In contrast to acute stressors, the amine reductions may be absent following chronic stressors. Among other things, a chronic, predictable stressor regimen may result in the synthesis of amines being increased, and the excessive utilization being moderated, so that levels of the transmitter are maintained. In fact, the augmented amine synthesis may persist for days following a chronic stressor, whereas the enhanced utilization drops off quickly upon stressor termination, as a result of which amine levels may be elevated. Additionally, chronic stressors may be accompanied by down-regulation of β -NE receptor sensitivity, and such effects may persist for as long as 24 hr following stressor termination (Stone, 1983). Interestingly, the neurochemical alterations associated with chronic predictable stressors may be absent following an unpredictable stressor regimen (Molina et al., 1990). Likewise, while chronic predictable stressors may result in behavioral adaptation (Weiss and Simson, 1985; Zacharko and Anisman, 1991), persistent behavioral disturbances may be evident with chronic unpredictable stressors (Willner, 1987). Thus, variables which promote amine reductions might be expected to increase vulnerability to depression (e.g., chronic unpredictable stressors), whereas chronic predictable stressors, which lead to neurochemical adaptation would not be associated with behavioral dysfunction.

Inasmuch as dysthymia is a chronic illness, often of mild-moderate severity and typically without clear precipitating events, this disorder is unlikely to be related to a single strong stressor experience. To the contrary, such a stressor might be more closely aligned with an acute major depressive episode. It is more likely that dysthymia reflects the actions of more sustained, variable, and probably less intense stressors, coupled with the use of inadequate or inappropriate methods of coping. Such a stressor schedule may culminate in permanent variations of endocrine and neurochemical responsivity to psychological insults,

and perhaps phenotypic alterations in the characteristics of cells that affect HPA functioning (e.g., such as AVP expression in CRH cells). Since the response to stressors is subject to sensitization, it would not be altogether surprising to find that double depression may be related to the superimposition of a further stressor on the backdrop of dysthymia, possibly reflecting the exacerbation of central amine disturbances. Furthermore, if a chronic stressor culminates in phenotypic variations of CRH/AVP neurons, then it might be expected that even after alleviation of the major depression in double depressive patients treated with antidepressant medication, the dysthymic profile would again emerge. In effect, we are suggesting that dysthymia may reflect a chronic state of altered endocrine and central neurotransmitter functioning which may be related to sustained stressor experiences together with inadequate coping. Indeed, even with the remission of symptoms, endocrine disturbances may persist, thereby increasing the probability of symptom recurrence. Figure 7 provides a provisional model showing the effects of different stressor regimens on HPA functioning and the presumed variations of affective state. Although neuroendocrine factors are the focus of the figure, the stressor-induced neurotransmitter variations are also thought to be fundamental to the illnesses.

In relating stressful events to the mechanisms underlying dysthymia and recurrent depression, we have defined stressors in a fairly broad way. As discussed earlier, it has been posited that, among other things, the immune system acts like a sensory organ informing the brain of antigenic challenge (Blalock, 1984, 1994). Furthermore, given the nature of the neurochemical changes elicited by antigens and cytokines, it was suggested that immune activation may be interpreted by the CNS as a stressor (Anisman and Merali, 1998; Anisman et al., 1993; Dunn, 1990; Dunn et al., 1989). To be sure, the effects of systemic stressors (e.g., those associated with viral insults, bacterial endotoxins, cytokines) are not entirely congruous with those elicited by processive stressors (i.e., those involving higher-order sensory processing, e.g., fear conditioning, exposure to a predator or novel environment) (Herman and Cullinan, 1997). Nevertheless, cytokines may be part

Figure 7. Hypothesized variations of HPA functioning and mood changes associated with various stress regimens. In addition to the particular HPA alterations denoted in the figure, stressors have also been shown to produce several central neurotransmitter alterations. These changes may be secondary to or independent of the HPA alterations, but may contribute to depressive symptomatology. An acute mild stressor provokes CRH (CRF) release from the PVN, which gives rise to pituitary ACTH and adrenal corticoid secretion. The magnitude of the endocrine variations are more pronounced following a severe stressor, and may be associated with acute depression. With a protracted stressor phenotypic changes of CRH neurons occur, such that they now coproduce and release AVP, leading to more pronounced ACTH and corticoid secretion. In addition, under such a stress-induced sensitized state the individual may be more susceptible to prolonged physiological arousal, major depression and dysthymia. The phenotypic change of CRH neurons persists following the termination of the stressor experience, may represent or underlie dysthymic states. In dysthymic individuals further encounters with acute stressors of moderate severity may elicit profound endocrine and neurochemical variations and the promotion of double depression. Treatment of the double depression may lead to abatement of the excessive HPA alterations, but the dysthymic characteristics may persist owing to the phenotypic CRH-AVP changes. As indicated earlier, sensitization effects following stressor experiences are not limited to HPA functioning, but have been documented with respect to several central neurotransmitters. Thus, the characteristics of dysthymia and double depression may also be ascribed to these central transmitter alterations (Provisional Model; adapted from Ravindran et al, 1997).

of a regulatory loop that, by virtue of their effects on CNS functioning, might influence behavioral outputs and may even contribute to the symptoms of behavioral pathologies, including mood and anxiety-related disorders (Anisman and Merali, 1998; Anisman et al., 1993; Crnic, 1991). It is certainly the case that both processive and systemic stressors effectively increase HPA activity. However, while processive stressors do so via limbic circuits, the HPA alterations elicited by systemic stressors result from limbic-independent processes (Herman and Cullinan, 1997).

The results of our investigations have indicated that dysthymia is associated with increased production of IL-1 β . To be sure, it remains to be determined whether the IL-1 β variations are secondary to the illness or, in fact, play an etiological role. It is of particular significance, however, that IL-1 β not only affects HPA functioning, as described earlier, but may influence hippocampal 5-HT activity, as well as that of NE and DA in hypothalamus, locus coeruleus and mesolimbic sites (Lacosta et al., 1998; Song et al., 1998). This cytokine is known to provoke behavioral changes, some of which are reminiscent of the characteristics of atypical depression, including increased sleep, lethargy and reduced locomotor activity (Kent et al., 1996), and may provoke anxiety (Anisman and Merali, 1998). Further to this point, the alterations of IL-2 that have been reported in major depressive patients have also been shown to affect mesolimbic DA activity (Song et al., 1998), and have been shown to elicit anhedonic responses (Anisman et al. 1996, 1998) and disturbances of cognitive functioning (Anisman and Merali, 1998; Hanisch and Quirion, 1996). In effect, it seems that cytokine alterations which have been associated with depressive illnesses may themselves come to affect central neurotransmitter disturbances that have been implicated in depression.

It has been proposed that alterations of HPA activity in depressive disorders could be the primary abnormality in depression, rather than simply an illness response (De Kloet and Reul, 1987; Dinan, 1994). Moreover, it has been suggested that among biologically predisposed individuals chronic stressors may come to promote sustained HPA activation

which leads to adverse effects (Checkley, 1992). As alluded to earlier, the view has been offered that the monoamine variations often associated with depression may actually stem from endocrine alterations. It should be emphasized, however, that dysthymia and illnesses associated with atypical depressive features (including bulimia and chronic fatigue syndrome) ought to be distinguished from that of major depression. In particular, these syndromes may be characterized by low cortisol levels, but possibly elevated ACTH concentrations (Demitrack et al., 1991; Levitan et al., 1996). Likewise, as discussed earlier, atypical major depressives exhibited elevated ACTH levels, as well as increased serum IL-1 β (Griffiths et al., 1996). The predominance of atypical symptoms in dysthymia, low frequency of a positive DST, and low or normal mean plasma cortisol levels, may lead to the speculation that dysthymia is associated with altered sensitivity/activity of HPA functioning. In fact, there is some evidence that serotonergic agents may reverse some of these parameters of underactivity (Ravindran et al., 1994a). The possibility needs to be considered that the chronicity of dysthymia may contribute to elevated ACTH secretagogues, but may result in adrenal insensitivity. It is necessary at this juncture to assess the effects of various challenges (e.g., ACTH, CRH, AVP, as well as serotonergic acting agents) on different aspects of HPA functioning in dysthymic patients.

It is apparent that the paucity of data from human studies, and the lack of a suitable animal model for dysthymia has limited the conclusions that can be drawn concerning the etiology of this disorder. Nevertheless, the available data have made it clear that determinations of the mechanisms underlying dysthymia, and the development of adequate treatment strategies, will require that several fundamental features be included in any experimental analysis. Foremost in this respect is the need to subtype subjects according to definite criteria. In particular, it will be of obvious advantage to distinguish between pure dysthymia, double depression, and other forms of chronic depression. Additionally, however, patients need to be characterized into homogeneous subgroups (e.g., early- vs.

late-onset; subaffective vs. character spectrum), and the symptom profile of the dysthymic patients ought to be considered (vis-à-vis the presence of typical or atypical neurovegetative symptoms).

10. References

- Abercrombie ED, Keefe KA, DiFrischia DS, Zigmond MJ. Differential effects of stress on in vivo dopamine release in striatum, nucleus accumbens and medial frontal cortex. *J Neurochem* 1989; **52**: 1655-1658.
- Abraham E. Effects of stress on cytokine production. In: Jasmin G, Cantin M (eds). *Stress Revisited. Neuroendocrinology of Stress*. Karger: Basel, 1991, pp 45-62.
- Abramson LY, Seligman MEP, Teasdale JD. Learned helplessness in humans: Critique and reformulation. *J Abn Psychol*. 1978; **87**: 49-74.
- Agosti V, Stewart JW, Quitkin FM. Life satisfaction and psychosocial functioning in chronic depression: effect of acute treatment with antidepressants. *J Affect Dis* 1991; **23**: 35-41.
- Agren H. Symptom patterns in UP and BP depression corellating with monoamine metabolites in CSF. *Psychiatry Res* 1980; **2**: 225-232.
- Akil HA, Morano MI. Stress. In: Bloom FE, Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven Press: New York, 1995, pp. 773-785.
- Akiskal HS. Dysthymic and cyclothymic depressions: therapeutic considerations. *J Clin Psychiatry* 1994; **55** (Suppl): 46-52.
- Akiskal HS. Towards a definition of dysthymia: Boundaries with personality and mood disorders. In: Burton SW, Akiskal HS (eds). *Dysthymic Disorders*. Gaskell: London, 1990, pp 1-12.
- Akiskal HS. Dysthymic disorder: psychopathology of proposed chronic depressive subtypes. *Am J Psychiat* 1983; **140**: 11-20.
- Akiskal HS, Bolis CL, Cazzullo C, Costa e Silva JA, Gentil V, Lecrubier Y, Licinio J, Linden M, Lopez-Ibor JJ, Ndiaye IP, Pami L, Prilipko L, Robertson MM, Robinson RG, Starkstein SE, Thomas P, Wang Y, Wong M-L. Dysthymia in neurological disorders. *Mol Psychiatry* 1996; **1**: 478-491.

- Akiskal HS, Judd LL, Gillin C, Lemmi H. Subthreshold depressions: clinical and polysomnographic validation of dysthymic, residual and masked forms. *J Affective Disord* 1997; **45**: 53-63.
- Akiskal HS, King D, Rosenthal TL, Robinson S, Scott-Strauss A. Chronic depressions, 1: clinical and familial characteristics in 137 probands. *J Affect Dis* 1981; **3**: 297-315.
- Akiskal HS, Lemmi H, Dickson H, King D, Yerevanian B, Van Valkenberg C. Chronic depressions. Part 2. Sleep EEG differentiation of primary dysthymic disorders from anxious depressions. *J Affective Disord* 1984; **6**: 287-295.
- Akiskal HS, Rosenthal TL, Haykal RF, Lemmi H, Rosenthal RT, Scott-Strauss A. Characterological depressions: clinical and sleep EEG findings separating "subaffective dysthymias" from "character spectrum" disorders. *Arch Gen Psychiatry* 1980; **37**: 777-783.
- Akiskal HS, Weise RE. The clinical spectrum of so-called "minor" depressions. *Amer J Psychotherapy* 1992; **46**: 9-22.
- Albrecht J, Helderman JH, Schlessner MA, Rush AJ. A controlled study of cellular immune function in affective disorders before and after somatic therapy. *Psychiat Res* 1985; **15**: 185-193.
- Alloy LB, Abramson LY. (1988) Depressive realism: Four theoretical perspectives. In: Alloy LB (ed). *Cognitive Processes in Depression*, Guilford Press: New York, 1988. pp 223-265.
- Alloy LB, Clements CM. Illusion of control: Invulnerability to negative affect and depressive symptoms after laboratory and natural stressors. *J Abn Psychol* 1992; **2**: 234-245.
- American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*. (4th ed). American Psychiatric Association: Washington, D.C., 1994.
- American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*. (3rd ed). American Psychiatric Association: Washington, D.C., 1980.

- Anderson RL, Klein DN, Riso LP, Ouimette PC, Lizardi H, Schwartz JE. The subaffective-character spectrum subtyping distinction primary early-onset dysthymia: a clinical and familial study. *J Affect Dis* 1996; **38**:13-22.
- Andreoli AV, Keller SE, Rabaeus M, Marin P, Bartlett JA, Taban C. Depression and immunity: age, severity, and clinical course. *Brain Behav Imm* 1993; **7**: 279-292.
- Anisman H, Kokkinidis L, Borowski T, Merali Z. Differential effects of IL-1, IL-2 and IL-6 on responding for rewarding lateral hypothalamic stimulation. *Brain Research* 1998; **779**: 177-187.
- Anisman H, Merali Z. Cytokines and stress in relation to anxiety and anhedonia. In: R. Dantzer R (ed). *Cytokines, Stress and Depression*. Plenum Press: New York, 1998, in press.
- Anisman H, Merali Z. Chronic stressors and animal models of depression: distinguishing characteristics and individual profiles. *Psychopharmacology* 1997; **134**: 330-332.
- Anisman H, Merali Z, Kokkinidis L. Influence of interleukin-2 on responding for electrical brain stimulation from the hypothalamus. *Brain Research* 1996; **731**: 1-11.
- Anisman H, Sklar LS. (1979) Catecholamine depletion upon reexposure to stress: Mediation of the escape deficits produced by inescapable shock. *J Comp Physiol Psychol*. 1979; **93**: 610-625.
- Anisman H, Zacharko RM. Multiple neurochemical and behavioral consequences of stressors: Implications for depression. *Pharmacol Therap* 1989; **46**: 119-136.
- Anisman H, Zalcman S, Shanks N, Zacharko RM (1991). Multisystem regulation of performance deficits induced by stressors: An animal model of depression. In: Boulton A, Baker G, Martin-Iverson M (eds). *Neuromethods, vol. 19: Animal Models of Psychiatry, II*. Humana Press: New Jersey, 1991, pp 1-59.
- Anisman H, Zalcman S, Zacharko RM. The impact of stressors on immune and central transmitter activity: Bidirectional communication. *Rev Neurosci* 1993; **4**: 147-180.

- Antelman SM. Time-dependent sensitization as the cornerstone for a new approach to pharmacotherapy: drugs as foreign/stressful stimuli. *Drug Dev Res* 1988; **14**: 1-30.
- Antelman SM, Chiodo LA. Amphetamine as a stressor. In: Creese I (ed). *Stimulants: Neurochemical, Behavioral and Clinical Perspectives*, Raven Press: New York, 1983, pp 269-300.
- Arana GW, Baldessarini RJ, Ornstein M. The dexamethasone suppression test for diagnosis and prognosis in psychiatry. *Arch Gen Psychiat* 1985; **42**: 1193-1204.
- Asnis GM, McGinn LK, Sanderson WC. Atypical depression: Clinical aspects and noradrenergic function. *Am J Psychiat* 1995; **152**: 31-36.
- Bachen EA, Manuck SB, Marsland AL, Cohen S, Malkoff SB, Muldoon MF, Rabin BS. Lymphocyte subset and cellular immune responses to a brief experimental stressor. *Psychosom Med* 1992; **54**: 673-679.
- Bakish D, Lapierre YD, Weinstein R, Klein J, Wiens A, Jones B, Horn E, Browne M, Bourget O, Blanchard A. Ritanserin, imipramine and placebo in the treatment of dysthymic disorder. *J Clin Psychopharmacol*. 1993; **13**: 409-414.
- Baldwin D, Rudge S, Thomas S. Dysthymia: Options in Pharmacotherapy. *CNS Drugs* 1995; **4**: 422-431.
- Banki CM, Arato M, Pap Z, Kuvez M. Biochemical markers in suicidal patients: Investigations with CSF amine metabolites and neuroendocrine tests. *J Affective Disord* 1984; **6**: 341-346.
- Banki CM, Bissette G, Arato M, Nemeroff CB. Elevation of immunoreactive CSF TRH in depressed patients. *Am J Psychiatry* 1988; **145**: 1526-1531.
- Banki CM, Karmacsi L, Bissette G, Nemeroff CB. CSF corticotropin-releasing hormone and somatostatin in major depression: response to antidepressant treatment and relapse. *Eur. Neuropsychopharmacol* 1992; **2**: 107-113.

- Baroja ML, Cueppens JL. More exact quantification of interleukin-2 production by addition of anti-Tac monoclonal antibody to cultures of stimulated lymphocytes. *J Immunol Meth* 1987; 98: 267-270.
- Bartanusz V, Jezova D, Bertini LT, Tilders FJ, Aubry JM, Kiss JZ. Stress-induced increase in vasopressin and corticotropin-releasing factor expression in hypophysiotrophic paraventricular neurons. *Endocrinology* 1993; **132**: 895-902.
- Bartrop RW, Lazarus L, Luckhurst E, Kiloh LG, Penney R. Depressed lymphocyte function after bereavement. *Lancet* 1977; **1**: 834-836.
- Baumhackl U, Biziere K, Fischbach R, Geretsegger C, Hebenstreit G, Radmayr E, Stabl M. Efficacy and tolerability of moclobemide compared with imipramine in depressive disorder (DSM-III): an Austrian double-blind multicentre study. *Br J Psychiatry* 1989; **suppl 6**: 78-83.
- Bebbington PE. Life events and schizophrenia: The WHO collaborative study. *Social Psychiatry* 1987; **22**: 179-180.
- Bebbington PE, Tennant C, Hurry J. Adversity and the nature of psychiatric disorder in the community. *J Affect Dis* 1981; **3**: 345-366.
- Beck AT. *Depression: Clinical, Experimental and Theoretical Aspects*. Harper & Row: New York, 1967.
- Beck AT. *Cognitive Therapy and the Emotional Disorders*. International Universities Press: New York, 1976.
- Beck AT, Ward CH, Mendelson M, Mock J, Erbaugh J. An inventory for measuring depression. *Arch Gen Psychiat* 1961; **4**: 561-569.
- Beckham EE, Adams RL. Coping behavior in depression: Report on a new scale. *Behav Res Ther* 1984; **22**: 71-75.
- Beckham EE, Leber WR, Youll LK. The diagnostic classification of depression. In Beckham EE, WR Leber (eds). *Handbook of Depression..* Guilford Press: New York, 1995, pp 39-60.

- Benjamini E, Leskowitz S. *Immunology: A Short Course* (2nd Edition). Wiley-Liss: New York, 1991.
- Benjaminsen S. Stressful life events preceding the onset of neurotic depression. *Psychol Med* 1981; **11**: 369-378.
- Benschop RJ, Jabaaij L, Oostveen FG, Vingerhoets JJM, Kirschbaum C, Duivenvoorden HJ, Ballieux RE. Psychobiological factors related to human natural killer cell activity and hormonal modulation of NK cells in vivo. *Life Sci* 1993a; **52**: 1825-1834.
- Benschop RJ, Nieuwenhuis EES, Tromp EAM, Godaert GLR, Ballieux RE, van Doornen LJP. Effects of β -adrenergic blockade on immunologic and cardiovascular changes induced by mental stress. *Circulation* 1994; **89**: 762-769.
- Benschop RJ, Oostveen FG, Heijnen CJ, Ballieux RE. β_2 -adrenergic stimulation causes detachment of natural killer cells from cultured endothelium. *Eur J Immunol* 1993b **23**: 3242-3247.
- Bersani G, Pozzi F, Marini S, Grispi A, Pasini A, Ciani N. 5-HT₂ receptor antagonism in dysthymic disorder: a double-blind placebo-controlled study with ritanserin. *Acta Psychiatr Scand* 1991; **83**, 244-248.
- Billings AG, Moos RH. Life stressors and social resources affect posttreatment outcomes among depressed patients. *J Abn Psychol* 1985; **94**: 140-153.
- Billings AG, Moos RH. Psychological theory and research on depression: An integrative framework and review. *Clin Psychol Rev* 1982; **2**, 213-237.
- Blackburn IM, Eunson KM, Bishop S. A two year naturalistic follow-up of depressed patients treated with cognitive therapy, pharmacotherapy and a combination of both. *J Affect Dis* 1986; **10**: 67-75.
- Blalock JE. The syntax of immune-neuroendocrine communication. *Immunol Today* 1994; **15**: 504-511.
- Blalock JE. The immune system as a sensory organ. *J Immunol* 1984; **132**: 1067-1070.

- Botte L, Evrard JL, Gilles C, Stenier P, Wolfrum C. Controlled comparison of RO 11-1163 (mocobemide) and placebo in the treatment of depression. *Acta Psychiat Belg* 1992; 92: 355-369.
- Brosschot JR, Benschop RJ, Godaert GLR, de Smet MBM, Olff M, Heijnen CJ, Ballieux RE. Effects of experimental psychological stress on distribution and function of peripheral blood cells. *Psychosom Med* 1992; 54: 394-406.
- Brosschot JF, Benschop RJ, Godaert GLR, Olff M, de Smet M, Heijnen CJ, Ballieux RE. Influence of life stress on immunologic reactivity to mild psychological stress. *Psychosom Med* 1994; 56: 216-224.
- Brown AS, Gershon S. Dopamine and depression. *J Neurotrans* 1993; 91: 75-109.
- Brown GW. Life events and affective disorder: Replications and limitations. *Psychosom Med* 1993; 55: 248-259.
- Brown GW, Bifulco A, Harris TO. Life events, vulnerability and onset of depression: Some refinements. *Brit J Psychiatry* 1987; 150: 30-42.
- Brown GW, Harris TO. *Life Events and Illness*. Guilford Press: New York, 1989.
- Brown GW, Harris TO. *Social Origins of Depression: A Study of Psychiatric Disorder in Women*. Free Press: New York, 1978.
- Brown GW, Nibhrolchain M, Harris TO. Psychotic and neurotic depression. Part 3. Aetiological and background factors. *J Affect Dis* 1979; 1: 195-211.
- Bunney WE Jr, Davis JM. Norepinephrine in depressive reactions: A review. *Arch Gen Psychiatry*, 1965, 13, 483-494.
- Calabrese JR, Kling MA, Gold PW. Alterations in immunocompetence during stress, bereavement, and depression: Focus on neuroendocrine regulation. *Am J Psychiatry* 1987; 144: 1123-1134.
- Caldwell CL, Irwin M, Lohr J. Reduced natural killer cell cytotoxicity in depression but not in schizophrenia. *Biol Psychiatry* 1991; 30: 1131-1138.
- Carlson NR. *Physiology of Behavior* (3rd Edition). Allyn and Bacon: Boston, 1986.

- Carr DJJ. Neuroendocrine peptide receptors on cells of the immune system. In: Ishizaka K, Lachmann PJ, Waksman BH (eds). *Chemical Immunology* Karger: Basel, 1992, pp 84-105.
- Cassano GB, Akiskal HS, Musetti L, Perugi G, Soriani A, Mignani V. Psychopathology, temperament, and past course in primary major depression. 2. Toward a redefinition of bipolarity with a new semistructured interview for depression. *Psychopathology* 1989; **22**: 278-288.
- Cassano GB, Perugi G, Marenmani I, Akiskal HS. Social adjustment in dysthymia. In: Burton SW, Akiskal HS (eds). *Dysthymic Disorders*. Gaskell: London, 1990, pp 78-85.
- Cassens G, Roffman M, Kuruc A, Orsulak PJ, Schildkraut JJ. Alterations of brain norepinephrine metabolism induced by environmental stimuli paired with inescapable shock. *Science* 1980; **209**: 1138-1140.
- Checkley SA. Neuroendocrine mechanisms and the precipitation of depression by life events. *Br J Psychiatry* 1992; **160**: 7-17.
- Checkley S, Arendt J. Pharmacoenocrine studies of GH, PRL and melatonin in patients with affective illness. In: Brown GM, Koslow SH, Reichlin S (eds). *Neuroendocrinology in Psychiatric Disorder*. Raven Press: New York, 1984, pp 165-190.
- Cohen S, Kamarck T, Mermelstein R. A global measure of perceived stress. *J Health Soc Beh* 1983; **24**: 385-396.
- Comstock LM, Hooper EM, Goodwin JM, Goodwin JS. Physician behavior that correlates with patient satisfaction. *J Med Educ* 1982; **57**: 105-112.
- Connor TJ, Leonard BE. Depression, stress and immunological activation: The role of cytokines in depressive disorders. *Life Sciences* 1998; **62**: 583-606.
- Conte HR, Karasu TB. A review of treatment studies of minor depression: 1980-1991. *Am J Psychiatry* 1992; **46**: 58-74.

- Coppen A, Prange AJ Jr, Whybrow PC, Noguera R. Abnormalities of indolamines in affective disorders. *Arch Gen Psychiatry* 1972; **26**: 474-478.
- Corona GL, Cucchi ML, Frattini P, Santagostino G, Schinelli S, Comincioli V, Zaerbi F, Fenoglio L, Savoldi F. Noradrenergic output and clinical response in depressed women during amitriptyline therapy. *Pharmacopsychiatry* 1989; **22**: 144-151.
- Coryell W, Endicott J, Andreasen N, Keller MB, Clayton PJ, Hirschfeld RM, Scheftner WA, Winokur G. Depression and panic attacks: the significance of overlap as reflected in follow-up and family study data. *Am J Psychiatry* 1988; **145**: 293-300.
- Costa-e-Silva JA. Treatment of dysthymic disorder with low-dose amisulpride. A comparative study of 50 mg/day amisulpride versus placebo. *Ann Psychiatry* 1990; **5**: 242-249.
- Cosyns P, Maes M, Vandewoude M, Stevens WJ, De Clerck LS, Schotte C. Impaired mitogen-induced lymphocyte responses and the hypothalamic-pituitary-adrenal axis in depressive disorders. *J Affect Disord* 1989; **16**: 41-48, 1989.
- Cox A, Hopkinson K, Rutter M. Psychiatric interviewing techniques. II Naturalistic Study: Eliciting factual information. *Br J Psychiatry* 1981; **138**: 283-291.
- Cox A, Rutter M, Holbrook D. Psychiatric interviewing techniques - A second experimental study: Eliciting feelings. *Br J Psychiatry* 1988; **152**: 64-72.
- Crary B, Hauser SL, Borysenko M, Kutz I, Hoban C, Ault KA, Weiner HL, Benson H. Epinephrine-induced changes in the distribution of lymphocyte subsets in peripheral blood of humans. *J Immunol* 1983; **131**: 1178-1181.
- Crnic LS. Behavioral consequences of viral infection. In: Ader R, Felten DL, Cohen N. (eds). *Psychoneuroimmunology*. San Diego: Academic Press, 1991, pp 749-770.
- Cruse JM, Lewis RR. *Illustrated Dictionary of Immunology*. CRC Press: Boca Raton, 1995.

- Cunnick JE, Lysle DT, Armfield A, Rabin BS. Shock-induced modulation of lymphocyte responsiveness and natural killer activity: Differential mechanisms of induction. *Brain Behav Immun* 1988; **2**: 102-113.
- Cunnick JE, Lysle DT, Kucinski BJ, Rabin BS. Evidence that shock-induced immune suppression is mediated by adrenal hormones and peripheral β -adrenergic receptors. *Pharmacol Biochem Behav* 1990; **36**: 645-651.
- Dallman MF. Stress Update. Adaptation of the hypothalamic-pituitary-adrenal axis to chronic stress. *Trends Endocrinol Metab* 1993; **4**: 62-69.
- Darko DF, Gillin JC, Risch SC, Bulloch K, Golshan S, Tasevska Z, Hamburger RN. Mitogen-stimulated lymphocyte proliferation and pituitary hormones in major depression. *Biol Psychiatry* 1989; **26**: 145-155.
- Darko DF, Irwin MR, Risch SC, Gillin JC. Plasma beta-endorphin and natural killer cell activity in major depression: A preliminary study. *Psychiat Res* 1992; **43**: 111-119.
- Darko DF, Lucas AH, Gillin JC. Replication of lower lymphocyte blastogenesis in depression. *Am J Psychiatry* 1986; **143**: 1492-1493.
- Darko DF, Lucas AH, Gillin JC, Risch SC, Golshan S, Hamburger R, Silverman MB, Janowsky DS. Cellular immunity and the hypothalamic-pituitary axis in major affective disorder: a preliminary study. *Psychiatry Res* 1988; **25**: 1-9.
- Darko DF, Wilson NW, Gillin JC, Golshan S. A critical appraisal of mitogen-induced lymphocyte proliferation in depressed patients. *Am J Psychiatry* 1991; **148**: 337-344.
- Daruna JH, Morgan JE. Psychosocial effects on immune function: Neuroendocrine pathways. *Psychosomatics* 1990; **31**: 4-12.
- Davidson J, Pelton S. Forms of atypical depression and their response to antidepressant drugs. *Psychiatry Res* 1986; **17**: 87-95.
- De Groote D, Gevaert Y, Lopez M, Gathy R, Fauchet F, Dehart I, Jadoul M, Radoux D, Franchimont P. Novel method for the measurement of cytokine production by a one-stage procedure. *J Immunol Methods* 1993; **163**: 259-267.

- De Kloet ER, Reul JMH. Feedback action and tonic influence of corticosteroids on brain function: A concept arising from heterogeneity of brain receptor systems. *Psychoneuroendocrinology* 1987; **12**: 83-105.
- De Lisio G, Maremmanni I, Perugi G, Cassano GB, Deltito J, Akiskal HS. Impairment of work and leisure in depressed outpatients. *J Affect Disord* 1986; **10**: 79-84.
- Demitrack MA, Dale JK, Straus SE, Laue L, Listwak SJ, Kruesi MJP, Chrousos GP, Gold PW. Evidence for impaired activation of the hypothalamic-pituitary-adrenal axis in patients with chronic fatigue syndrome. *J Clin Endocrinol Metab* 1991; **148**: 337-344.
- Derecho CN, Wetzler S, McGinn LK, Sanderson WC, Asnis GM. Atypical depression among psychiatric inpatients: clinical features and personality traits. *J Affect Disord* 1996; **39**, 55-59.
- Deutch AY, Roth RH. The determinants of stress-induced activation of the prefrontal cortical dopamine system. In: Uylings HBM, Van Eden CG, DeBruin JPC, Corner MA, Feenstra NJP. (eds). *Prog Brain Res* 1990; **85**, Elsevier: New York, pp 357-393.
- Deutch AY, Tam SY, Roth RH. Footshock and conditioned stress increase 3,4-dihydroxyphenylacetic acid (DOPAC) in the ventral tegmental area but not substantia nigra. *Brain Research* 1985; **333**: 143-146.
- Dhabhar FS, Miller AH, McEwen BS, Spencer RL. Effects of stress on immune cell distribution: Dynamic and hormonal mechanisms. *J Immunology* 1995; **154**: 5511-5527.
- Dinan TG. Glucocorticoids and the genesis of depressive illness: A psychobiological model. *Br J Psychiat.* 1994; **164**: 365-371.
- Doherty MD, Gratton A. High-speed chronoamperometric measurements of mesolimbic and nigrostriatal dopamine release associated with repeated daily stress. *Brain Research* 1992; **586**: 295-302.

- Dohrenwend BS, Dohrenwend BP, Dodson M, Shrout PE. Symptoms, hassles, social supports and life events: A problem of confounded measures. *J Abn Psychol* 1984; **93**: 222-230.
- Donaldson SK, Klein DN, Riso LP, Schwartz JE. Comorbidity between dysthymic and major depressive disorders: a family study analysis. *J Affect Disord* 1997; **42**: 103-111.
- Dorian BJ, Garfinkel PE. Stress immunity and illness: A review. *Psychol Med* 1987; **17**: 393-407.
- Duarte A, Camozzi CR. Moclobemide versus fluoxetine in the treatment of dysthymia. *9th World Congress of Psychiatry* abstract, Rio de Janeiro, 1993
- Dunn AJ. Interactions between the nervous system and the immune system: Implications for psychopharmacology. In: Bloom FE, Kupfer DJ (eds), *Psychopharmacology: The Fourth Generation of Progress*, Raven Press: New York, 1995, pp 719-731.
- Dunn AJ. Interleukin-1 as a stimulator of hormone secretion. *Prog NeuroEndocrinImmunol* 1990; **3**: 26-34.
- Dunn AJ, Powell ML, Meitin C, Small PA. Virus infection as a stressor: Influenza virus elevates plasma concentrations of corticosterone, and brain concentrations of MHPG and tryptophan. *Physiol Behav* 1989; **45**: 591-594.
- Dura JR, Stukenberg KW, Kiecolt-Glaser JK. Chronic stress and depressive disorder in older adults. *J Abn Psychology* 1990; **99**: 284-290.
- Eisemann M. Contact difficulties and experience of loneliness in depressed patients and nonpsychiatric controls. *Acta Psychiat Scand* 1984; **70**: 160-165.
- Evans DL, Folds JD, Petitto JM, Golden RN, Pedersen CA, Corrigan M, Gilmore JH, Silva SG, Quade D, Ozer H. Circulating natural killer cell phenotypes in men and women with major depression: Relation to cytotoxic activity and severity of depression. *Arch Gen Psychiatry* 1992; **49**: 388-395.

- Evans DL, Pederson CA, Folds JD. Major depression and immunity: preliminary evidence of decreased natural killer cell populations. *Prog Neuropsychopharmacol Biol Psychiatry* 1988; **12**: 739-748.
- Fawcett J. Antidepressants: partial response in chronic depression. *Br J Psychiatry Suppl* 1994; **26**: 37-41.
- Fibiger HC. Neurobiology of depression: Focus on dopamine. In: Gessa GL, Fratta W, Pani L, Serra G (eds). *Depression and Mania: From Neurobiology to Treatment*. Lippincott-Raven: Philadelphia, 1995, pp 1-42.
- Finlay-Jones R, Brown GW. Types of stressful life events and the onset of anxiety and depressive disorders. *Psychol Med* 1981; **11**: 803-816.
- Folkman S, Lazarus RS. *Manual for the Ways of Coping Questionnaire*. Consulting Psychologists Press: Palo Alto, CA., 1988.
- Freeman HL. Historical and nosological aspects of dysthymia. *Acta Psychiat Scand* 1994; **89**: 7-11.
- Friedman RA. Social impairment in dysthymia. *Psychiat Ann* 1993; **23**, 632-637.
- Friedman RA, Markowitz JC, Parides M, Kocsis JH. Acute response of social functioning in dysthymic patients with desipramine. *J Affect Disord* 1995a; **34**: 85-88.
- Friedman RA, Mitchell J, Kocsis JH. Retreatment for relapse following desipramine discontinuation in dysthymia. *Am J Psychiatry* 1995; **152**: 926-928
- Friedman RA, Parides M, Baff R, Moran M, Kocsis JH. Predictors of response to desipramine in dysthymia. *J Clin Psychopharm* 1995b; **15**: 280-283.
- Futterman AD, Kemeny ME, Shapiro D, Fahey JL. Immunological and physiological changes associated with induced positive and negative mood. *Psychosom Med* 1994; **56**: 499-511.
- Futterman AD, Kemeny ME, Shapiro D, Fahey JL. Immunological variability associated with experimentally-induced positive and negative affective states. *Psychol Med* 1992; **54**: 133-148.

- Ghaziuddin M, Ghaziuddin N, Stein GS. Life events and the recurrence of depression. *Can J Psychiatry* 1990; **35**: 239-242.
- Glaser R, Kiecolt-Glaser JK, Bonneau RH, Malarkey W, Kennedy S, Hughes J. Stress-induced modulation of the immune response to recombinant hepatitis B vaccine. *Psychosom Med* 1992; **54**: 22-29.
- Glaser R, Kiecolt-Glaser JK, Speicher CE, Holliday JE. Stress, loneliness, and changes in herpesvirus latency. *J Behav Med* 1985; **8**: 249-260.
- Glaser R, Rice CE, Sheridan J, Fertel R, Stout J, Speicher C, Pinsky D, Kotur M, Post A, Beck M, Kiecolt-Glaser J. Stress-related immune suppression: Health implications. *Brain Behav Immun* 1987; **1**: 7-20.
- Glavin GB. Stress and brain noradrenaline: A review. *Neurosci Biobehav Rev* 1985; **9**: 233-243.
- Gold PW, Chrousos GP, Kellner C, Post RM, Roy A, Augerinas P, Schulte H, Oldfield EH, Loriaux DL. Psychiatric implications of basic and clinical studies with corticotropin-releasing factor. *Am J Psychiatry* 1984; **141**: 619-627.
- Gold PW, Licinio J, Wong ML, Chrousos GP. Corticotropin releasing hormone in pathophysiology of melancholic and atypical depression and in the mechanism of action of antidepressant drugs. *Ann NY Acad Sci* 1995; **771**: 716-729.
- Goldstein RB, Weissman MM, Adams PB, Horwath E, Lish JD, Charney D, Woods SW, Sobin C, Wickramaratne PJ. Psychiatric disorders in relatives of probands with panic disorder and/or major depression. *Arch Gen Psychiat*. 1994; **51**, 383-394.
- Goodman DW, Barnhill J. Family and genetic epidemiologic studies. In: Kocsis JH, Klein DN (eds). *Diagnosis and treatment of chronic depression..* Guilford Press: New York, 1995, pp 103-123.
- Grant I, Sweetwood HL, Yager J, Gerst M. (1981) Quality of life events in relation to psychiatric symptoms. *Arch Gen Psychiat* 1981; **38**: 335-339.

- Griffiths J, Ravindran AV, Merali Z, Anisman H. Immune and behavioral correlates of typical and atypical depression. *Soc Neurosci Abst.* 1996; **22**: 1350.
- Griffiths J, Ravindran AV, Waddell C, Anisman H. Immune status associated with dysthymia, major depression and stressful life events. *Soc Neurosci Abst.* 1992; **18**: 717.
- Guy W. ECDEU assessment manual for psychopharmacology. National Institute for Mental Health, Psychopharmacology Research Branch: Maryland, 1976.
- Hamilton M. Development of a rating scale for primary depressive illness. *Brit J Soc Clin Psychol* 1967; **6**: 278-296.
- Hamilton M. The assessment of anxiety states by rating. *Brit J Med Psychol* 1959; **32**: 50-55.
- Hammen C. Generation of stress in the course of unipolar depression. *J Abn Psychol* 1991; **100**: 555-561.
- Hammen C, Davila J, Brown G, Ellicott A, Gitlin M. Psychiatric history and stress: Predictors of severity of unipolar depression. *J Abn Psychol* 1992; **101**: 45-52.
- Hammen C, Ellicott A, Gitlin M. Vulnerability to specific life events and prediction of course of disorder in unipolar depressed patients. *Can J Behav Sci* 1989; **21**, 377-388.
- Hammen C, Mayol A, DeMayo R, Marks T. Initial symptom levels and the life-event - depression relationship. *J Abn Psychol* 1986; **95**: 114-122.
- Hanisch U-W, Quirion R. Interleukin-2 as a neuroregulatory cytokine. *Brain Res Rev* 1996; **21**: 246-284.
- Harrison W, Rabkin J, Stewart JW, McGrath PJ, Tricamo E, Quitkin F. Phenelzine for chronic depression: a study of continuation treatment . *J Clin Psychiatry* 1986; **47**, 346-349.

- Heinsbroek R, van Haaren, F, Feenstra M, van de Poll N. Changes in dopamine and noradrenaline activity in the frontal cortex produced by controllable and uncontrollable shock. *Behav Pharmacol* 1989; **1**, 61.
- Hellerstein DJ, Samstag LW, Cantillon M, Maurer M, Rosenthal J, Yanowitch P, Winston A. Follow-up assessment of medication-treated dysthymia. *Prog Neuro-Psychopharmacol Biol Psychiatry* 1996; **20**: 427-442
- Hellerstein DJ, Yanowitch P, Rosenthal J, Samstag LW, Maurer M, Dasch K, Burrows L, Poster M, Cantillon M, Winston A. A randomized double blind study of fluoxetine versus placebo in the treatment of dysthymia. *Am J Psychiatry* 1993; **150**: 1169-1175.
- Herbert TB, Cohen S. Depression and immunity: A meta-analytic review. *Psychol Bull* 1993; **113**: 472-486.
- Herman JP, Cullinan WE. Neurocircuitry of stress: Central control of hypothalamo-pituitary-adrenocortical axis. *Trends Neurosci* 1997; **20**, 78-84.
- Herman JP, Guillonneau D, Dantzer R, Scatton B, Semerdjian-Rouquier L, LeMoal M. Differential effects of inescapable footshock and stimuli previously paired with inescapable footshocks on dopamine turnover in cortical and limbic areas of the rat. *Life Sci* 1982; **30**: 2207-2214.
- Herman JP, Stinus L, Le Moal M. Repeated stress increases locomotor response to amphetamine. *Psychopharmacology* 1984; **84**: 431-435.
- Hickie I, Hickie C, Lloyd A, Silove D, Wakefield D. Impaired in vivo immune responses in patients with melancholia. *Br J Psychiatry* 1993; **162**: 651-657.
- Hirschfeld RMA. Personality and dysthymia. In: Burton SW, Akiskal HS (eds). *Dysthymic Disorders*. Gaskell: London, 1990, pp 69-77.
- Holahan CJ, Moos RH. Life stressors, personal and social resources, and depression: A 4-year structural model. *J Abn Psychol* 1991; **100**: 31-38.
- Hollon SD, Shelton RC, Loosen PT. Cognitive therapy and pharmacotherapy for depression. *J Consul Clin Psychol* 1991; **59**, 88-99.

- Holsboer F. Neuroendocrinology of Mood Disorders. In: Bloom FE, Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven Press: New York, 1995, pp 957-969.
- Howland RH. Pharmacotherapy of dysthymia: A Review. *J Clin Psychopharmacol* 1991; **11**: 83- 92.
- Howland RH, Thase ME. Biological studies of dysthymia. *Biol Psychiatry* 1991; **30**: 283-304.
- Inui TS, Carter WB, Kukull WA, Haigh VH. Outcome based doctor-patient interaction analysis. *Medical Care* 1982; **20**: 537-549.
- Irwin J, Ahluwalia P, Anisman H. Sensitization of norepinephrine activity following acute and chronic footshock. *Brain Research* 1986; **379**: 98-103.
- Irwin J, Livnat S. Behavioral influences on the immune system: Stress and conditioning. *Prog Neuropsychopharmacol Biol Psychiatry* 1987; **11**: 137-143.
- Irwin M. Psychoneuroimmunology of depression. In: Bloom FE, Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven Press: New York, 1995, pp 983-998.
- Irwin M, Brown M, Patterson T, Hauger R, Maseovich A, Grant I. Neuropeptide Y and natural killer cell activity: Findings in depression and Alzheimer caregiver stress. *FASEB* 1991; **5**: 3100-3107.
- Irwin M, Caldwell C, Smith TL, Brown S, Schuckit MA, Gillin JC. Major depressive disorder, alcoholism, and reduced natural killer cytotoxicity: Role of severity of depressive symptoms and alcohol consumption. *Arch Gen Psychiatry* 1990a; **47**: 713-719.
- Irwin M, Daniels M, Bloom E, Smith TL, Weiner H. Life events, depressive symptoms, and immune function. *Am J Psychiatry* 1987a; **144**: 437-441.
- Irwin M, Daniels M, Bloom E, Weiner H. Life events and natural killer cell activity. *Psychopharm Bull* 1986; **22**: 1093-1096.

- Irwin M, Daniels M, Smith TL, Bloom E, Weiner H. Impaired natural killer cell activity during bereavement. *Brain Behav Immun* 1987b; **1**: 98-104.
- Irwin M, Gillin JC. Impaired natural killer cell activity among depressed patients. *Psychiatr Res* 1987; **20**: 181-182.
- Irwin M, Hauger RL. Adaptation to stress: Temporal patterns of immune and neuroendocrine correlates. *Neuropsychopharmacology* 1988; **1**: 239-242.
- Irwin M, Mascovich A, Gillin JC, Willoughby R, Pike J, Smith TL. Partial sleep deprivation reduces natural killer cell activity in humans. *Psychosom Med* 1994; **56**: 493-498.
- Irwin M, Patterson T, Smith TL, Caldwell C, Brown SA, Gillin JC, Grant I. Reduction of immune function in life stress and depression. *Biol Psychiatry* 1990c; **27**: 22-30.
- Irwin M, Smith TL, Gillin JC. Electroencephalographic sleep and natural killer cell activity in depressed patients and control subjects. *Psychosom Med* 1992; **54**: 10-21.
- Jessop JJ, Gale K, Bayer BM. Enhancement of rat lymphocyte proliferation after prolonged exposure to stress. *J Neuroimmunol* 1987; **16**: 261-271.
- Jimerson DC. Role of dopamine mechanisms in the affective disorders. In: Meltzer HY (ed). *Psychopharmacology: The Third Generation of Progress*. Raven: New York, 1987, pp 505-511.
- Jimerson DC, Post RM. Psychomotor stimulants and dopamine agonists in depression. In: Post RM, Ballenger JC (eds). *Neurobiology of Mood Disorders*, Williams & Wilkins: Baltimore, 1984, pp 619-628.
- Joffe RT. A perspective on the thyroid and depression. *Can J Psychiatr* 1990; **35**: 754-758.
- Joffe RT, Bagby RM, Levit AJ. The thyroid and melancholia. *Psychiatr Res* 1992, **42**, 73-80.
- Joseph-Vanderpool JR, Rosenthal NE, Chrousos GP, Wehr TA, Skwerer R, Kasper S, Gold PW. Abnormal pituitary-adrenal responses to corticotropin-releasing hormone in

- patients with seasonal affective disorder: Clinical and pathophysiological implications. *J Clin Endocrinol Metab* 1991; **72**: 1382-1387.
- Judd LL, Akiskal HS, Paulus MP. The role and clinical significance of subsyndromal depressive symptoms (SSD) in unipolar major depressive disorder. *J Affective Disord* 1997; **45**: 5-18.
- Kalivas PW, Stewart J. Dopamine transmission in the initiation and expression of drug- and stress-induced sensitization of motor activity. *Brain Res Rev* 1991; **16**: 223-244.
- Kanner AD, Coyne JC, Schaefer C, Lazarus RS. Comparison of two modes of stress measurement: Daily hassles and uplifts versus major life events. *J Behav Med* 1981; **4**: 1-39.
- Kapur S, Mann JJ. Role of the dopaminergic system in depression. *Biol Psychiat* 1992; **32**: 1-17.
- Katschnig H, Berner P. The poly-diagnostic approach in psychiatric research. In: *Proceedings of the International Conference on Diagnosis and Classification of Mental Disorder and Alcohol and Drug Related Problems* World Health Organization: Geneva, 1984.
- Keller MB, Lavori PW, Endicott J, Coryell W, Klerman GL. Double depression: Two year follow up. *Am J Psychiatry* 1983; **140**: 689-694.
- Keller MB, Lavori PW, Mueller TI, Endicott J, Coryell W, Hirschfeld RM, Shea T. Time to recovery, chronicity, and levels of psychopathology in major depression. A 5-year prospective follow-up of 431 subjects. *Arch Gen Psychiatry* 1992; **49**: 809-816.
- Keller MB, Sessa FM. Dysthymia: Development and Clinical Course. In: Burton SW, HS Akiskal (eds). *Dysthymic Disorders*. Gaskell: London, 1990, pp 13-23.
- Keller MB, Shapiro RW. Double depression: superimposition of acute depressive episodes on chronic depressive disorders. *Am J Psychiatry* 1982; **139**: 438-442.

- Keller MB, Klein DN, Hirschfeld RMA, Docsis JH, McCullough JP, Miller I, First MB, Holzer III CP, Keiner GI, Marin Db, Shea T. Results of the DSM-IV Mood Disorders Field Trial. *Am J Psychiatry* 1995; **152**: 843-849.
- Keller MB, Klerman GL, Lavori PW, Coryell W, Endicott J, Taylor J. Long-term outcome of episodes of major depression. Clinical and public health significance. *JAMA* 1984; **252**: 788-792.
- Keller SE, Schleifer SJ, Demetrikopoulos MK. Stress-induced changes in immune function in animals: Hypothalamo-pituitary-adrenal influences. In: Ader R, Felten DL, Cohen N (eds). *Psychoneuroimmunology*. Academic: San Diego, 1991, pp 771-787.
- Kendler KS, Neale MC, Kessler RC, Heath AC, Eaves LJ. Childhood parental loss and adult psychopathology in women: A twin study perspective. *Arch Gen Psychiatr* 1992; **49**: 109-116.
- Kent S, Bluthé RM, Kelley KW, Dantzer R. Sickness behavior as a new target for drug development. *Trends Pharmacol Sci* 1992; **13**: 24-28.
- Kent, S., Bret-Dibat, J.L., Kelley, K.W., & Dantzer, R. (1996). Mechanisms of sickness-induced decreases in food-motivated behavior. *Neuroscience and Biobehavioral Reviews*, **20**, 171-175.
- Kessler RC, Price RH, Wortman CB. Social factors in psychopathology: Stress, social support and coping processes. *Ann Rev Psychol* 1985; **36**: 531-572.
- Kiecolt-Glaser JK, Cacioppo JT, Malarkey WB, Glaser R. Acute psychological stressors and short-term immune changes: What, why, for whom, and to what extent? *Psychosom Med* 1992; **54**: 680-685.
- Kiecolt-Glaser JK, Dura JR, Speicher CE, et al. Spousal caregivers of dementia victims: Longitudinal changes in immunity and health. *Psychosom Med* 1991; **53**: 345-362.
- Kiecolt-Glaser JK, Garner W, Speicher C, Penn GM, Holliday J, Glaser R. Psychosocial modifiers of immunocompetence in medical students. *Psychosom Med* 1984; **46**: 15-24.

- Kiecolt-Glaser JK, Glaser R. Stress and immune function in humans. In: Ader R, Felten DL, Cohen N (eds), *Psychoneuroimmunology*. Academic: San Diego, 1991a, pp 849-867.
- Kiecolt-Glaser JK, Glaser R. Psychosocial factors, stress, disease and immunity. In: Ader R, Felten DL, Cohen N (eds), *Psychoneuroimmunology*. Academic: San Diego, 1991b, pp 868-885.
- Kirschbaum C, Hellhammer DH. Salivary cortisol in psychoneuroendocrine research: recent developments and applications. *Psychoneuroendocrinology* 1994; **19**: 313-333.
- Kirschbaum C, Pruessner JC, Stone AA, Federenko I, Gaab J, Lintz D, Schommer N, Hellhammer DH. Persistent high cortisol responses to repeated psychological stress in a subpopulation of healthy men. *Psychosom Med* 1995; **57**: 468-474.
- Klein DN, Riso LP, Donaldson SK, Schwartz JE, Anderson RL, Ouimette PL, Lizardi H, Aronson TA. Family study of early-onset dysthymia. Mood and personality disorders in relatives of outpatients with dysthymia and episodic major depression and normal controls. *Arch Gen Psychiatry* 1995; **52**: 487-496.
- Klein DN, Taylor EB, Dickstein S, Harding K. Primary early-onset dysthymia: comparison with primary non-bipolar non-chronic major depression on demographic, clinical, familial, personality, and socioenvironmental characteristics and short-term outcome. *J Abn Psychol* 1988a; **97**: 387-398.
- Klein DN, Taylor EB, Dickstein S, Harding K. The early-late onset distinction in DSM-III-R dysthymia. *J Affect Disord* 1988b; **14**: 25-33.
- Knapp PH, Levy EM, Giorgi RG, Black PH, Fox BH, Heeren TC. Short-term immunological effects of induced emotion. *Psychosom Med* 1992; **54**: 133-148.
- Kocsis JH, Frances AJ. A critical discussion of DSM-III dysthymic disorder. *Am J Psychiatry* 1987; **144**, 1534-1542.
- Kocsis JH, Frances AJ, Voss C, Mann JJ, Mason BJ, Sweeney J. Imipramine treatment for chronic depression. *Arch Gen Psychiatry* 1988; **45**, 253-257.

- Kocsis JH, Friedman RA, Markowitz JC, Leon AC, Miller NL, Gniwesch L, Parides M. Maintenance therapy for chronic depression. A controlled clinical trial of desipramine. *Arch Gen Psychiatry* 1996; **53**: 769-774.
- Kocsis JH, Sutton BM, Frances AJ. Long-term follow-up of chronic depression treated with imipramine. *J Clin Psychiatry* 1991; **52**: 56-59.
- Kocsis JH, Thase M, Koran L, Halbreich U, Yonkers K. Pharmacotherapy of pure dysthymia: Sertraline vs. imipramine and placebo. *Eur Neuropsychopharm.* 1994; **4** (suppl 3): S204.
- Kocsis JH, Zisook S, Davidson J, Shelton R, Yonkers K, Hellerstein DJ, Rosenbaum J, Halbreich U. Double-blind comparison of sertraline, imipramine, and placebo in the treatment of dysthymia: Psychosocial outcomes. *Am J Psychiatry* 1997; **154**: 390-395.
- Kovacs M, Feinberg TL, Crouse-Novak MA, Paulauskas SL, Finkelstein R. Depressive disorders in childhood, I: a longitudinal prospective study of characteristics and recovery. *Arch Gen Psychiatry* 1984; **41**: 229-237.
- Kronfol Z, House JD. Depression, cortisol, and immune function. *Lancet* 1984; **1**: 1026-1027.
- Kronfol Z, Nair M, Goodson J, Goel K, Haskett R, Schwartz S. Natural killer cell activity in depressive illness: Preliminary report. *Biol Psychiatry* 1989; **26**: 753-756.
- Kronfol Z, Sylva J, Greden J, Dembinski S, Carroll BJ. Cell-mediated immunity in melancholia. *Psychosom Med* 1982; **44**: 304-307.
- Kronfol Z, Sylva J, Greden J, Dembinski S, Gardner R, Carroll B. Impaired lymphocyte function in depressive illness. *Life Sciences* 1983; **33**: 241-247.
- Krueger RB, Levy EM, Cathcart ES. Lymphocyte subsets in patients with major depression: Preliminary findings. *Advances*, 1984; **1**: 5-9.
- Kvetnansky R. Recent progress in catecholamines under stress. In: Usdin E, Kvetnansky R, Kopin IJ (eds). *Catecholamines and Stress: Recent Advances*. New York: Elsevier. 1980; pp 7-20.

- Lacosta S, Merali Z, Anisman H. Influence of interleukin-1 on exploratory behaviors, plasma ACTH and cortisol, and central biogenic amines in mice. *Psychopharmacology* 1998; 137: 351-361.
- Landmann RMA, Muller FB, Perini CH, Wesp M, Erne P, Buhler FR. Changes in immunoregulatory cells induced by psychological and physical stress: Relationship to plasma catecholamines. *Clin Exp Immunol* 1984; 58: 127-135.
- Lazarus RS. Coping theory and research: Past, present, and future. *Psychosom Med* 1993; 55: 234-247
- Lazarus RS. Theory-based stress measurement and commentaries. *Psychol Inquiry* 1990; 1: 3-51.
- Lazarus RS, Folkman S. *Stress, Appraisal, and Coping*. Springer: New York, 1984.
- Leader JB, Klein DN. Social adjustment in dysthymia, double depression and episodic major depression. *J Affect Dis* 1996; 37: 91-101.
- Leahy RL, Beck AT. Cognitive therapy of depression and mania. In: Georgotas A, Cancro R (eds). *Depression and Mania* 1988, pp 517-537.
- Lechin F, van der Dijs B, Orozco B, Lechin AE. Plasma neurotransmitters, blood pressure, and heart rate during supine resting, orthostasis, and moderate exercise in dysthymic depressed patients. *Biol Psychiatry* 1994; 37: 884-891.
- Lecrubier Y, Boyer P, Turjanski S, Rein W, the Amisulpride Study Group. Amisulpride versus imipramine and placebo in dysthymia and major depression. *J Affect Dis* 1997; 43: 95-103.
- Lecrubier Y, Guelfi JD. Efficacy of reversible inhibitors of monoamine oxidase-A in various forms of depression. *Acta Psychiatr Scand* 1990; 82: 18-23.
- Leon CA, Vigoya J, Conde S, Campo G, Castrillon E, Leon A. Comparison of the effect of amisulpride and viloxazine in the treatment of dysthymia. *Acta Psiquiatr Psicol Am Lat* 1994; 40: 41-49.

- Levitan RD, Kaplan AS, Brown GG. Atypical depression associated with low cortisol bulimia nervosa. *Am Psychiat Assoc Conference*, 1996, San Diego.
- Levitan RD, Kaplan AS, Brown GM, Joffe RT, Levitt AJ, Vaccarino FJ, Kennedy SH. Low plasma cortisol in bulimia nervosa patients with reversed neurovegetative symptoms of depression. *Biol Psychiat* 1997; **41**: 366-368.
- Levitt AJ, Joffe RT, Sokolov STH. Does the chronological relationship between the onset of dysthymia and major depression influence subsequent response to antidepressants? *J Affect Dis*. 1998; **47**: 169-175.
- Levy EM, Borrelli DJ, Mirin SM, Salt P, Knapp PH, Peirce C, Fox BH, Black PH. Biological measures and cellular immunological function in depressed psychiatric patients. *Psychiat Res* 1991; **36**: 157-167.
- Levy SM, Fernstrom J, Herberman RB, Whiteside L, Lee J, Ward M, Massoudi M. Persistently low natural killer cell activity and circulating levels of plasma beta-endorphin: Risk factors for infectious disease. *Life Sciences* 1991; **48**: 107-116.
- Linn MW, Linn BS, Jensen J. Stressful events, dysphoric mood and immune responsiveness. *Psychol Rev* 1984; **54**: 219-222.
- Locke S, Kraus L, Kutz I. Altered natural killer activity during norepinephrine infusion in humans. In: Spector NH (ed). *Neuroimmunomodulation: Proceedings of the First International Workshop on Neuroimmuno modulation*. Bethesda, 1984, pp 297-312.
- Lopez Ibor JJ, Frances A, Jones C. Dysthymic disorder: a comparison of DSM-IV and ICD-10 and issues in differential diagnosis. *Acta Psychiatr Scand* 1994; **89** (suppl 383), 12-18.
- Lora A, Fava E. Provoking agents, vulnerability factors and depression in an Italian setting: A replication of Brown and Harris's model. *J Affect Disord* 1992; **24**: 227-235.
- Lupien SJ, Gaudreau S, Tchiteya BM, Maheu F, Sharma S, Nair NPV, Hauger RL, McEwen BS, Meaney MJ. Stress-induced declarative memory impairment in health

- elderly subjects: Relationship to cortisol reactivity. *J Clin Endocrinol Metab* 1997; **82**, 2070-2075.
- Lyons JM, Eisen SA, Goldberg J, True W, Lin N, Meyer JM, Toomey R, Faraone SV, Merla-Ramos M, Tsuang MT. A registry-based twin study of depression in men. *Arch Gen Psychiatry* 1998; **55**: 468-472.
- Maas JW. Biogenic amines and depression. *Arch Gen Psychiatry* 1975; **32**: 1357-1361.
- Maes M. The inflammatory response system in major depression. In: Dantzer R (ed). *Cytokines, stress and depression*. Plenum Press: New York, 1998, in press.
- Maes M. Evidence for an immune response in major depression: A review and hypothesis. *Prog Neuropsychopharmacol Biol Psychiat* 1995; **19**: 11-38.
- Maes M, Bosmans E, Meltzer HY, Scharpe S, Suy E. Interleukin-1 β : A putative mediator of HPA axis hyperactivity in major depression? *Am J Psychiatry* 1993a; **150**: 1189-1193.
- Maes M, Bosmans E, Suy E, Minner B, Raus J. Immune cell parameters in severely depressed patients: negative findings. *J Affect Disord* 1989; **17**: 121-128.
- Maes M, Bosmans E, Suy E, Vandervorst C, de Jonckheere C, Minner B, Raus J. Depression-related disturbances in mitogen-induced lymphocyte responses and interleukin-1 β and soluble interleukin-2 receptor production. *Acta Psychiatr Scand* 1991; **84**: 379-386.
- Maes M, Lambrechts J, Bosmans E, Jacobs J, Suy E, Vandervorst C, DeJonckheere C, Minner B, Raus J. Evidence for a systemic immune activation during depression: Results of leukocyte enumeration by flow cytometry in conjunction with monoclonal antibody staining. *Psychol Med* 1992a; **22**, 45-53.
- Maes M, Lambrechts J, Suy E, Vandervorst C, Bosmans E. Absolute number and percentage of circulating natural killer, non-MHC-restricted T cytotoxic, and phagocytic cells in unipolar depression. *Neuropsychobiology* 1994a; **29**:157-163.

- Maes M, Meltzer HY. The serotonin hypothesis of major depression. In: Bloom FE, Kupfer DJ (eds), *Psychopharmacology: The Fourth Generation of Progress*, Raven Press: New York, 1995, pp 933-944.
- Maes M, Meltzer HY, Bosmans E, Bergmans R, Vandoolaeghe E, Ranjan R, Desnyder R. Increased plasma concentrations of interleukin-6, soluble interleukin-6, soluble interleukin-2 and transferrin receptor in major depression. *J Affect Disord* 1995; **34**: 301-309.
- Maes M, Meltzer HY, Cosyns P, Calabrese J, D'Hondt P, Blockx P. Adrenocorticotrophic hormone, beta-endorphin and cortisol responses to oCRH in unipolar depressed patients pretreated with dexamethasone. *Prog Neuropsychopharmacol Biol Psychiatry* 1994b; **18**: 1273-1292.
- Maes M, Meltzer HY, Stevens W, Calabrese J, Cosyns P. Natural killer cell activity in major depression: Relation to circulating natural killer cells, cellular indices of the immune response, and depressive phenomenology. *Prog Neuro Psychopharmacol Biol Psychiat* 1994c; **18**: 717-730.
- Maes M, Meltzer HY, Stevens W, Cosyns P, Blockx P. Multiple reciprocal relationships between in vivo cellular immunity and hypothalamic-pituitary-adrenal axis in depression. *Psychol Med* 1994df; **24**: 167-177.
- Maes M, Stevens WJ, Peeters D, DeClerck LS, Bridts CH, Peeters D, Schotte C, Cosyns P. A significantly increased expression of T cell activation markers (interleukin-2 and HLA-DR) in depression: Additional evidence for an inflammatory process during that illness. *Prog Psychopharmacol Biol Psychiat* 1993b; **17**: 214-255.
- Maes M, Stevens W, Peeters D, DeClerck L, Scharpe S, Bridts C, Schotte C, Cosyns P. A study on the blunted natural killer cell activity in severely depressed patients. *Life Sci* 1992b; **50**: 505-513.

- Magliozzi JR, Gietzen D, Maddock RJ, Haack D, Doran AR, Goodman T, Weiler PG. Lymphocyte beta-adrenoreceptor density in patients with unipolar depression and normal controls. *Biol Psychiatr* 1989; 26: 15-25.
- Maier W, Benkert O. Treatment of chronic depression with sulpiride: evidence of efficacy in placebo-controlled single case studies. *Psychopharmacology* 1994; 115: 495-501.
- Malarkey WB, Peral DK, Demers LM, Kiecolt-Glaser JK, Glaser R. Influence of academic stress and season on 24-hour mean concentrations of ACTH, cortisol, and β -endorphin. *Psychoneuroendocrinology* 1995; 20: 499-508.
- Mann JJ, Arango V, Underwood MD. Serotonin and suicidal behavior. *Ann NY Acad Sci* 1990; 600: 476-485.
- Manuck SB, Cohen S, Rabin BS, Muldoon, MF, Bachen EA. Individual differences in cellular immune response to stress. *Psychol Sci* 1991; 2: 111-115.
- Marin DB, Kocsis JH, Frances AJ, Parides M. Desipramine for the treatment of "pure" dysthymia versus "double" depression. *Am J Psychiatry* 1994; 151: 1079-1080.
- Markowitz JC. Psychotherapy of dysthymia. *Am J Psychiat* 1994; 151: 1114-1121.
- Markowitz JC, Friedman RA, Miller N, Spielman LA, Moran ME, Kocsis JH. Interpersonal improvement in chronically depressed patients treated with desipramine. *J Affect Dis* 1996; 41: 59-62.
- Marsland AL, Manuck SB, Fazzari TV, Stewart CJ, Rabin BS. Stability of individual differences in cellular immune responses to acute psychological stress. *Psychosom Med* 1995; 57: 295-298.
- McCullough JP, Kornstein SG, McCullough JP, Belyea-Caldwell S, Kaye AL, Roberts C, Plybon JK, Kruus LK. Differential diagnosis of chronic depressive disorders. *Psychiatr Clin North Am* 1996; 19: 41-53.
- McEwen BS. The plasticity of the hippocampus is the reason for its vulnerability. *Sem Neurosci* 1994; 6: 239-246.

- McGinn LK, Asnis GM, Robinson E. Biological and clinical validation of atypical depression. *Psychiat Res* 1996; **60**: 191-198.
- McNaughton ME, Patterson TL, Irwin MR, Grant, I. The relationship of life adversity, social support, and coping to hospitalization with major depression. *J Nerv Ment Dis* 1992; **180**: 491-497.
- Meador-Woodruff JH, Gurguis G, Grunhaus L, Haskett RF, Greden JF. Multiple depressive episodes and plasma postdexamethasone cortisol levels. *Biol Psychiatry* 1987; **22**: 583-592.
- Meltzer HY, Lowy MT. The serotonin hypothesis of depression. In: Meltzer HY (ed). *Psychopharmacology: The Third Generation of Progress*. Raven Press: New York, 1987, pp 513-26.
- Meyers CA, Valentine AD. Neurological and psychiatric adverse effects of immunological therapy. *CNS Drugs* 1995; **3**: 56-68.
- Miller AH, Spencer RL, McEwen BS, Stein M. Depression, adrenal steroids, and the immune system. *Ann Med* 1993; **25**: 481-487.
- Mills PJ, Ziegler MG, Dimsdale JE, Parry BL. Enumerative immune changes following acute stress: Effect of the menstrual cycle. *Brain Behav Immun* 1995; **9**: 190-195.
- Mohl PC, Huang L, Bowden C, Fischbach M, Vogtsberger K, Talal N. Natural killer cell activity in major depression. *Am J Psychiatry* 1987; **144**: 1619.
- Moldofsky H. Sleep and the immune system. *Int J Immunopharmac* 1995; **17**: 649-654.
- Molina VA, Volosin M, Cancela L, Keller E, Murua VS, Basso AM. Effect of chronic variable stress on monoamine receptors: Influence of imipramine administration. *Pharmacol Biochem Behav* 1990; **35**: 335-340.
- Monroe SM, Bellack AS, Hersen M, Himmelhoch JM. Life events, symptom course, and treatment outcome in unipolar depressed women. *J Consult Clin Psychol* 1983; **51**: 604-615.

- Monroe SM, Depue RA. Life stress and depression. In: Psychosocial Aspects of Depression, J. Becker and A. Kleinman (eds), Erlbaum: Hillsdale, 1991, pp 101-130.
- Monroe SM, Kupfer DJ, Frank E. Life stress and treatment course of recurrent depression: 1. Response during index episode. *J Consult Clin Psychol* 1992; **5**: 718-724.
- Monroe SM, Simons AD. Diathesis-stress theories in the context of life stress research: Implications for the depressive disorders. *Psychol Bull* 1991; **110**: 406-425.
- Monroe SM, Simons AD, Thase ME. Onset of depression and time to treatment entry: Roles of life stress. *J Consult Clin Psychol* 1991; **59**: 566-573.
- Monroe SM, Thase ME, Hersen M, Himmelhoch JM, Bellack AS. Life events and the endogenous - non-endogenous distinction in the treatment and posttreatment course of depression. *Comp Psychiatry* 1985; **26**: 175-186.
- Montgomery SA, Asberg M. A new depression scale designed to be sensitive to change. *Br J Psychiatry* 1979; **134**: 382-389.
- Mullar N, Ackenheil M. Psychoneuroimmunology and the cytokine action in the CNS: implications for psychiatric disorders. *Prog Neuropsychopharm Biol Psychiatry* 1998; **22**: 1-33.
- Munck A, Guyre PM. Glucocorticoids and immune function. In: Ader R, Felten DL, Cohen N. (eds). *Psychoneuroimmunology*. Academic Press: New York, 1991. pp 447-462.
- Munck A, Guyre PM, Holbrook NJ. Physiological functions of glucocorticoids in stress and their relation to pharmacological actions. *Endocr Rev* 1984; **5**: 25-44.
- Naliboff BD, Benton D, Solomon GF, Morley JE, Fahey JL, Bloom ET, Makinodan T, Gilmore SL. Immunological changes in young and old adults during brief laboratory stress. *Psychosom Med* 1991; **53**: 121-132.
- Naliboff BD, Solomon GF, Gilmore SL, Fahey JL, Benton D, Pine J. Rapid changes in cellular immunity following a confrontational role-play stressor. *Brain Behav Immun* 1995; **9**: 207-219.

- Nassberger L, Traskman-Bendz L. Increased soluble interleukin-2 receptor concentrations in suicide attempters. *Acta Psychiatr Scand* 1993; **88**: 48-52.
- Needles DJ, Abramson LY. Positive life events, attributional style and hopefulness: Testing a model of recovery from depression. *J Abn Psychol* 1990; **99**: 156-165.
- Nemeroff CB. The corticotropin-releasing factor (CRF) hypothesis of depression: New findings and new directions. *Mol Psychiatry* 1996; **1**: 336-342.
- Nemeroff CB, Evans DL. Correlation between the dexamethasone suppression test in depressed patients and clinical response. *Am J Psychiatry* 1984; **141**: 247-249.
- Nerozzi D, Santoni A, Bersani G, Magnani A, Bressan A, Pasini A, Antonozzi I, Frajese G. Reduced natural killer cell activity in major depression: Neuroendocrine implications. *Psychoneuroendocrinology* 1989; **14**: 295-302.
- Nisenbaum LK, Abercrombie ED. Enhanced tyrosine hydroxylation in hippocampus of chronically stressed rats upon exposure to a novel stressor. *J Neurochem* 1992; **58**, 276-281.
- Nisenbaum LK, Zigmond MJ, Sved AF, Abercrombie ED. Prior exposure to chronic stress results in enhanced synthesis and release of hippocampal norepinephrine in response to a novel stressor. *J Neurosci* 1991; **11**: 1478-1484.
- O'Leary A. Stress, emotion and human immune function. *Psychol Bull* 1990; **108**: 363-382.
- O'Neil MK, Lancee WJ, Freeman SJ. Loss and depression: A controversial link. *J Nerv Ment Dis* 1987; **175**: 354-357.
- Ottaway CA, Husband AJ. Central nervous system influences on lymphocyte migration. *Brain Behav Immun* 1992; **6**: 97-116.
- Overholser JC. Emotional reliance and social loss: Effects on depressive symptomatology. *J Pers Asses* 1990; **55**: 618-629.
- Overholser JC, Norman WH, Miller IW. Life stress and social supports in depressed inpatients. *Behav Med* 1990; **16**: 125-132.

- Parker GB, Brown LB. Coping behaviors that mediate between life events and depression. *Arch Gen Psychiat* 1982; **39**: 1386-1391.
- Parker G, Brown L, Blignault I. Coping behaviors as predictors of the course of clinical depression. *Arch Gen Psychiat* 1986; **43**: 561-565.
- Parry G, Shapiro DA. Social support and life events in working class women. Stress buffering or independent effects? *Arch Gen Psychiatr* 1986; **43**: 315-323.
- Paykel ES, Prusoff BA, Uhlenhuth EH. Scaling of life events. *Arch Gen Psychiatry* 1971; **25**: 340-347.
- Pepper CM, Klein DN, Anderson RL, Riso LP, Crosby Ouimette P, Lizardi H. DSM-III-R axis II comorbidity in dysthymia and major depression. *Am J Psychiatry* 1995; **152**: 239-247.
- Perris, H. Life events and depression. Part 2. Results in diagnostic subgroups, and in relation to the recurrence of depression. *J Affect Disord* 1984; **7**: 25-36.
- Petty F, Sherman A. A neurochemical differentiation between exposure to stress and the development of learned helplessness. *Drug Devel Res* 1982; **2**: 43-45.
- Petursson H. Studies of reversible and selective inhibitors of monoamine oxidase A in dysthymia. *Acta Psychiatr Scand* 1995; **91**(suppl 386): 36-39.
- Pini S, Cassano GB, Simonini E, Savino M, Russo A, Montgomery SA. Prevalence of anxiety disorders comorbidity in bipolar depression, unipolar depression and dysthymial. *J Affect Dis* 1997; **42**: 145-153.
- Plotsky PM, Owens MJ, Nemeroff CB. Neuropeptide alterations in mood disorders. In: Bloom FE, Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven Press: New York, 1995, pp 971-981.
- Post RM. Transduction of psychosocial stress into the neurobiology of recurrent affective disorder. *Amer J Psychiat* 1992; **149**: 999-1010.

- Post RM, Rubinow DR, Ballenger JC. Conditioning, sensitization, and kindling: Implications for the course of affective illness. In: Post RM, Ballenger JC (eds). *Neurobiology of Mood Disorders*. Williams & Wilkins: Baltimore, 1984.
- Post RM, Weiss SRB. The neurobiology of treatment-resistant mood disorders. In Bloom FE, Kupfer DJ (eds), *Psychopharmacology: The Fourth Generation of Progress*, New York, Raven Press, 1995, pp. 1155-1170.
- Prange AJ, Loosen PT, Wilson IC, Lipton MA. The therapeutic use of hormones of the thyroid axis in depression. In: Post RM, Ballenger JC. *Neurobiology of Mood Disorders*. Williams & Wilkins: Baltimore, 1984, pp 311-322.
- Pruessner JC, Wolf OT, Hellhammer DH, Buske-Kirschbaum A, von Auer K, Jobst S, Kaspers F, Kirschbaum C. Free cortisol levels after awakening: A reliable biological marker for the assessment of adrenocortical activity. *Life Sci* 1997; **61**: 2539-2549
- Price JS. Change or homeostasis? A systems theory approach to depression. *Br J Med Psychol* 1991; **64**: 331-344.
- Quitkin FM, Harrison WM, Stewart JW, McGrath PJ, Tricamo E, Ocepek-Welikson K, Rabkin JJ, Wager SG, Nunes E, Klein DF. Response to phenelzine and imipramine in placebo responders with atypical depression. *Arch Gen Psychiatry* 1991; **48**: 319-323.
- Quitkin FM, Stewart JW, McGrath PJ, Liebowitz MR, Harrison WM, Tricamo E, Klein DF, Rabkin JJ, Markowitz JS, Wager SG. Phenelzine vs imipramine in the treatment of probable atypical depression: Defining syndrome boundaries of selective MAOI responders. *Am J Psychiatry* 1988; **145**: 306-311.
- Rabin BS, Cohen S, Ganguli R, Lysle DT, Cunnick JE. Bidirectional interaction between the central nervous system and the immune system. *Crit Rev Immunol* 1989; **9**: 279-312.
- Ravicki DA, Turner R, Brown R, Martindale JJ. Battelle Quality of Life Scale. *Qual Life Res* 1992; **1**: 257-266.

- Ravindran AV, Anisman H, Merali Z, Charbonneau Y, Telner J, Bialik RJ, Wiens A, Ellis J, Griffiths J. Treatment of primary dysthymia with group cognitive therapy and pharmacotherapy: clinical symptoms and functional impairments. *Am J Psychiatry*, submitted.
- Ravindran AV, Bialik RJ, Brown GM, Lapierre YD. Primary early onset dysthymia, biochemical correlates of the therapeutic response to fluoxetine: II. Urinary metabolites of serotonin, norepinephrine, epinephrine and melatonin. *J Affect Disord* 1994b; **31**: 119-123.
- Ravindran AV, Bialik RJ, Lapierre YD. Primary early onset dysthymia, biochemical correlates of the therapeutic response to fluoxetine: 1. Platelet monoamine oxidase and the dexamethasone suppression test. *J Affect Dis* 1994a; **31**: 111-117.
- Ravindran AV, Bialik RJ, Lapierre YD. Therapeutic efficacy of specific serotonin reuptake inhibitors (SSRIs) in dysthymia. *Canad J Psychiatry* 1994c; **39**: 21-26.
- Ravindran, AV, Griffiths J, Merali Z, Anisman H. Primary dysthymia: A study of several psychosocial, endocrine and immune correlates. *J Affect Dis* 1996; **40**: 73-84.
- Ravindran AV, Griffiths J, Waddell C, Lapierre YD, Anisman H. Major depression and dysthymia following serotonin reuptake blockade: Modification of stressor perception and coping style. *Soc Neurosci Abst* 1993; **19**: 1884.
- Ravindran AV, Merali Z, Anisman H. Dysthymia: A Biological Perspective. In: Licinio J, Bolis CL, Gold P (eds). *Dysthymia: from clinical neuroscience to treatment..* World Health Organization: Geneva, 1997, pp 21-44.
- Regier DA, Boyd JH, Burke JD, Rae DS, Myers JK, Kramer M, Robins LN, George LK, Karno M, Locke B. One-month prevalence of mental disorders in the United States. *Arch Gen Psychiatry* 1988; **45**: 977-986.

- Regier DA, Narrow WE, Rae DS, Manderscheid RW, Locke BZ, Goodwin FK. The de facto U.S. mental and addictive disorders service system: Epidemiologic Catchment Area prospective 1-year prevalence rates of disorders and services. *Arch Gen Psychiatry* 1993; **50**: 85-94.
- Remick RA, Sadovnick AD, Lam RW, Zis AP, Yee IML. Major depression, minor depression, and double depression: Are they distinct clinical entities. *Am J Med Gen* 1996; **67**: 347-353
- Reno RM, Hillaris AE. The relationship between life stress and depression in an endogenous sample. *Compr Psychiatry* 1990; **31**: 25-33.
- Reynolds CF, Frank E, Kupfer DJ, Thase ME, Perel JM, Mazumdar S, Houck PR. Treatment outcome in recurrent major depression: a post hoc comparison of elderly ("young old") and midlife patients. *Am J Psychiatry* 1996; **153**: 1288-1292.
- Riso LP, Klein DN, Ferro T, Kasch KL, Pepper CM, Schwartz JE, Aronson TA. Understanding the comorbidity between early-onset dysthymia and cluster B personality disorders: a family study. *Am J Psychiatry* 1996; **153**: 900-906.
- Ritter S, Pelzer NL. Magnitude of stress induced norepinephrine depletion varies with age. *Brain Research* 1978; **152**: 1701-1705.
- Rivier C. Effect of peripheral and central cytokines on the hypothalamic-pituitary-adrenal axis of the rat, *Ann NY Acad Sci* 1993; **697**: 97-105.
- Robins CJ. Congruence of personality and life events in depression. *J Abn Psychol* 1990; **99**: 393-397.
- Robins CJ, Block P. Personal vulnerability, life events, and depressive symptoms: A test of a specific interactional model. *J Pers Soc Psychol* 1988; **54**: 847-852.
- Roth RH, Tam S-Y, Ida Y, Yang J-X, Deutch Y. Stress and mesocorticolimbic dopamine system. *Ann NY Acad Sci* 1988; **537**: 138-147.
- Roy A. Early parental separation and adult depression. *Arch Gen Psychiatr* 1985; **42**: 987-991.

- Roy A. Early parental death and adult depression. *Psychol Med* 1983; **13**: 861-865.
- Roy-Burne PP, Vitaliano PP, Cowley DS, Luciano G, Zheng Y, Dunner DL. Coping in panic and major depressive disorder: Relative effects of symptom severity and diagnostic comorbidity. *J Nerv Ment Dis* 1992; **180**: 179-183.
- Rubin RT, Poland RE. The dexamethasone suppression test in depression: Advantages and limitation. In Burrows GD, Norman TR, McGuire KP (eds). *Biological Psychiatry: Recent Studies*. Libbey: London, 1984, pp 76-83.
- Russell D, Peplau LA, Cutrona CE. The revised UCLA Loneliness Scale: Concurrent and discriminant validity evidence. *J Pers Soc Psychol* 1980; **39**: 472-480.
- Safran JD, Segal ZV. *Interpersonal Process in Cognitive Therapy*. Basic Books: New York, 1990.
- Sansone RA, Wiederman MW, Sansone LA, Touchet B. Early-onset dysthymia and personality disturbance among patients in a primary care setting. *J Nerv Ment Dis* 1998; **186**: 57-58.
- Schatzberg AF, Schildkraut, JJ. Recent studies on norepinephrine systems on mood disorders. In: Bloom FE, Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven: New York, 1995, pp 911-920.
- Schedlowski M, Falk A, Rohne A, Wagner TO, Jacobs R, Tewes U, Schmidt RE. Catecholamines induce alterations of distribution and activity of human natural killer (NK) cells. *J Clin Immunol* 13: 344-351, 1993a.
- Schedlowski M, Jacobs R, Stratmann G, Richter S, Hadicke A, Tewes U, Wagner TO, Schmidt RE. Changes of natural killer cells during acute psychological stress. *J Clin Immunol* 1993b; **13**: 119-126.
- Schildkraut JJ. Current status of the catecholamine hypothesis of affective disorders. In: Lipton MA, DiMascio A, Killam KF (eds). *Psychopharmacology: A Generation of Progress*. Raven Press: New York, 1978, pp 1223-1234.

- Schildkraut JJ. The catecholamine hypothesis of affective disorders: A review of supporting evidence. *Am J Psychiatry* 1965; **122**: 509-522.
- Schleifer SJ, Keller SE, Bond RN, Cohen J, Stein M. Major depressive disorder and immunity: Role of age, sex, severity and hospitalization. *Arch Gen Psychiatry* 1989; **46**: 81-87.
- Schleifer SJ, Keller SE, Camerino M, Thornton JC, Stein M. Suppression of lymphocyte stimulation following bereavement. *JAMA* 1983; **250**: 374-377.
- Schleifer SJ, Keller SE, Meyerson AT, Raskin, MJ, Davis KL, Stein M. Lymphocyte function in major depressive disorder. *Arch Gen Psychiatry* 1984; **41**: 484-486.
- Schleifer SJ, Keller SE, Siris SG, Davis KL, Stein M. Depression and immunity. *Arch Gen Psychiatry* 1985; **42**: 129-133.
- Seegal RF, Brosch KO, Bush B. High-performance liquid chromatography of biogenic amines and metabolites in brain, cerebrospinal fluid, urine and plasma. *J Chromatog* 1986; **377**: 141-144.
- Segal ZV, Shaw BF, Vella DD, Katz R. Cognitive and life stress predictors of relapse in remitted unipolar depressed patients: Test of the congruency hypothesis. *J Abn Psychol* 1992; **101**: 26-36.
- Seidel A, Arolt V, Huntstiger M, Rink L, Behnisch A, Kirchner H. Increased CD56+ natural killer cells and related cytokines in major depression. *Clin Immunol Immunopathol* 1996; **78**: 83-85.
- Seligman MEP. *Helplessness: On depression, development and death*. Freeman: San Francisco, 1975.
- Sengar DPS, Waters BGH, Dunne JV, Bouer IN. Lymphocyte subpopulations and mitogenic responses of lymphocytes in manic depressive disorders. *Biol Psychiatry* 1982; **17**: 1017-1022.
- Sheehan D, Lecrubier Y, Janavs J, Knapp E, Weiller E, Sheehan M, Lepine JP, Teherani M. Mini-international Neuropsychiatric Interview, Clinician Rated, version 2.1, 1992.

- Shelton RC, Davidson J, Yonkers KA, Kora L, Thase ME, Pearlstein T, Halbreich U. The undertreatment of dysthymia. *J Clin Psychiatry* 1997; **58**: 59-65.
- Siever LJ. Role of noradrenergic mechanisms in the etiology of the affective disorders. In: Meltzer HY (ed). *Psychopharmacology: The Third Generation of Progress*, Raven: New York, 1987, pp. 493-504.
- Slater E, Cowie V. *The genetics of mental disorder*. Oxford University Press: London, 1971.
- Sluzewska A, Rybakowski JK, Laciak M, Mackiewicz A, Sobieska M, Wiktorowicz K. Interleukin-6 serum levels in depressed patients before and after treatment with fluoxetine. *Ann NY Acad Sci* 1995; **762**: 474-476.
- Smeraldi E. Amisulpride versus fluoxetine in patients with dysthymia or major depression in partial remission. A double-blind, comparative study. *J Affect Dis* 1998; **48**, 47-56.
- Smith RS. The macrophage theory of depression. *Med Hypoth* 1991; **35**: 298-306.
- Smyth JM, Ockenfels MC, Gorin AA, Catley D, Porter LS, Kirschbaum C, Hellhammer DH, Stone AA. (1997) Individual differences in the diurnal cycle of cortisol. *Psychoneuroendocrinology* 22, 89-105.
- Song C, Dinan T, Leonard BE. Changes in immunoglobulin, complement and acute phase protein levels in depressed patients and normal controls. *J Affect Disord* 1994; **30**: 283-288.
- Song C, Merali Z, Anisman H. Systemically administered IL-1, IL-2 and IL-6 and mild stress influence in vivo monoamine variations in the nucleus accumbens. *Neuroscience* 1998, in press.
- Stein M, Keller SE, Schleifer SJ. The role of brain and the neuroendocrine system in immune regulation-potential links to neoplastic diseases. In: Levy SM (ed). *Biological Mediators of Behavior and Disease: Neoplasia*. Elsevier: New York, 1982, pp 147-174.

- Stein M, Miller AH, Trestman RL. Depression, the immune system, and health and illness. *Arch Gen Psychiatry* 1991; **48**: 171-177.
- Stein M, Miller AH, Trestman RL. Depression and the immune system. In: Ader R, Felten DL, Cohen N (eds) *Psychoneuroimmunology*. Academic Press: New York, 1991, pp 897-930.
- Stewart JW, McGrath PJ, Liebowitz MR. Treatment outcome validation of DSM-III depressive subtypes: clinical usefulness in outpatients with mild to moderate depression. *Arch Gen Psychiatry* 1985; **42**: 1148-1153.
- Stewart JW, McGrath PJ, Quitkin FM. Relevance of DSM-III depressive subtype and chronicity to antidepressant efficacy in atypical depression. *Arch Gen Psychiatry* 1989; **46**: 1080-1087.
- Stewart JW, McGrath PJ, Quitkin FM, Rabkin J, Harrison W, Wager S, Nunes S, Ocepek-Welikson K, Tricamo E. Chronic depression: Response to placebo, imipramine, and phenelzine. *J Clin Psychopharmacology* 1993; **13**, 391-396.
- Stewart JW, Quitkin FM, McGrath PJ, Rabkin JG, Markowitz JS, Tricamo E, Klein DF. Social functioning in chronic depression: Effect of 6 weeks of antidepressant treatment. *Psychiat Res* 1988; **25**: 213-222.
- Stokes PE, Sikes CR. Hypothalamic-pituitary-adrenal axis in affective disorders. In: Meltzer HY (ed). *Psychopharmacology: The Third Generation of Progress*. Raven Press: New York, 1987, pp 589-608.
- Stone EA. Problems with current catecholamine hypotheses of antidepressant agents. *Behav Brain Res* 1983; **6**: 535-578. .
- Stone EA. Central cyclic-AMP-linked noradrenergic receptors: New findings on properties as related to the actions of stress. *Neurosci Biobehav Rev* 1987; **11**: 391-398.
- Strauman TJ, Lemieux AM, Coe CL. Self-discrepancy and natural killer cell activity: Immunological consequences of negative self-evaluation. *J Pers Soc Psychol* 1993; **64**: 1042-1052.

- Sulser F. Antidepressant treatments and regulation of norepinephrine-receptor-coupled adenylate cyclase systems in brain. In: Usdin E, Asberg M, Bertilsson L, Sjoqvist F (eds). *Frontiers in Biochemical and Pharmacological Research in Depression*, Raven: New York, 1984, pp 249-262.
- Swann AC, Secunda SK, Stokes PE, Croughan J, Davis JM, Koslow SH, Maas JW. Stress, depression and mania: Relationship between perceived role of stressful events and clinical and biochemical characteristics. *Acta Psychiatr Scand* 1990; 81: 389-397.
- Syvalahti E, Eskola J, Ruuskanen O, Laine T. Nonsuppression of cortisol in depression and immune function. *Prog Neuropsychopharmacol Biol Psychiatry* 1985; 9: 14-22.
- Szadoczky E, Sazekas I, Rihmer Z, Arato M. The role of psychosocial and biological variables in separating chronic and non-chronic major depression and early-late onset dysthymia. *J Affect Disord* 1994; 32: 1-11.
- Tanaka M, Kohno Y, Nakagawa R, et al., Time related differences in noradrenaline turnover in rat brain regions by stress. *Pharm Biochem Behav* 1982; 16: 315-319.
- Targum SD, Marshall LE, Fischman P, Martin D. Lymphocyte subpopulations in depressed elderly women. *Biol Psychiatry* 1989; 26: 581-589.
- Taylor SE, Brown JD. Illusion and well-being: A social psychological perspective on mental health. *Psychol Bull* 1988; 103: 193-210.
- Thase ME, Fava M, Halbreich U, Kocsis JH, Koran L, Davidson J, Rosenbaum J, Harrison W. A placebo-controlled, randomized clinical trial comparing sertraline and imipramine for the treatment of dysthymia. *Arch Gen Psychiatry* 1996; 53: 777-784.
- Thase ME, Kupfer DJ, Buysse DJ, Frank E, Simons AD, McEachran AB, Rashid KF, Grochocinski VJ. Electroencephalographic sleep profiles in single-episode and recurrent unipolar forms of major depression: I. Comparison during acute depressive states. *Biol Psychiatry* 1995; 38: 506-515.
- Thase ME, Sullivan LR. Relapse and recurrence of depression: A practical approach for prevention. *CNS Drugs* 1995; 4: 261-277.

- Tilders FJH, Schmidt ED, De Goeij DCE: Phenotypic plasticity of CRF neurons during stress. *Ann NY Acad Sci* 697: 39-52, 1993.
- Tyrer P, Seivewright N, Murphy S, Ferguson B, Kingdon D, Barczak P, Brothwell J, Darling C, Gregory S, Johnson AL. The Nottingham study of neurotic disorder: comparison of drug and psychological treatments. *Lancet* 1988; 2 (8605): 235-240.
- Vallejo J, Gasto C, Catalan R, Salamero M. Double blind study of imipramine versus phenelzine in melancholias and dysthymic disorders. *Br J Psychiatry* 1987; **151**: 639-642.
- Vander AJ, Serman JH, Luciano DS. *Human Physiology: The Mechanisms of Body Function* (3rd Edition). McGraw-Hill: New York, 1980.
- Vannelle JM, Attar-Levy D, Poirier MF, Bouhassira M, Blin P, Olie JP. Controlled efficacy study of fluoxetine in dysthymia. *Br J Psychiatry* 1997; **170**: 345-350.
- van Praag HM. Depression, suicide and serotonin metabolism in the brain. In: Post RM, Ballenger JC (eds). *Neurobiology of Mood Disorders*. Williams and Wilkins: Baltimore, 1984, pp. 601-618.
- van Praag HM. Amine hypotheses of affective disorders. In: Iversen LL, Iversen, SD, Snyder SH (eds). *Handbook of Psychopharmacology*. Plenum: New York, 1978. pp. 187-298.
- van Praag HM, Korf J. Retarded depression and the dopamine metabolism. *Psychopharmacology* 1971; **19**: 199-203.
- Versiani M. Pharmacotherapy of dysthymia: A controlled study with imipramine, moclobemide or placebo. *Neuropsychopharmacology* 1993; **10**(3S), 298.
- Versiani M, Amrein R, Stabl M, the International Collaborative Study Group. Moclobemide and imipramine in chronic depression (dysthymia): an international double-blind, placebo-controlled trial. *Int Clin Psychopharm* 1997; **12**: 183-193.

- Versiani M, Nardi D, Capponi R, Costa DA, Magistris H, Ucha Udabe R. Moclobemide compared with imipramine in the treatment of chronic depression (dysthymia DSM-III-R): A double-blind placebo-controlled trial. *Clin Neuropharmacol* 1992; **15** (suppl 1): 148b.
- Vetulani J. Studies on the neurochemical basis of action of antidepressant drugs and electroconvulsive treatment. *Pol J Pharmacol Pharm* 1984; **36**: 101-116.
- Wahlund B, Saaf J, Wetterberg L. Clinical symptoms and platelet monoamine oxidase in subgroups and different states of affective disorders. *J Affect Dis* 1995; **35**, 75-87.
- Walker V, Streiner DL, Novosel S, Rocchi A, Levine MAH, Dean DM. Health-related quality of life in patients with major depression who are treated with moclobemide. *J Clin Psychopharmacol*. 1995; **15**: 60S-67S.
- Weiss JM, Goodman PA, Losito BG, Corrigan S, Charry JM, Bailey WH. Behavioral depression produced by an uncontrollable stressor: Relationship to norepinephrine, dopamine and serotonin levels in various regions of rat brain. *Brain Res Rev* 1981; **3**: 161-191.
- Weiss JM, Simson PE. Electrophysiology of the locus coeruleus: Implications for stress-induced depression. In: Koob GF, Ehlers CL, Kupfer DJ (eds). *Animal Models of Depression*. Birkhauser: Boston, 1989, pp 111-134.
- Weiss JM, Simson PE. Neurochemical mechanisms underlying stress-induced depression. In: Field T, McCabe P, Schneiderman N (eds). *Stress and Coping*. Erlbaum: New York, 1985, pp 93-116.
- Weisse CS. Depression and immunocompetence: A review of the literature. *Psychol Bull* 1992; **111**: 475-489.
- Weissman MM, Leaf PJ, Bruce ML, Florio L. The epidemiology of dysthymia in five communities: Rates, risks, comorbidity and treatment. *Am J Psychiat* 1988; **145**: 815-819.

- Wells KB, Stewart A, Hays RD, Burnam MA, Rogers W, Daniels M, Berry S, Greenfield S, Ware J. The functioning and well-being of depressed patients. *JAMA*.1989; **262**: 914-919.
- Willner P. Dopaminergic mechanisms in depression and mania. In: Bloom FE, D.J. Kupfer DJ (eds). *Psychopharmacology: The Fourth Generation of Progress*. Raven Press: New York, 1995, pp 921- 931.
- Willner P. The anatomy of melancholy: the catecholamine hypothesis of depression revisited. *Rev Neurosci*. 1987; **1**: 77-99
- Wise RA. The anhedonia hypothesis: mark III. *Behav Brain Sci* 1985; **8**: 178-186.
- Yee CM, Deldin PJ, Miller GA. Early stimulus processing in dysthymia and anhedonia. *J Abn Psychol* 1992; **101**: 230-233.
- Yee CM, Miller GA. A dual-task analysis of resource allocation in dysthymia and anhedonia. *J Abn Psychol* 1994; **103**: 625-636.
- Yee CM, Miller GA. Emotional information processing: Modulation of fear in normals and dysthymics. *J Abn Psychol* 1988; **97**: 54-63.
- Zacharko RM, Anisman H. Stressor-induced anhedonia in the mesocorticolimbic system. *Neurosci Biobehav Rev* 1991; **15**: 1-15.
- Zacharko RM, Anisman H. Pharmacological, biochemical and behavioral analyses of depression: Animal models. In: Koob GF, Ehlers CL, Kupfer DJ (eds). *Animal Models of Depression*. Birkhauser: Boston, 1989, pp 204-238.
- Zalcman S, Richter M, Anisman H. Alterations of immune functioning following exposure to stressor related cues. *Brain Behav Immun* 1989; **3**: 99-109.
- Zuroff DC, Mongrain M. Dependency and self criticism: Vulnerability factors for depressive affective states. *J Abn Psychol* 1987; **96**: 14-22.

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