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Insufficient glucocorticoid signaling and elevated inflammation in coronary heart disease patients with comorbid depression

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ABSTRACT

Coronary heart disease (CHD) and depression are very common and often co-existing disorders. In addition to psychological and social morbidity, depression exacerbates adverse cardiac outcomes in CHD patients. Inflammation has been proposed as one of the mechanisms involved in the association between these two debilitating diseases. Therefore, the present study aimed to evaluate inflammatory responses as well as to investigate the pathophysiological mechanisms underlying the putative inflammatory activation in CHD patients with and without depression, by assessing the function of two important biological factors regulating inflammation, the hypothalamus–pituitary–adrenal (HPA) axis and the glucocorticoid receptor (GR). Eighty-three CHD patients with ($n = 28$) and without ($n = 55$) comorbid depression were recruited from primary care services in South London. Depression status was assessed by means of Clinical Interview Schedule Revised for diagnosis of depression, and Beck Depression Inventory for the presence of depressive symptoms. Serum C-reactive protein (CRP), plasma vascular endothelial growth factor (VEGF), and plasma and salivary cortisol were measured using commercially available ELISA kits. Gene expression of GR and interleukin-6 (IL-6) were conducted via qPCR. GR sensitivity was evaluated *in vitro* in isolated peripheral blood mononuclear cells using the dexamethasone inhibition of lipopolysaccharide-stimulated IL-6 levels. Serum levels of kynurenine pathway metabolites were measured using high performance liquid chromatography. Our results show that CHD patients with depression had higher levels of CRP, IL-6 gene expression, and VEGF compared with CHD non-depressed, as well as lower plasma and saliva cortisol levels. The CHD depressed group also exhibited a reduction in GR expression and sensitivity. Finally, tryptophan levels were significantly lower in patients with depression, who also showed an increased kynurenine/tryptophan ratio. In conclusion, CHD patients with depression had elevated levels of inflammation in the context of HPA axis hypoactivity, GR resistance, and increased activation of the kynurenine pathway. Reduced cortisol bioavailability and attenuated glucocorticoid responsiveness due to decreased expression and sensitivity of GR may lead to insufficient glucocorticoid signaling and thus elevation of inflammation in these patients.

1. Introduction

Heart disease and depression are two very common and often co-existing disorders affecting the population worldwide. According to the World Health Organization Global Burden of Disease Survey, coronary heart disease (CHD) and major depressive disorder (MDD) are currently the first and second causes of disability in developed countries, and it is estimated that this will apply to all countries throughout the world by the year 2020 (Blazer, 2000; Licinio et al., 2002). The prevalence of depression among patients with established CHD is considerably higher as compared to the general population (Carney and Freedland, 2003). Depression exacerbates adverse cardiac outcomes in CHD patients, besides worsening the psychological and social morbidity. Indeed, depression has been recognized as a negative prognostic indicator and an independent factor greatly increasing the risk of cardiovascular

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related morbidity and mortality, regardless of etiology and other known cardiac risk factors (Jiang et al., 2002; Meijer et al., 2011; Miller et al., 2002).

The mechanisms associated with the link between the two diseases are not completely understood and identifying the pathophysiological mechanisms underlying the increased incidence of depression in patients with CHD remains a big challenge (Whooley and Wong, 2013). Inflammation has been recognized as a common link between these two mental and physical disorders (Halaris, 2013; Kop and Gottdiener, 2005; Maes et al., 2011; McCaffery et al., 2006). Indeed, failure of regulatory responses in the control of inflammation could be particularly important in the development of depression in CHD patients, when the immune system fails to induce an appropriate anti-inflammatory response to inhibit excessive release of pro-inflammatory cytokines (Davidson, 2012).

Inflammation plays a central role in pathogenesis of CHD and is involved in all stages of atherosclerosis including initiation, propagation, and activation of atherosclerotic plaque, ultimately leading to thrombogenesis. Indeed, CHD patients show higher circulating pro-inflammatory cytokines, higher clinical markers of inflammation like C-reactive protein (CRP), and association with higher expression of inflammatory genes (Hansson, 2005; Libby, 2006). Downregulation of the anti-inflammatory cytokine IL-10 and upregulation of the pro-inflammatory cytokines IL-6 and TNF-α have been reported in chronic heart failure patients with depressive symptoms (Parissis et al., 2004), and patients with myocardial infarction have increased plasma IL-6 and CRP concentrations and altered response to the anti-inflammatory properties of glucocorticoids, which independently correlate with depressive symptoms (Pizzi et al., 2008). Activated inflammatory responses are obvious candidate for the increased comorbidity with depression. Increased inflammation has been suggested to induce depressive symptoms not only by directly affecting the brain but also modulating the serotonergic system (Dantzer et al., 2011; Wichers and Maes, 2004) through indoleamine-2,3-dioxygenase (IDO) enzyme overstimulation, possibly leading to tryptophan (TRP) depletion (Wichers and Maes, 2002). In fact, in response to inflammatory challenges, TRP metabolism shifts towards the formation of kynurenine (KYN) metabolites (Lapin and Oxenkrug, 1969) with an increased production of 3-hydroxykynurenine (3-HK) and the depressogenic, quinolinic acid (NMDA receptor agonist, neurotoxic metabolite) rather than kynurenic acid (KYNA) (N-methyl d-aspartate (NMDA) receptor antagonist, neuroprotective metabolite) (Myint and Kim, 2003).

Hypothalamus–pituitary–adrenal (HPA) axis also plays a fundamental role as a regulatory system in stress responses and inflammation. Alteration of the HPA axis is observed in a significant proportion of patients with major depression and seems to reflect an impaired ability of cortisol to exert its physiological effects including the negative feedback on the HPA axis itself as well as the anti-inflammatory effects at the peripheral level (Pariante, 2006; Pariante and Miller, 2001). Indeed, excessive inflammation due to impaired glucocorticoid receptor (GR) sensitivity has been found in patients with MDD (Pace et al., 2007; Stewart et al., 2009), in elderly individuals (Rohleder et al., 2002), and in at-risk patients for cardiovascular disease (Carvalho et al., 2015). It has been also postulated that glucocorticoid resistance itself may occur as a result of chronic stress and prolonged exposure to inflammatory cytokines (Miller et al., 1999).

The present study focuses on the potential mechanisms underlying the pathophysiology of depression in heart disease patients, to further advance our current understanding of the link between these two debilitating conditions by investigating the possible biological pathways involved through assessing inflammatory response, HPA axis activity, GR sensitivity and kynurenine pathway, all in the same population.

2. Materials and methods

2.1. Subjects

The study subjects were recruited from participants of the UPBEAT UK research programme led by Prof. Andre Tylee (Institute of Psychiatry, Kings College London) examining patients with CHD and evaluating psychosocial predictors of depression in CHD. In the UPBEAT cohort study (Tylee et al., 2011), patients were recruited from sixteen practices across South London. Patients were eligible if they were registered on the primary care CHD registers which were kept by general practices under the Quality and Outcomes Framework. Patients were on the GP CHD registers because of documented history of myocardial infarction (MI) (around 50%) up to 20 years previously or had had angina or an intervention at some time (angioplasty, stent, bypass surgery etc.). Depression was assessed through baseline interview for psychiatric evaluation of mental state using the Clinical Interview Schedule-Revised (CIS-R) (Lewis et al., 1992) based on ICD-10 code (International Classification of Diseases-10) (Walters et al., 2014). CHD patients who had given prior authorization to be contacted for related-studies were invited to participate in the present biological project. The priori exclusion criteria included patients taking corticosteroid medications such as prednisolone, patients with other relevant medical co-morbidity including asthma, cancer, and arthritis, as well as those suffering from acute infections. Furthermore, based on the CIS-R assessment patients with a primary diagnosis of other psychiatric conditions (panic, obsessive–compulsive disorder, phobias, generalized anxiety disorder, and mixed depression anxiety disorder) were also excluded. The depression status was defined based on the CIS-R assessment (Lewis et al., 1992) for a diagnosis of depression as well as the concurrent presence of depressive symptoms, using the Beck depression inventory (BDI) score (Beck et al., 1961) obtained at the time of the biological sample collection. The cut-off BDI scores $\geqslant 10$ was used to indicate the presence of depressive symptoms (Beck et al., 1988). Therefore, CHD patients who were both CIS-R and BDI positive were categorized as CHD depressed (CHD-D), and CHD patients who were both CIS-R and BDI negative were categorized as CHD non depressed (CHD). This project was approved by the National Research Ethics Service, East Kent Local Research Ethics committee (reference number 09/H1103/19); The Institution Review Boards at the Institute of Psychiatry, King's College London (reference numbers 07/H0809/38 and 322/2003); and the University of Roehampton Ethics Committee (reference number BHS 10/025). Patients were interviewed, and asked to complete the BDI followed by collecting peripheral venous blood. Non-fasting blood samples were collected between 8:30 am and 10:30 am. Written informed consent was obtained from all participants. A total of $n = 28$ CHD-D patients and $n = 55$ CHD patients were included in the study. The demographic information for the participants is presented in Table 1. The effect of age, gender, employment statues were analyzed in all subsequent analysis. Some CHD-D patients were taking selective serotonin reuptake inhibitor or tricyclic antidepressants, and again their effects were analyzed.

2.2. Salivary cortisol

Saliva samples were collected from participants according to our previously published procedure (Mondelli et al., 2010). Participants were asked to complete the BDI questionnaire on the day of sampling. Individuals who described problems during sample collection in the self-recorded questionnaire provided, or who did not respect the time-intervals required, were taken out of the

Table 1
Characteristics of the coronary heart disease subjects with and without depression.

	CHD (n = 55)	CHD-D (n = 28)	Test and significance
Sociodemographic			
Age (years) (mean)	70 ± 1	69 ± 2	t (81) = 0.72, p = 0.48
Gender, male (%)	89	54	χ^2 = 13.26, p = 0.001**
Ethnicity (% of white)	91	82	χ^2 = 1.35, p = 0.25
Marital status [n (%)]			
Married/divorced/widowed/single	75/9/7/9	59/11/15/15	χ^2 = 2.28, p = 0.52
Educational level [n (%)]			
⩽10 years/>11 years	43/57	59/41	χ^2 = 2.00, p = 0.16
Employment status [n (%)]			
Employed/retired/unemployed	16/80/4	11/70/19	χ^2 = 5.26, p = 0.07
Smoking status [n (%)]			
Current/Ex/Never	33/53/14	32/47/21	χ^2 = 0.67, p = 0.72
Psychometric			
CIS-R (%)	0	100	
BDI score (mean ± SEM)			
At the time of blood collection	4.2 ± 0.4	19.0 ± 1.3	
At the time of saliva collection	4.3 ± 0.4	18.2 ± 1.7	
History of depression (%)	0	54	
Family history of depression (%)	13	36	
Cardiac factors and diagnoses			
Myocardial infarction (%)	47	36	χ^2 = 1.01, p = 0.32
Heart rate (mean ± SEM)	58.1 ± 1.5	67.0 ± 4.0	t (52) = −2.09, p = 0.047*
Hypertension (%)	69	79	χ^2 = 0.92, p = 0.34
BMI (kg/m^2) (mean ± SEM)	29.2 ± 0.8	30.2 ± 1.5	U = 733.0, z = −0.23, p = 0.82
High cholesterol (%)	66	56	χ^2 = 0.84, p = 0.36
Family history of heart problems (%)	65	70	χ^2 = 0.25, p = 0.62
Diabetic CHD (%)	18	29	χ^2 = 1.18, p = 0.28
Concomitant medications			
Antidepressants (%)	0	39	
Statin (%)	89	80	χ^2 = 0.99, p = 0.32
ASA (%)	71	80	χ^2 = 0.69, p = 0.41
Anticoagulants (%)	21	20	χ^2 = 0.01, p = 0.91
Antihypertensive (%)	94	92	χ^2 = 0.14, p = 0.71
Beta-blockers (%)	60	52	χ^2 = 0.40, p = 0.53
ACE-inhibitors (%)	67	52	χ^2 = 1.69, p = 0.19
Ca^{2+}-channel inhibitors (%)	25	20	χ^2 = 0.24, p = 0.63

BMI = body mass index, ACE = angiotensin-converting-enzyme inhibitor, ASA = acetylsalicylic acid.

analysis. Salivary cortisol levels were measured using a commercially available high sensitivity salivary cortisol enzyme immunoassay kit (Salimetrics). SoftMax Pro 4.8 software was used to calculate the cortisol values, following a 4-parameter fit. The analytical sensitivity was set to 0.33 nmol/l. Inter and intra-assay co-efficient of variations ranged 8–11% and 6–10%, respectively. To investigate the responsiveness of the HPA axis, the repeated measures of salivary cortisol was used to calculate the area under the curve (AUC) of the cortisol awakening response (CAR) and diurnal cortisol with respect to the increase (AUC_I) as well as with respect to the ground (AUC_G). The formulas for the calculations of the AUC were derived from the trapezoidal formula introduced by Pruessner et al. (2003).

2.3. Peripheral cortisol and CRP measurements

Blood samples collected in sodium-heparin and clot activator containing tubes were centrifuged to separate plasma and serum, respectively. The samples were stored at −80 °C until assayed. The samples were analyzed at the biochemistry laboratory, KingsPath, at King's College Hospital. The levels of peripheral CRP were determined performing the Cormay high sensitivity CRP (HsCRP) assay (P.Z. Cormay, Lublin, Poland). The minimum detectable concentration of hsCRP was 0.01 mg/dL. Inter and intra-assay co-efficient of variations were <10%. Cortisol reagents were supplied by Siemens Healthcare Diagnostics Ltd., Camberley, Surrey, UK. The minimum detectable concentration was 30 nmol/L. Inter and intra-assay co-efficient of variations were <10%.

2.4. Gene expression analysis

Blood samples for gene expression analysis of IL-6 and GR mRNA were collected using PAXgene Tubes (Qiagen). Isolation of RNA was performed using the PAXgene Blood RNA Kit (Qiagen) according to manufacturer's instructions. The RNA quality and quantity were assessed by evaluation of the A260/280 and A260/230 ratios using a Nanodrop spectrometer (NanoDrop Technologies, USA). Samples were kept frozen at −80 °C until further use. One microgram of total RNA was used for cDNA synthesis and for subsequent gene expression analysis in quantitative Real Time PCR (qPCR) performed using HOT FIREPol EvaGreen qPCR (Solis BioDyne, Tartu, Estonia) according to the SYBR Green method. For each target primer set a validation experiment was performed to demonstrate that PCR efficiencies were within the range of 90–100% and approximately equal to the efficiencies of the reference genes; glyceraldehyde 3-phosphate dehydrogenase (GAPDH), beta-actin (ACTB), and beta-2-microglobulin (B2M). Each sample was assayed in duplicate and each target gene was normalized to the geometric mean of the expression of the three housekeeping genes as reference. Pfaffl equation was used to

quantify the relative expression levels of each target gene (Pfaffl, 2001). This method has been previously used in our group (Anacker et al., 2011; Cattaneo et al., 2013).

2.5. Vascular endothelial growth factor (VEGF) measurement in plasma

The quantitative determination of vascular endothelial growth factor (VEGF) concentrations in plasma samples were measured by a commercially available ELISA kit (R&D systems, Minneapolis, MN, USA) (Rosti et al., 2007). Microplate reader (Beckman Coulter DTX 880) was used to determine optical density at 450 nm with correction at 570 nm. SoftMax Pro 4.8 software was used to calculate the VEGF values. Inter- and intra-assay variations were <10%.

2.6. GR function evaluation in vitro

Sodium-heparinized blood was diluted 1:1 with phosphate buffered saline (PBS). Peripheral blood mononuclear cells (PBMC) were isolated by Ficoll-Paque density gradient centrifugation, as per previous publications (Knijff et al., 2007; Sarkar et al., 2003). The procedure was carried out by layering diluted blood on top of Ficoll (2:1 e.g. 30 ml diluted blood on 15 ml of Ficoll) in a 50 ml polypropylene tube. The tube was centrifuged at 1000 g for 15 min at room temperature, and PBMC were re-suspended in PBS to the total volume of 50 ml, and centrifuged at 800 g for 10 min at room temperature. The supernatant was discarded and PBMC rich pellet was re-suspended in a cryovial in 1 ml cold cell culture fluid 1 (CF1) medium consisted of 10% FCS (fetal calf serum), and 1% PEN-STREP (Penicillin–Streptomycin), and 90% RPMI 1640 with 25 mM Hepes and Ultraglutamin-1 medium. Freshly prepared cell culture fluid 2 (CF2) containing 80% CF1 and 20% DMSO were then added 1:1 slowly in droplets to the cell suspension. The cells were distributed in 1 ml aliquots in precooled cryovials ready for storage using slow temperature-lowering method by incubating them in an isopropyl alcohol bath in Cryo 1 °C freezing container (Mr. Frosty polyethylene vial holder). The cryovials in the freezing container were placed at −80 °C overnight and then transferred into liquid nitrogen for long-term storage until future use. GR sensitivity was analyzed directly on isolated and stored PBMC using the "dexamethasone inhibition of lipopolysaccharide (LPS)-stimulated IL-6 levels" technique according to a previously published method with slight modifications (Carvalho et al., 2008, 2010). Recovery and viability of PBMC were examined by trypan blue exclusion technique, and cryopreserved and thawed cells contained greater than 95% viable recovered cells. PBMC were cultured in a 96-well round-bottomed tissue culture plate (Falcon, 3077) at a concentration of 50×10^4 cells/ml (100 K/well) in CF1. Cells were cultured without stimulation (Unst), or stimulated by LPS (Sigma) (1 ng/ml). Next, dexamethasone (DEX) (Sigma) was used at different concentrations (10^{-6}, 10^{-7}, 3×10^{-8}, 10^{-8}, 10^{-9} M) to inhibit the production of IL-6 mediated by the GR. Each condition was assayed in duplicate. PBMC were then incubated for 24 h at 37 °C, under 5% CO_2 and humidified air. After 24 h, the plate was centrifuged at 1000 g for 10 min at 4 °C. The supernatants from each well were harvested and stored at −20 °C until they were assayed the following day for measurements of IL-6 levels via ELISA as per previously published work with slight modifications (Knijff et al., 2007). A 96-well ELISA plate (Costar 3590) was coated overnight at 4 °C adding 100 μl diluted (1:1000) IL-6 specific capture antibody (Invitrogen: Ms mAb Anti-Hu IL-6) to each well. After overnight incubation, the wells were then emptied and tapped on a paper filter. Next, 300 μl block buffer, PBS containing 0.5% BSA (bovine serum albumin, Sigma) solution, was added to each well, and the plate was incubated for 2 h in room temperature. After blocking step, the plate was washed 4 times using the plate washer machine and freshly prepared wash buffer (MilliQ water containing 0.9% Nacl and 0.05% Tween20 solution). The supernatant samples were diluted to a measurable solution (1:2, 1:100, and 1:50 for Unst, LPS, and DEX, respectively) in working buffer (PBS containing 0.5% BSA). The standard solutions of IL-6 (R&D: recombinant human IL-6) were prepared in serial dilution (500, 250, 125, 62.5, 31.25, 15.63, 7.8, 3.9, 1.95, 0 ng/ml) for the standard curve. 100 μl of diluted samples and standard solutions were aliquoted in duplicate in the appropriate wells. 50 μl diluted (1:500) IL-6 detection antibody (Invitrogen: Ms mAb Anti-Hu IL-6 Biotin) were thereafter added to each well followed by incubation for 2 h on a plate shaker at room temperature. After another washing step, 100 μl diluted (1:15,000) streptavidin poly HRP (Thermo scientific) was added to enhance the reaction, and the plate was incubated for 30 min under continuous shaking at room temperature. The plate was then washed 5 times. The reaction was developed in dark after adding 100 μl of TMB substrate (Thermo Scientific), and then stopped by adding 100 μl 1 M H2SO4 when the color of Blank and the standard solution in the lowest concentration started to turn slightly blue (approximately after 3–5 min). The absorbance at 450/620 nm was measured using ELISA reader (MULTISKANEX, Thermo Electron Corporation). The concentrations of IL-6 in samples of supernatants were determined using GraphPad Prism Software version 4.0 for windows, Inc., San Diego, California, USA. Inter- and intra-assay variations were <10%. The analytical sensitivity was set to 2 ng/mL. The values obtained from the levels of IL-6 of the unstimulated cells were deducted from the LPS-stimulated IL-6 levels for each condition. Glucocorticoid suppression was calculated by normalizing the data to LPS-stimulated IL-6 levels (in the absence of dexamethasone), which was expressed as 100% (Carvalho et al., 2010).

2.7. Tryptophan and kynurenine pathway analysis

Serum samples were sent to Dr. Aye Mu Myint and her colleagues in Germany (Laboratory for Psychoneuroimmunology, Ludwig-Maximilians University, Munich) to determine the levels of TRP, KYN, KYNA, and 3-HAA metabolites, using high performance liquid chromatography (HPLC) method of Herve et al. (1996) with some modifications as well as the recently published method by Oades et al. (2010) to measure 3-HK. The following is a brief description of the method used: KYN was detected spectrophotometrically at 365 nm. KYNA was detected fluorimetrically at an excitation wavelength of 334 nm and an emission wavelength of 388 nm. KYNA was analyzed in plasma that was deproteinised using perchloric acid. 3-HK was measured at a wavelength of 365 nm by UV detection. All of the analyses were conducted using HPLC with a reversed phase C-18 column. The 3-HK analysis method has been validated showing an absolute recovery of 85.8%, intra-day precision of 3.9%, and inter-day precision of 7.5%. A time series demonstrated total stability of the analyte 3-HK during the extraction and analysis steps for up to 3× repeated freezing/thawing cycles, which ensured the validity of the analysis in samples that had already been thawed and frozen again. The intra- and inter-assay coefficients of variation ranged from 5% to 7% for all of the metabolites. The kynurenine to tryptophan (KYN/TRP) ratio was calculated.

2.8. Data analysis

All statistical analyses were performed using SPSS software version 20.0 and GraphPad Prism 4.0 for Windows. The data were reported as the mean ± SEM. All data were tested for suitability for parametric or non-parametric analysis. For comparing the biological factors assessed in this study between the two groups of

CHD patients without depression and CHD patients with depression, independent *t*-test or Mann–Whitney *U*-test was used, as appropriate. Dichotomous variables were compared using Chi-squared test. Correlations were assessed using Pearson's product moment correlation or Spearman's correlation coefficient where the data violated parametric assumptions. For comparison of more than two groups, One-way ANOVA analysis or Kruskal–Wallis test were performed, as appropriate. General linear model (GLM) was performed for taking into account the effect of covariates (ANCOVA) such as gender, age, employment status, and antidepressant use on the findings. The *p*-values of <0.05 were considered as significant.

3. Results

3.1. CHD patients with depression show higher levels of inflammation

Both CHD groups, with and without depression, exhibited mean CRP levels greater than 3 mg/L, considered as being in high-risk group for cardiovascular events according to the American Heart Association (see Table 2). However, mean inflammation was found to be even higher (5.2 mg/L) in CHD patients with depression ($p < 0.05$). CRP levels were found to be positively correlated with heart rate in CHD patients ($r_s = 0.37$, $p = 0.008$, $n = 50$). In order to evaluate further the inflammatory response, we analyzed gene expression levels of IL-6 mRNA; its expression was significantly increased in CHD depressed patients as compared with non-depressed (~2.8-fold increase, $p < 0.01$, Table 2). There were no differences in the number of monocytes between the two groups: (mean ± SEM); CHD $0.51 \pm 0.03 \times 10\text{^}9/L$, CHD-D $0.48 \pm 0.03 \times 10\text{^}9/L$, $t\ (50) = 0.64$, $p = 0.5$). Gender and age were found to have a significant effect on IL-6 expression; $F(1,36) = 4.226$, $p = 0.047$ and $F(1,36) = 6.167$, $p = 0.018$, respectively. However, the result held true after controlling for the effect of these covariates ($F(3,35) = 4.54$, $p = 0.04$). We also analyzed VEGF as another inflammatory marker, since its release is stimulated by IL-6 and inhibited by glucocorticoids. Comparing the two groups, CHD depressed patients showed significantly higher (almost twofold) plasma VEGF levels than CHD non-depressed individuals ($p < 0.05$, Table 2).

Table 2
Inflammatory biomarkers measured in CHD patients with and without depression.

Inflammatory biomarkers	CHD (mean ± SEM)	CHD-D (mean ± SEM)	Comparison analysis
Serum CRP (mg/L)	3.34 ± 0.75 $n = 33$	5.20 ± 0.99 $n = 21$	$U = 224.50$, $Z = -2.17$, $p = 0.030^{*}$
IL-6 gene expression	1.14 ± 0.13 $n = 23$	3.22 ± 0.63 $n = 16$	$U = 79.50$, $Z = -2.98$, $p = 0.003^{**}$
Plasma VEGF (pg/mL)	94.28 ± 16.22 $n = 29$	173.13 ± 36.46 $n = 21$	$U = 193.00$, $Z = -2.19$, $p = 0.028^{*}$

CRP = C-reactive protein, IL-6 = interleukin-6, VEGF = vascular endothelial growth factor.

3.2. CHD patients with depression show lower cortisol in both plasma and saliva

As demonstrated in Fig. 1a, CHD patients with depression had lower (−20%) plasma cortisol than non-depressed group ($p = 0.026$). In addition, the depressed group exhibited significantly decreased cortisol levels in the first hour after awakening compared to CHD non-depressed (Two-way ANOVA, $F(1,3) = 19.45$, $n = 51$, $p < 0.0001$), and post hoc tests indicated significant findings at 30 min and 60 min (Fig. 1b). The results from calculating the area under the curve for the cortisol awakening response in respect

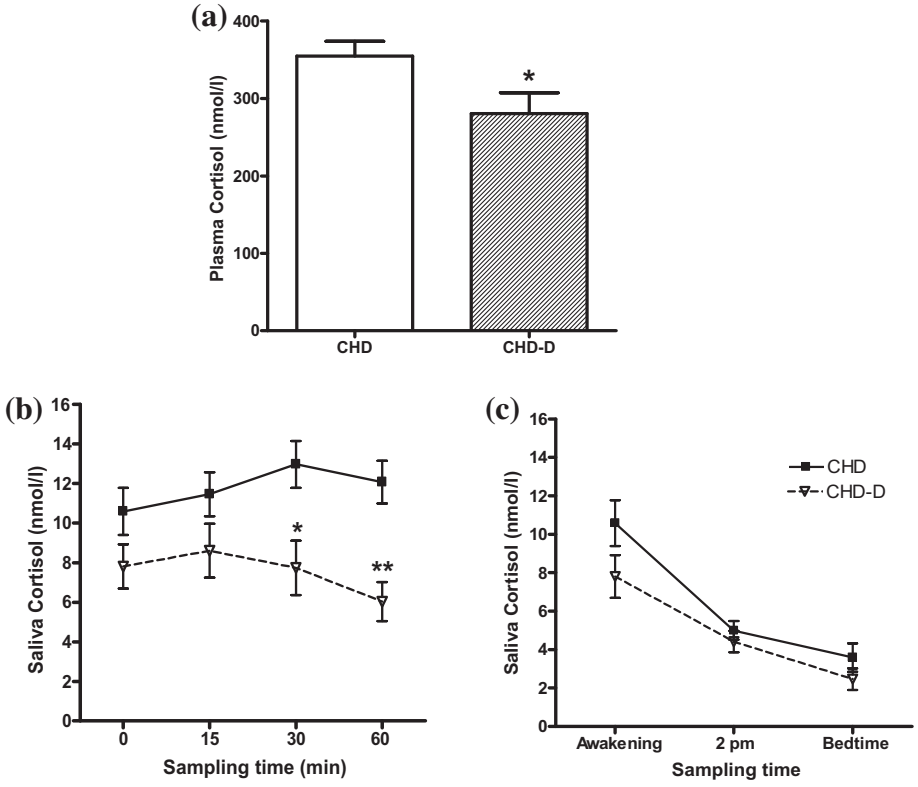

Fig. 1. Plasma cortisol levels (nmol/L) (a); mean salivary cortisol levels for the first hour after awaking at four sampling points on awakening and 15, 30, and 60 min later (b); and mean salivary cortisol levels over the day at three sampling points on awakening, 2 pm, and bedtime (c) in CHD patients with and without depression. Data expressed as mean ± SEM. $^{*}p < 0.05$, $^{**}p < 0.01$.

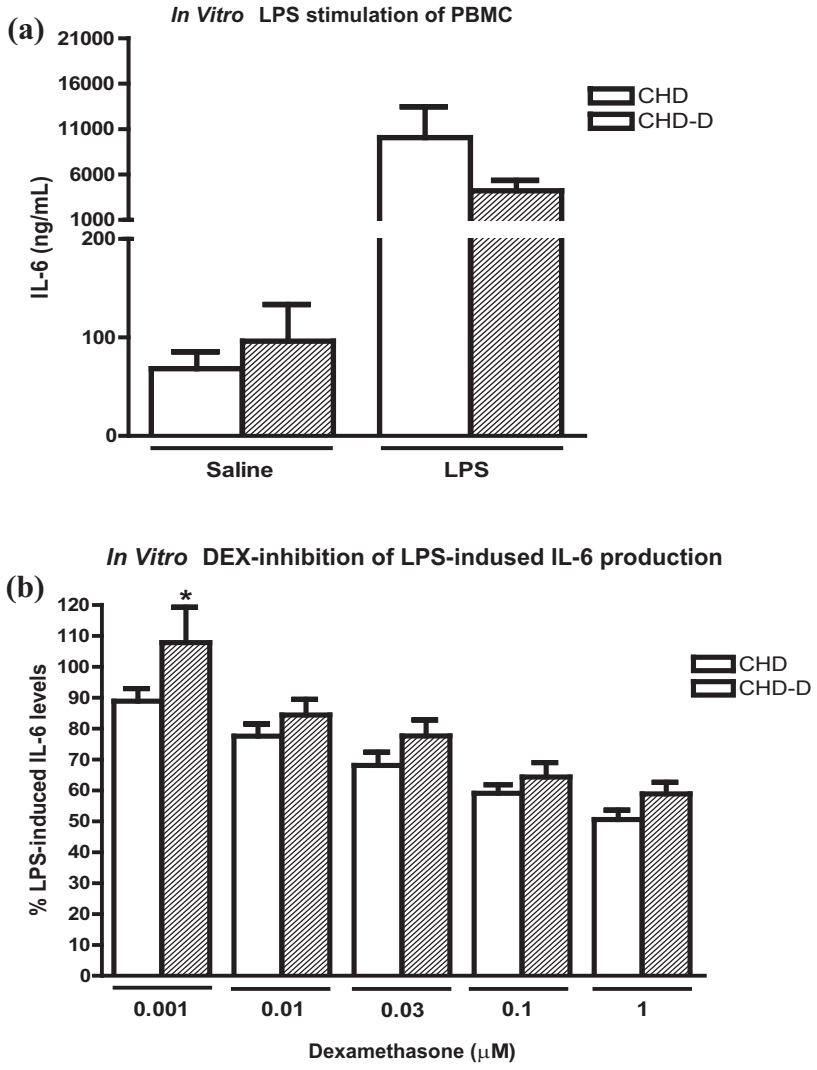

Fig. 2. IL-6 levels (ng/mL) before and after *in vitro* LPS stimulation (a) and dexamethasone inhibition of LPS-stimulated IL-6 production (b) in PBMC of CHD patients with and without depression. Data expressed as mean ± SEM. $^{*}p < 0.05$.

to the ground (distance from zero) also revealed significantly lower cortisol output during the first hour post awakening: CAR AUC_G, ($U = 176.0$, $p = 0.016$). Calculating the area under the curve taking into account the measurements of the first hour post awakening in respect to the increase from the baseline measurement (awakening) showed a trend towards a decreased cortisol response in CHD depressed compared to non-depressed group: CAR AUC_I ($U = 205.0$, $p = 0.0796$). Although, diurnal cortisol levels showed lower cortisol values at each time point (awakening, 2 pm, and bedtime) in CHD patients with depression compared with CHD without depression (Fig. 1c), no statistically significant difference was found on the diurnal cortisol and the result only reached trend-level in the Two-way ANOVA ($F(1,2) = 3.39$, $p = 0.068$). There was no difference in awakening time between the two groups: (mean ± SEM); CHD 7:04 ± 0:09 am, CHD-D 7:03 ± 0:22 am, t (51) = 0.013, $p = 0.990$).

3.3. CHD patients with depression show decreased GR expression and sensitivity

CHD depressed individuals exhibited significantly reduced expression of GR mRNA when compared with CHD non-depressed (−28%, $p < 0.05$). As expected, following *in vitro* stimulation of PBMC by LPS, IL-6 levels were remarkably increased in both CHD depressed and non-depressed patients (Fig. 2a). There was no difference between LPS-induced IL-6 levels between depressed and non-depressed patients (mean ± SEM); CHD 10,050 ± 3415 ng/mL, CHD-D 4195 ± 1142 ng/mL, t (25) = 1.38, $p = 0.18$. *In vitro* dexamethasone inhibition of LPS-stimulated IL-6 production in PBMC was measured as percentage suppression, to eliminate any potential effects of this baseline difference in stimulated IL-6 levels. As illustrated in Fig. 2b, both CHD patients with and without depression showed suppression of LPS-stimulated IL-6 levels in a concentration-dependent manner. However, across all concentrations of DEX, CHD depressed individuals exhibited higher levels of IL-6 (two-way ANOVA, $F(1,5) = 11.19$, $n = 27$, $p = 0.001$), with post hoc analyses showing significant differences for 1 nM ($t = 2.91$, $p < 0.05$).

3.4. Activation of the kynurenine pathway in CHD patients with depression

We investigated the tryptophan metabolism pathway by measuring the circulatory levels of several metabolites. As presented in Table 3, CHD depressed patients showed lower serum levels of tryptophan when compared with CHD non-depressed individuals

Table 3
Serum levels of kynurenine pathway metabolites in CHD patients with and without depression.

	CHD (n = 29) (mean ± SEM)	CHD-D (n = 15) (mean ± SEM)	Comparison analysis
TRP (μg/ml)	12.64 ± 0.64	10.42 ± 0.58	t (42) = 2.25, p = 0.03*
KYN (ng/ml)	843.50 ± 54.37	746.53 ± 42.29	U = 187.0, Z = −0.76, p = 0.45
KYN/TRP	62.21 ± 2.88	74.43 ± 5.26	t (42) = −2.22, p = 0.03*
KYNA (ng/ml)	15.57 ± 1.18	12.02 ± 1.43	t (42) = 1.83, p = 0.07
3-HK (ng/ml)	18.85 ± 1.78	18.40 ± 1.49	U = 192.5, Z = −0.62, p = 0.54
3-HAA (ng/ml)	34.31 ± 3.35	33.72 ± 5.48	U = 202.0, Z = −0.38, p = 0.70

TRP = tryptophan, KYN = Kynurenine, KYNA = Kynurenic acid, 3-HK = 3-hydroxykynurenin, 3-HAA = 3-hydroxyanthranilic acid.

(p = 0.03). In addition, kynurenine/tryptophan (KYN/TRP) ratio, the indicator of activated IDO, was significantly higher in depressed CHD compared with non-depressed patients (p = 0.03). Although not statistically significant, the depressed group showed a trend towards less production of kynurenic acid (p = 0.07). The levels of 3-HK and 3-HAA did not differ between the two groups (p > 0.05). CRP levels were associated with KYN/TRP ratio (r_s = 0.35, p = 0.028, n = 40).

3.5. Covariate analyses

We analyzed the effects of a priori selected confounders (age, gender, employment status, and antidepressants use) on serum CRP, plasma VEGF, gene expression of IL-6 and GR, plasma cortisol, cortisol awakening response, diurnal salivary cortisol, and circulatory tryptophan and kynurenine levels. We found no effects of any of these potential confounders (all p > 0.05).

4. Discussion

The present results show that, in CHD patients, depression is accompanied by elevated levels of inflammation in the context of low cortisol output, GR resistance, and increased activation of the kynurenine pathway. Although CHD patients without depression already exhibit an activation of the inflammatory system as measured by circulatory levels of CRP, inflammation is even higher in the presence of depression comorbidity. Consistent with high CRP, CHD depressed individuals also display increased IL-6 gene expression and plasma VEGF. While the association between CHD and depression in relation to inflammation has been reported before, the present study extends the findings by showing that the depressed group exhibit insufficient glucocorticoid signaling due to both decreased hormone bioavailability as well as attenuated glucocorticoid responsiveness. Indeed, CHD patients with depression exhibit lower levels of cortisol as shown by a single measurement in plasma and repeated measures in saliva. In addition, CHD patients with depression have significantly lower expression levels of GR mRNA and less functional ability of the GR to respond to glucocorticoids. Our study also confirms that elevated inflammation is associated with effects on tryptophan metabolism through activation of the kynurenine pathway, as revealed by lower levels of tryptophan together with higher KYN/TRP ratio. Again, higher CRP is associated with increased activity of IDO enzyme. Therefore, the results suggest that an activated inflammatory response may lead to increased rates of tryptophan degradation.

CRP is known as a mediator and marker of atherosclerosis and has been consistently reported to be a potent predictor of future vascular serious events including myocardial infarction, ischemic stroke, and sudden cardiac death. This predictive value has been shown to be stronger than LDL cholesterol and also independent of sex, age, blood pressure, cholesterol, smoking, and diabetes (Ridker, 2003, 2007). On the other hand, elevation of inflammation and increased CRP concentration has been implicated in pathogenesis of depression. Indeed, patients with major depression exhibit evidence of activated inflammatory responses and elevated levels of inflammatory cytokines such as TNF-α, IL-6 and CRP (Empana et al., 2005). The phenomenon is linked to the alteration of the central nervous system and contributes to depressive symptoms (Irwin and Miller, 2007). Recently, attention has been also paid to disruption of the blood–brain-barrier (BBB) as another potential mechanism underlying the pathogenesis of depression in relation to inflammation. The hypothesis suggests breakdown of BBB leads to the penetration of inflammatory molecules into the brain (Shalev et al., 2009). CRP has been shown to be associated with increased BBB permeability (Hsuchou et al., 2012), and therefore the (relatively) high CRP levels (average >3 mg/L) even in the CHD patients without depression may indicate not only an increased cardiovascular risk but also an increased risk of developing depression in the future. Furthermore, the even higher CRP levels (average >5 mg/L) in the CHD depressed group is a significant cause of concern, since even a small elevation in CRP levels greatly increases the risk of heart attack, angina, and more severe cardiovascular complications. Indeed, we find that heart rate, which is increased in the depressed group, is also positively correlated with CRP. Increased heart rate in depressed patients with CHD has been described before (Carney et al., 1988, 1993), and it has been shown to be an independent prognostic risk factor for morbidity and mortality related to CHD (Dyer et al., 1980). Moreover, the correlation between heart rate and CRP would support the notion of a bidirectional influence between inflammation and cardiac function, but the cross-sectional nature of the data limits the possibility of speculating on the direction of the causation.

Moreover, IL-6 (also higher in CHD depressed patients) is believed to be a contributing factor in the stimulation of the processes leading to the formation and propagation of coronary plaque, including release of adhesion molecules from endothelium and platelet aggregation (Barton, 1996). Indeed, expression and plasma concentration of IL-6 and its by product CRP have been reported to be indicator of the intensity of inflammatory induced coronary plaque and are associated with instability of the plaque and vulnerability to rupture (Ikeda et al., 2001). Overexpression of IL-6 is also shown to be associated with aging, possibly accounting for the development of chronic inflammatory conditions and other physical complications, such as muscle atrophy, arthritis, and osteoporosis (Ferrucci et al., 1999; Ershler and Keller, 2000). In addition, elevation in the levels of IL-6 and CRP has been shown to be associated with a remarkably increased mortality in older people (Harris et al., 1999). These findings are important considering the age of the present participants, who are therefore at increased risk of disability, cardiovascular events, and mortality.

Finally, higher VEGF in CHD patients with depression may also contribute to worsening their cardiovascular condition. Originally identified for its ability to promote vascular permeability (Senger et al., 1986), VEGF is the most potent growth factor for endothelial cells and a main regulator of angiogenesis and vasculogenesis (Byrne et al., 2005; Keck et al., 1989) and has been shown to be

increased in patients with coronary atherosclerosis (Blann et al., 2002), with some data suggesting an association between prognosis of coronary atherosclerosis and elevated VEGF (Fleisch et al., 1999). Taken together, our findings indicate that CHD patients with comorbid depression are truly at high risk of future cardiovascular events due to the widespread increase in potent inflammatory biomarkers. The role of VEGF expands beyond the vasculature, specifically in the brain. Known as a neurotrophic factor, VEGF is thought to exert neuroprotection (Storkebaum et al., 2004) and increase neurogenesis (Jin et al., 2002) in addition to vasculogenesis. The direct neurotrophic activity of VEGF in the brain has shown to be exerted through stimulating axonal outgrowth and enhancing cell survival (Sondell et al., 1999), protecting hippocampal neurons against glutamate mediated toxicity (Matsuzaki et al., 2001), and influencing synaptic plasticity (Licht et al., 2011) and transmission (McCloskey et al., 2005). Due to the crucial involvement in the vascular system as well as neurological processes, and considering the relationship between CHD and depression, VEGF has been suggested to be one of the molecular links underlying the comorbidity of these two disorders (Warner-Schmidt and Duman, 2008). However, there are considerable discrepancies among studies investigating VEGF in depression. While some studies have reported increased VEGF mRNA (Berent et al., 2014; Iga et al., 2007), serum (Berent et al., 2014; Kahl et al., 2009) or plasma (Lee and Kim, 2012; Takebayashi et al., 2010) levels in patients with MDD, others have found significant decreases in VEGF peripheral levels in depression (Dome et al., 2009; Isung et al., 2012; Katsuura et al., 2011) or no difference between patients with MDD and healthy controls (Kotan et al., 2012; Ventriglia et al., 2009). A recent review assessing clinical studies on VEGF and depression speculated that elevation in VEGF levels in patients with MDD appears to be due to the response to the perceived stress associated with depression resulting in an attempted neuroprotective effect, whereas decreased levels of VEGF seem to be observed in treatment-resistant depressed patients whose brains are less able to undergo neurogenesis processes (Clark-Raymond and Halaris, 2013).

In our study, compared to CHD non-depressed, CHD patients with depression, show lower levels of cortisol both in plasma and saliva, together with more blunted response to the stress of awakening. Chronic inflammatory exposure in CHD is one of the stressful challenges that these patients face, and adaptation to such condition require involvement of neural–endocrine–immune mechanisms (Chrousos and Gold, 1992; McEwen, 1998). The impact of exposure to chronic or extreme stress on the neurobiology of the HPA axis is well documented. Indeed, prolonged activation of the HPA axis has an adverse effect on health outcomes both physically and emotionally (Sapolsky et al., 2000). Despite the popularity of studies showing higher levels of cortisol in MDD as a result of hyperactivity of the HPA axis and presence of GR resistance (Carvalho et al., 2008; Pariante and Miller, 2001), not all the studies have found the same results. While some studies reported depressed patients show increased level of cortisol in cerebrospinal fluid, plasma and urine, consistent with overactivity of the HPA axis; and enlargement of pituitary and adrenal glands (Otte et al., 2004; Pariante and Miller, 2001; Stewart et al., 2009), others explored the opposite findings. Indeed, decreased morning cortisol in both serum and saliva samples have been reported in depressed as well as vulnerable individuals in the community (Strickland et al., 2002). There are also studies suggesting hypoactivity of the HPA axis and hypocortisolism associated with late-life depression (Morrison et al., 2000; Oldehinkel et al., 2001) and in patients with physical comorbidity such as diabetes (Carvalho et al., 2015). In fact, late-life depressive symptoms have been suggested to be associated with both hyper and hypoactivity of the HPA axis (Penninx et al., 2007), with studies suggesting that physical frailty and exhaustion are the underlying mechanisms associated with HPA axis insufficiency among elderly with depression (Fries et al., 2005; Morrison et al., 2001; Oldehinkel et al., 2001). Moreover, some studies have shown an association of hypocortisolemia with physical complains and chronic fatigue after long term HPA axis overstimulation under chronic physical stress (Fries et al., 2005; Hellhammer et al., 2004). Decreased levels of cortisol have been consistently shown in other stress-related conditions including atypical depression (Gold and Chrousos, 2002), PTSD (Yehuda, 2001), chronic fatigue syndrome and fibromyalgia (Crofford et al., 1994; Demitrack and Crofford, 1998). Some inconsistent findings have also been reported in regards to the association of cortisol with depression in patients with cardiovascular diseases. In the Heart and Soul Study, Otte and colleagues have found an association between depression and elevated urinary cortisol in CHD patients. However, increased cortisol has not been found to be related to the worsening of cardiac function in these patients (Otte et al., 2004). In another study in CHD patients, depression was associated with flatter diurnal cortisol rhythms, which in turn contributed to prognosis of coronary atherosclerosis (Bhattacharyya et al., 2008). However, lack of association between depressive symptoms and diurnal cortisol profile has been also reported in patients following hospitalization with an acute coronary syndrome (Molloy et al., 2008).

Based on our results, CHD individuals with depression also show lower levels of GR mRNA expression, indicating less GR available for responding to the glucocorticoids, as well as impaired GR sensitivity in PBMC. Although the etiology of GR resistance is unknown, pro-inflammatory cytokines may generate glucocorticoid resistance by directly affecting functional capacity of GR at multiple levels. Studies suggest that prolonged inflammation has a direct effect in reducing GR sensitivity through the interaction of cytokine signaling pathway with GR signaling pathway (Pace et al., 2007). In addition, elevated inflammatory responses and pro-inflammatory cytokines are known to increase expression of GRβ, which is a relatively inert isoform, affecting the active isoform GRα involved in regulation of gene expression. GRβ has been suggested to decrease GR function by attenuating GRα transcriptional activity (Webster et al., 2001; Wichers and Maes, 2002). Moreover, cytokines may trigger glucocorticoid resistance by reducing GR ligand and DNA binding capacities, inhibiting GR translocation to the nucleus and influencing GR protein–protein interactions as seen for example by activating the mitogen-activated protein kinase (MAPK) signaling pathway in cytoplasm which leads to phosphorylation of the receptor protein, thus diminishing GR transcriptional activity (Pace et al., 2007; Raison and Miller, 2003). GR resistance, as a result of chronic stress, can in turn further contribute to a dysregulation of inflammation (Cohen et al., 2012). This coexistence of GR resistance and increased inflammation at a molecular level is consistent with a study on chronic stress in humans, where caregivers to cancer patients were found to have decreased expression of GR-dependent transcripts together with increased expression of inflammatory genes and biomarkers (Miller et al., 2008). Therefore, it appears that endogenous cortisol secretion is insufficient to limit inflammation in CHD patients with depression, due to hypoactivity of the HPA axis and GR resistance, leading to higher inflammatory response in these patients, which in turn further maintains GR resistance. However, it is also important to highlight one study that found *increased*, rather than decreased, GR function in CHD patients, although a number of methodological differences between may explain this discrepancy: first, patients were within 3 month after a myocardial infarction (or a revascularization procedure), while our patients had chronic conditions, likely resulting in increased inflammation occurring over many years; second, the sample was slightly younger, aged on average 60 years compared with our average age of 70; and

finally the GR sensitivity protocol was different, using whole blood rather than isolated PBMCs (Miller et al., 2005).

Finally, we found activation of the kynurenine pathway and increased IDO activity in CHD patients with depression, leading to lower peripheral levels of tryptophan (which in turn can putatively lead to reduced serotonin synthesis in the brain). Indeed, evidence suggests that alterations in inflammatory response in relation to activation of the kynurenine pathway leading to the imbalance between neuroprotective and neurotoxic metabolites have been implicated in pathology of the psychiatric conditions, including MDD (Myint et al., 2007, 2012; Myint and Kim, 2003). On the other hand, previous studies on cardiovascular diseases have also shown decreased tryptophan levels and higher KYN/TRP ratio coincided with increased neopterin concentration (a marker of monocyte/macrophage activation) in CHD patients compared to healthy controls (Wirleitner et al., 2003). In line with our findings, a recently published study revealed an association between KYN/TRP ratio and CRP as well as neopterin in CHD patients, suggesting IDO activity to be related with immune activation (OZKAN et al., 2014). IDO activity is believed to be associated with risk factors for atherosclerosis and prognosis of cardiovascular disorders (Niinisalo et al., 2008; Pedersen et al., 2011; Pertovaara et al., 2007), and one study also showed an association between depressive symptoms and carotid atherosclerosis that involved IDO activation (Elovainio et al., 2011). Therefore, it appears that elevated inflammation in our CHD depressed patients not only can lead to further disruption in serotonergic system but also worsening the cardiac outcome in this population.

One of the limitations of this study is the small sample size of the group with depression. In addition, in some experiments, only subgroups of patients were analyzed due to missing/insufficient biological samples. Furthermore, some potentially confounding variables, such as left ventricular function, congestive heart failure, or physical activity, have not been measured in this study. Finally, it should be noted that the study participants were elderly, and the clinical overlapping between older age and CHD makes it difficult to dissect the biological interaction between age, CHD and depression, also considering the age-related risk factors contributing to depression, such as medical illnesses, frailty, functional decline, and general disability (Bruce, 2002; Yoshimura et al., 2013). Indeed, our findings may not necessarily extend to other populations of younger depressed patients, since late-life depression differs in terms of psychopathology as compared to depression in younger adults; for example, depression in late life is more common as compared to midlife, with a more chronic course, and these patients often only display some symptoms of depression, not always meeting the strict criteria of the Diagnostic and Statistical Manual of Mental Disorders for clinical depression (Beekman et al., 2002; Kohn and Epstein-Lubow, 2006).

This study was cross-sectional; prospective investigations would enable examining the longitudinal association between heart disease and depression in relation to inflammation, and whether high inflammation in non-depressed CHD patients makes them more at risk of future depression. Indeed, identification of biomarkers, which predict future development of depression, will have important translational significance leading to identification of subjects that may benefit from targeted therapeutic prophylactic intervention.

5. Conclusion

We propose that chronic inflammation in patients with CHD contributes to the development of depression, which in turn leads to further activation of inflammatory processes as reflected in joint elevation in IL-6 expression and CRP as well as VEGF levels, which is inadequately restrained by endogenous glucocorticoids that are both lower and less able to act on the GR, and which lead to further brain and metabolic dysfunction by activating the kynurenine pathway. These findings may provide a better understanding of the role of inflammation in the pathogenesis of depression in coronary heart disease and an ample support of the observations in regards to inflammation as a remarkable link between these two devastating disorders.

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References

Anacker, C., Zunszain, P.A., Cattaneo, A., Carvalho, L.A., Garabedian, M.J., Thuret, S., Price, J., Pariante, C.M., 2011. Antidepressants increase human hippocampal neurogenesis by activating the glucocorticoid receptor. Mol. Psychiatry 16, 738–750.

Barton, B.E., 1996. The biological effects of interleukin 6. Med. Res. Rev. 16, 87–109.

Beck, A.T., Ward, C.H., Mendelson, M., Mock, J., Erbaugh, J., 1961. An inventory for measuring depression. Arch. Gen. Psychiatry 4, 561–571.

Beck, A.T., Steer, R.A., Carbin, M.G., 1988. Psychometric properties of the Beck Depression Inventory: twenty-five years of evaluation. Clin. Psychol. Rev. 8, 77–100.

Beekman, A.T., Geerlings, S.W., Deeg, D.J., Smit, J.H., Schoevers, R.S., de Beurs, E., Braam, A.W., Penninx, B.W., van Tilburg, W., 2002. The natural history of late-life depression: a 6-year prospective study in the community. Arch. Gen. Psychiatry 59, 605–611.

Berent, D., Macander, M., Szemraj, J., Orzechowska, A., Galecki, P., 2014. Vascular endothelial growth factor A gene expression level is higher in patients with major depressive disorder and not affected by cigarette smoking, hyperlipidemia or treatment with statins. Acta Neurobiol. Exp. (Wars) 74, 82–90.

Bhattacharyya, M.R., Molloy, G.J., Steptoe, A., 2008. Depression is associated with flatter cortisol rhythms in patients with coronary artery disease. J. Psychosom. Res. 65, 107–113.

Blann, A.D., Belgore, F.M., McCollum, C.N., Silverman, S., Lip, P.L., Lip, G.Y., 2002. Vascular endothelial growth factor and its receptor, Flt-1, in the plasma of patients with coronary or peripheral atherosclerosis, or Type II diabetes. Clin. Sci. (Lond.) 102, 187–194.

Blazer 2nd, D.G., 2000. Controversies in community-based psychiatric epidemiology: let the data speak for themselves. Arch. Gen. Psychiatry 57, 227–228.

Bruce, M.L., 2002. Psychosocial risk factors for depressive disorders in late life. Biol. Psychiatry 52, 175–184.

Byrne, A.M., Bouchier-Hayes, D.J., Harmey, J.H., 2005. Angiogenic and cell survival functions of vascular endothelial growth factor (VEGF). J. Cell Mol. Med. 9, 777–794.

Carney, R.M., Freedland, K.E., 2003. Depression, mortality, and medical morbidity in patients with coronary heart disease. Biol. Psychiatry 54, 241–247.

Carney, R.M., Rich, M.W., Tevelde, A., Saini, J., Clark, K., Freedland, K.E., 1988. The relationship between heart rate, heart rate variability and depression in patients with coronary artery disease. J. Psychosom. Res. 32, 159–164.

Carney, R.M., Freedland, K.E., Rich, M.W., Smith, L.J., Jaffe, A.S., 1993. Ventricular tachycardia and psychiatric depression in patients with coronary artery disease. Am. J. Med. 95, 23–28.

Carvalho, L.A., Juruena, M.F., Papadopoulos, A.S., Poon, L., Kerwin, R., Cleare, A.J., Pariante, C.M., 2008. Clomipramine in vitro reduces glucocorticoid receptor function in healthy subjects but not in patients with major depression. Neuropsychopharmacology 33, 3182–3189.

Carvalho, L.A., Garner, B.A., Dew, T., Fazakerley, H., Pariante, C.M., 2010. Antidepressants, but not antipsychotics, modulate GR function in human whole blood: an insight into molecular mechanisms. Eur. Neuropsychopharmacol. 20, 379–387.
Carvalho, L.A., Urbanova, L., Hamer, M., Hackett, R.A., Lazzarino, A.I., Steptoe, A., 2015. Blunted glucocorticoid and mineralocorticoid sensitivity to stress in people with diabetes. Psychoneuroendocrinology 51, 209–218.
Cattaneo, A., Gennarelli, M., Uher, R., Breen, G., Farmer, A., Aitchison, K.J., Craig, I.W., Anacker, C., Zunsztain, P.A., McGuffin, P., Pariante, C.M., 2013. Candidate genes expression profile associated with antidepressants response in the GENDEP study: differentiating between baseline 'predictors' and longitudinal 'targets'. Neuropsychopharmacology 38, 377–385.
Chrousos, G.P., Gold, P.W., 1992. The concepts of stress and stress system disorders. Overview of physical and behavioral homeostasis. JAMA 267, 1244–1252.
Clark-Raymond, A., Halaris, A., 2013. VEGF and depression: a comprehensive assessment of clinical data. J. Psychiatr. Res. 47, 1080–1087.
Cohen, S., Janicki-Deverts, D., Doyle, W.J., Miller, G.E., Frank, E., Rabin, B.S., Turner, R.B., 2012. Chronic stress, glucocorticoid receptor resistance, inflammation, and disease risk. Proc. Natl. Acad. Sci. 109, 5995–5999.
Crofford, L.J., Pillemer, S.R., Kalogeras, K.T., Cash, J.M., Michelson, D., Kling, M.A., Sternberg, E.M., Gold, P.W., Chrousos, G.P., Wilder, R.L., 1994. Hypothalamic–pituitary–adrenal axis perturbations in patients with fibromyalgia. Arthritis Rheum. 37, 1583–1592.
Dantzer, R., O'Connor, J.C., Lawson, M.A., Kelley, K.W., 2011. Inflammation-associated depression: from serotonin to kynurenine. Psychoneuroendocrinology 36, 426–436.
Davidson, K.W., 2012. Depression and coronary heart disease. ISRN Cardiol. 2012, 743813.
Demitrack, M.A., Crofford, L.J., 1998. Evidence for and pathophysiologic implications of hypothalamic–pituitary–adrenal axis dysregulation in fibromyalgia and chronic fatigue syndrome. Ann. N. Y. Acad. Sci. 840, 684–697.
Dome, P., Teleki, Z., Rihmer, Z., Peter, L., Dobos, J., Kenessey, I., Tovari, J., Timar, J., Paku, S., Kovacs, G., Dome, B., 2009. Circulating endothelial progenitor cells and depression: a possible novel link between heart and soul. Mol. Psychiatry 14, 523–531.
Dyer, A.R., Persky, V., Stamler, J., Paul, O., Shekelle, R.B., Berkson, D.M., Lepper, M., Schoenberger, J.A., Lindberg, H.A., 1980. Heart rate as a prognostic factor for coronary heart disease and mortality: findings in three Chicago epidemiologic studies. Am. J. Epidemiol. 112, 736–749.
Elovainio, M., Hurme, M., Jokela, M., Pulkki-Raback, L., Kivimaki, M., Hintsanen, M., Hintsa, T., Lehtimaki, T., Viikari, J., Raitakari, O.T., Keltikangas-Jarvinen, L., 2011. Moderating effect of indoleamine 2,3-dioxygenase (IDO) activation in the association between depressive symptoms and carotid atherosclerosis: evidence from the Young Finns study. J. Affect. Disord. 133, 611–614.
Empana, J.P., Sykes, D.H., Luc, G., Juhan-Vague, I., Arveiler, D., Ferrieres, J., Amouyel, P., Bingham, A., Montaye, M., Ruidavets, J.B., Haas, B., Evans, A., Jouven, X., Ducimetiere, P., 2005. Contributions of depressive mood and circulating inflammatory markers to coronary heart disease in healthy European men: the Prospective Epidemiological Study of Myocardial Infarction (PRIME). Circulation 111, 2299–2305.
Ershler, W.B., Keller, E.T., 2000. Age-associated increased interleukin-6 gene expression, late-life diseases, and frailty. Annu. Rev. Med. 51, 245–270.
Ferrucci, L., Harris, T.B., Guralnik, J.M., Tracy, R.P., Corti, M.C., Cohen, H.J., Penninx, B., Pahor, M., Wallace, R., Havlik, R.J., 1999. Serum IL-6 level and the development of disability in older persons. J. Am. Geriatr. Soc. 47, 639–646.
Fleisch, M., Billinger, M., Eberli, F.R., Garachemani, A.R., Meier, B., Seiler, C., 1999. Physiologically assessed coronary collateral flow and intracoronary growth factor concentrations in patients with 1- to 3-vessel coronary artery disease. Circulation 100, 1945–1950.
Fries, E., Hesse, J., Hellhammer, J., Hellhammer, D.H., 2005. A new view on hypocortisolism. Psychoneuroendocrinology 30, 1010–1016.
Gold, P.W., Chrousos, G.P., 2002. Organization of the stress system and its dysregulation in melancholic and atypical depression: high vs low CRH/NE states. Mol. Psychiatry 7, 254–275.
Halaris, A., 2013. Inflammation, heart disease, and depression. Curr. Psychiatry Rep. 15, 400.
Hansson, G.K., 2005. Inflammation, atherosclerosis, and coronary artery disease. N. Engl. J. Med. 352, 1685–1695.
Harris, T.B., Ferrucci, L., Tracy, R.P., Corti, M.C., Wacholder, S., Ettinger Jr., W.H., Heimovitz, H., Cohen, H.J., Wallace, R., 1999. Associations of elevated interleukin-6 and C-reactive protein levels with mortality in the elderly. Am. J. Med. 106, 506–512.
Hellhammer, J., Schlotz, W., Stone, A.A., Pirke, K.M., Hellhammer, D., 2004. Allostatic load, perceived stress, and health: a prospective study in two age groups. Ann. N. Y. Acad. Sci. 1032, 8–13.
Herve, C., Beyne, P., Jamault, H., Delacoux, E., 1996. Determination of tryptophan and its kynurenine pathway metabolites in human serum by high-performance liquid chromatography with simultaneous ultraviolet and fluorimetric detection. J. Chromatogr. B Biomed. Appl. 675, 157–161.
Hsuchou, H., Kastin, A.J., Mishra, P.K., Pan, W., 2012. C-reactive protein increases BBB permeability: implications for obesity and neuroinflammation. Cell. Physiol. Biochem. 30, 1109–1119.
Iga, J., Ueno, S., Yamauchi, K., Numata, S., Tayoshi-Shibuya, S., Kinouchi, S., Nakataki, M., Song, H., Hokoishi, K., Tanabe, H., Sano, A., Ohmori, T., 2007. Gene expression and association analysis of vascular endothelial growth factor in major depressive disorder. Prog. Neuropsychopharmacol. Biol. Psychiatry 31, 658–663.
Ikeda, U., Ito, T., Shimada, K., 2001. Interleukin-6 and acute coronary syndrome. Clin. Cardiol. 24, 701–704.
Irwin, M.R., Miller, A.H., 2007. Depressive disorders and immunity: 20 years of progress and discovery. Brain Behav. Immun. 21, 374–383.
Isung, J., Mobarrez, F., Nordstrom, P., Asberg, M., Jokinen, J., 2012. Low plasma vascular endothelial growth factor (VEGF) associated with completed suicide. World J. Biol. Psychiatry 13, 468–473.
Jiang, W., Krishnan, R.R., O'Connor, C.M., 2002. Depression and heart disease: evidence of a link, and its therapeutic implications. CNS Drugs 16, 111–127.
Jin, K., Zhu, Y., Sun, Y., Mao, X.O., Xie, L., Greenberg, D.A., 2002. Vascular endothelial growth factor (VEGF) stimulates neurogenesis in vitro and in vivo. Proc. Natl. Acad. Sci. USA 99, 11946–11950.
Kahl, K.G., Bens, S., Ziegler, K., Rudolf, S., Kordon, A., Dibbelt, L., Schweiger, U., 2009. Angiogenic factors in patients with current major depressive disorder comorbid with borderline personality disorder. Psychoneuroendocrinology 34, 353–357.
Katsuura, S., Kamezaki, Y., Yamagishi, N., Kuwano, Y., Nishida, K., Masuda, K., Tanahashi, T., Kawai, T., Arisawa, K., Rokutan, K., 2011. Circulating vascular endothelial growth factor is independently and negatively associated with trait anxiety and depressive mood in healthy Japanese university students. Int. J. Psychophysiol. 81, 38–43.
Keck, P.J., Hauser, S.D., Krivi, G., Sanzo, K., Warren, T., Feder, J., Connolly, D.T., 1989. Vascular permeability factor, an endothelial cell mitogen related to PDGF. Science 246, 1309–1312.
Knijff, E.M., Nadine Breunis, M., Kupka, R.W., De wit, H.J., Ruwhof, C., Akkerhuis, G.W., Nolen, W.A., Drexhage, H.A., 2007. An imbalance in the production of IL-1β and IL-6 by monocytes of bipolar patients: restoration by lithium treatment. Bipolar Disord. 9, 743–753.
Kohn, R., Epstein-Lubow, G., 2006. Course and outcomes of depression in the elderly. Curr. Psychiatry Rep. 8, 34–40.
Kop, W.J., Gottdiener, J.S., 2005. The role of immune system parameters in the relationship between depression and coronary artery disease. Psychosom. Med. 67 (Suppl. 1), S37–S41.
Kotan, Z., Sarandol, E., Kirhan, E., Ozkaya, G., Kirli, S., 2012. Serum brain-derived neurotrophic factor, vascular endothelial growth factor and leptin levels in patients with a diagnosis of severe major depressive disorder with melancholic features. Ther. Adv. Psychopharmacol. 2, 65–74.
Lapin, I.P., Oxenkrug, G.F., 1969. Intensification of the central serotoninergic processes as a possible determinant of the thymoleptic effect. Lancet 1, 132–136.
Lee, B.H., Kim, Y.K., 2012. Increased plasma VEGF levels in major depressive or manic episodes in patients with mood disorders. J. Affect. Disord. 136, 181–184.
Lewis, G., Pelosi, A.J., Araya, R., Dunn, G., 1992. Measuring psychiatric disorder in the community: a standardized assessment for use by lay interviewers. Psychol. Med. 22, 465–486.
Libby, P., 2006. Inflammation and cardiovascular disease mechanisms. Am. J. Clin. Nutr. 83, 456S–460S.
Licht, T., Goshen, I., Avital, A., Kreisel, T., Zubedat, S., Eavri, R., Segal, M., Yirmiya, R., Keshet, E., 2011. Reversible modulations of neuronal plasticity by VEGF. Proc. Natl. Acad. Sci. 108, 5081–5086.
Licinio, J., Yildiz, B., Wong, M.L., 2002. Depression and cardiovascular disease: co-occurrence or shared genetic substrates? Mol. Psychiatry 7, 1031–1032.
Maes, M., Ruckoanich, P., Chang, Y.S., Mahanonda, N., Berk, M., 2011. Multiple aberrations in shared inflammatory and oxidative & nitrosative stress (IO&NS) pathways explain the co-association of depression and cardiovascular disorder (CVD), and the increased risk for CVD and due mortality in depressed patients. Prog. Neuropsychopharmacol. Biol. Psychiatry 35, 769–783.
Matsuzaki, H., Tamatani, M., Yamaguchi, A., Namikawa, K., Kiyama, H., Vitek, M.P., Mitsuda, N., Tohyama, M., 2001. Vascular endothelial growth factor rescues hippocampal neurons from glutamate-induced toxicity: signal transduction cascades. FASEB J. 15, 1218–1220.
McCaffery, J.M., Frasure-Smith, N., Dubé, M.-P., Théroux, P., Rouleau, G.A., Duan, Q., Lespérance, F., 2006. Common genetic vulnerability to depressive symptoms and coronary artery disease: a review and development of candidate genes related to inflammation and serotonin. Psychosom. Med. 68, 187–200.
McCloskey, D.P., Croll, S.D., Scharfman, H.E., 2005. Depression of synaptic transmission by vascular endothelial growth factor in adult rat hippocampus and evidence for increased efficacy after chronic seizures. J. Neurosci. 25, 8889–8897.
McEwen, B.S., 1998. Stress, adaptation, and disease. Allostasis and allostatic load. Ann. N. Y. Acad. Sci. 840, 33–44.
Meijer, A., Conradi, H.J., Bos, E.H., Thombs, B.D., van Melle, J.P., de Jonge, P., 2011. Prognostic association of depression following myocardial infarction with mortality and cardiovascular events: a meta-analysis of 25 years of research. Gen. Hosp. Psychiatry 33, 203–216.
Miller, A.H., Pariante, C.M., Pearce, B.D., 1999. Effects of cytokines on glucocorticoid receptor expression and function. Glucocorticoid resistance and relevance to depression. Adv. Exp. Med. Biol. 461, 107–116.
Miller, G.E., Stetler, C.A., Carney, R.M., Freedland, K.E., Banks, W.A., 2002. Clinical depression and inflammatory risk markers for coronary heart disease. Am. J. Cardiol. 90, 1279–1283.
Miller, G.E., Freedland, K.E., Carney, R.M., 2005. Depressive symptoms and the regulation of proinflammatory cytokine expression in patients with coronary heart disease. J. Psychosom. Res. 59, 231–236.

Miller, G.E., Chen, E., Sze, J., Marin, T., Arevalo, J.M., Doll, R., Ma, R., Cole, S.W., 2008. A functional genomic fingerprint of chronic stress in humans: blunted glucocorticoid and increased NF-kappaB signaling. Biol. Psychiatry 64, 266–272.
Molloy, G.J., Perkins-Porras, L., Strike, P.C., Steptoe, A., 2008. Type-D personality and cortisol in survivors of acute coronary syndrome. Psychosom. Med. 70, 863–868.
Mondelli, V., Dazzan, P., Hepgul, N., di Forti, M., Aas, M., D'Albenzio, A., di Nicola, M., Fisher, H., Handley, R., Marques, T.R., Morgan, C., Navari, S., Taylor, H., Papadopoulos, A., Aitchison, K.J., Murray, R.M., Pariante, C.M., 2010. Abnormal cortisol levels during the day and cortisol awakening response in first-episode psychosis: the role of stress and of antipsychotic treatment. Schizophr. Res. 116, 234–242.
Morrison, M.F., Redei, E., Tenhave, T., Parmelee, P., Boyce, A.A., Sinha, P.S., Katz, I.R., 2000. Dehydroepiandrosterone sulfate and psychiatric measures in a frail, elderly residential care population. Biol. Psychiatry 47, 144–150.
Morrison, M.F., ten Have, T., Freeman, E.W., Sammel, M.D., Grisso, J.A., 2001. DHEA-S levels and depressive symptoms in a cohort of African American and Caucasian women in the late reproductive years. Biol. Psychiatry 50, 705–711.
Myint, A.M., Kim, Y.K., 2003. Cytokine–serotonin interaction through IDO: a neurodegeneration hypothesis of depression. Med. Hypotheses 61, 519–525.
Myint, A.M., Kim, Y.K., Verkerk, R., Scharpe, S., Steinbusch, H., Leonard, B., 2007. Kynurenine pathway in major depression: evidence of impaired neuroprotection. J. Affect. Disord. 98, 143–151.
Myint, A.M., Schwarz, M.J., Muller, N., 2012. The role of the kynurenine metabolism in major depression. J. Neural Transm. 119, 245–251.
Niinisalo, P., Raitala, A., Pertovaara, M., Oja, S.S., Lehtimaki, T., Kahonen, M., Reunanen, A., Jula, A., Moilanen, L., Kesaniemi, Y.A., Nieminen, M.S., Hurme, M., 2008. Indoleamine 2,3-dioxygenase activity associates with cardiovascular risk factors: the Health 2000 study. Scand. J. Clin. Lab. Invest. 68, 767–770.
Oades, R.D., Dauvermann, M.R., Schimmelmann, B.G., Schwarz, M.J., Myint, A.M., 2010. Attention-deficit hyperactivity disorder (ADHD) and glial integrity: S100B, cytokines and kynurenine metabolism – effects of medication. Behav. Brain Funct. 6, 29.
Oldehinkel, A.J., van den Berg, M.D., Flentge, F., Bouhuys, A.L., ter Horst, G.J., Ormel, J., 2001. Urinary free cortisol excretion in elderly persons with minor and major depression. Psychiatry Res. 104, 39–47.
Otte, C., Marmar, C.R., Pipkin, S.S., Moos, R., Browner, W.S., Whooley, M.A., 2004. Depression and 24-hour urinary cortisol in medical outpatients with coronary heart disease: The Heart and Soul Study. Biol. Psychiatry 56, 241–247.
Ozkan, Y., Sukuroglu, M.K., Tulmac, M., Kisa, U., Simsek, B., 2014. Relation of kynurenine/tryptophan with immune and inflammatory markers in coronary artery disease. Clin. Lab. 3, 4.
Pace, T.W., Hu, F., Miller, A.H., 2007. Cytokine-effects on glucocorticoid receptor function: relevance to glucocorticoid resistance and the pathophysiology and treatment of major depression. Brain Behav. Immun. 21, 9–19.
Pariante, C.M., 2006. The glucocorticoid receptor: part of the solution or part of the problem? J. Psychopharmacol. 20, 79–84.
Pariante, C.M., Miller, A.H., 2001. Glucocorticoid receptors in major depression: relevance to pathophysiology and treatment. Biol. Psychiatry 49, 391–404.
Parissis, J.T., Adamopoulos, S., Rigas, A., Kostakis, G., Karatzas, D., Venetsanou, K., Kremastinos, D.T., 2004. Comparison of circulating proinflammatory cytokines and soluble apoptosis mediators in patients with chronic heart failure with versus without symptoms of depression. Am. J. Cardiol. 94, 1326–1328.
Pedersen, E.R., Midttun, Ø., Ueland, P.M., Schartum-Hansen, H., Seifert, R., Igland, J., Nordrehaug, J.E., Ebbing, M., Svingen, G., Bleie, Ø., 2011. Systemic markers of interferon-γ-mediated immune activation and long-term prognosis in patients with stable coronary artery disease. Arterioscler. Thromb. Vasc. Biol. 31, 698–704.
Penninx, B.W., Beekman, A.T., Bandinelli, S., Corsi, A.M., Bremmer, M., Hoogendijk, W.J., Guralnik, J.M., Ferrucci, L., 2007. Late-life depressive symptoms are associated with both hyperactivity and hypoactivity of the hypothalamo-pituitary–adrenal axis. Am. J. Geriatr. Psychiatry 15, 522–529.
Pertovaara, M., Raitala, A., Juonala, M., Lehtimaki, T., Huhtala, H., Oja, S.S., Jokinen, E., Viikari, J.S., Raitakari, O.T., Hurme, M., 2007. Indoleamine 2,3-dioxygenase enzyme activity correlates with risk factors for atherosclerosis: the Cardiovascular Risk in Young Finns Study. Clin. Exp. Immunol. 148, 106–111.
Pfaffl, M.W., 2001. A new mathematical model for relative quantification in real-time RT-PCR. Nucleic Acids Res. 29, e45.
Pizzi, C., Manzoli, L., Mancini, S., Costa, G.M., 2008. Analysis of potential predictors of depression among coronary heart disease risk factors including heart rate variability, markers of inflammation, and endothelial function. Eur. Heart J. 29, 1110–1117.
Pruessner, J.C., Kirschbaum, C., Meinlschmid, G., Hellhammer, D.H., 2003. Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change. Psychoneuroendocrinology 28, 916–931.
Raison, C.L., Miller, A.H., 2003. When not enough is too much: the role of insufficient glucocorticoid signaling in the pathophysiology of stress-related disorders. Am. J. Psychiatry 160, 1554–1565.
Ridker, P.M., 2003. Clinical application of C-reactive protein for cardiovascular disease detection and prevention. Circulation 107, 363–369.
Ridker, P.M., 2007. C-reactive protein and the prediction of cardiovascular events among those at intermediate risk: moving an inflammatory hypothesis toward consensus. J. Am. Coll. Cardiol. 49, 2129–2138.
Rohleder, N., Kudielka, B.M., Hellhammer, D.H., Wolf, J.M., Kirschbaum, C., 2002. Age and sex steroid-related changes in glucocorticoid sensitivity of pro-inflammatory cytokine production after psychosocial stress. J. Neuroimmunol. 126, 69–77.
Rosti, V., Massa, M., Campanelli, R., de Amici, M., Piccolo, G., Perfetti, V., 2007. Vascular endothelial growth factor promoted endothelial progenitor cell mobilization into the peripheral blood of a patient with POEMS syndrome. Haematologica 92, 1291–1292.
Sapolsky, R.M., Romero, L.M., Munck, A.U., 2000. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. Endocr. Rev. 21, 55–89.
Sarkar, S., Kalia, V., Montelaro, R.C., 2003. Caspase-mediated apoptosis and cell death of rhesus macaque CD4+ T-cells due to cryopreservation of peripheral blood mononuclear cells can be rescued by cytokine treatment after thawing. Cryobiology 47, 44–58.
Senger, D.R., Perruzzi, C.A., Feder, J., Dvorak, H.F., 1986. A highly conserved vascular permeability factor secreted by a variety of human and rodent tumor cell lines. Cancer Res. 46, 5629–5632.
Shalev, H., Serlin, Y., Friedman, A., 2009. Breaching the blood–brain barrier as a gate to psychiatric disorder. Cardiovasc. Psychiatry Neurol. 2009, 278531.
Sondell, M., Lundborg, G., Kanje, M., 1999. Vascular endothelial growth factor has neurotrophic activity and stimulates axonal outgrowth, enhancing cell survival and Schwann cell proliferation in the peripheral nervous system. J. Neurosci. 19, 5731–5740.
Stewart, J.C., Rand, K.L., Muldoon, M.F., Kamarck, T.W., 2009. A prospective evaluation of the directionality of the depression–inflammation relationship. Brain Behav. Immun. 23, 936–944.
Storkebaum, E., Lambrechts, D., Carmeliet, P., 2004. VEGF: once regarded as a specific angiogenic factor, now implicated in neuroprotection. BioEssays 26, 943–954.
Strickland, P.L., Deakin, J.F., Percival, C., Dixon, J., Gater, R.A., Goldberg, D.P., 2002. Bio-social origins of depression in the community. Interactions between social adversity, cortisol and serotonin neurotransmission. Br. J. Psychiatry 180, 168–173.
Takebayashi, M., Hashimoto, R., Hisaoka, K., Tsuchioka, M., Kunugi, H., 2010. Plasma levels of vascular endothelial growth factor and fibroblast growth factor 2 in patients with major depressive disorders. J. Neural Transm. 117, 1119–1122.
Tylee, A., Ashworth, M., Barley, E., Brown, J., Chambers, J., Farmer, A., Fortune, Z., Haddad, M., Lawton, R., Mann, A., Mehay, A., McCrone, P., Murray, J., Leese, M., Pariante, C.M., Rose, D., Rowlands, G., Smith, A., Walters, P., 2011. Up-beat UK: a programme of research into the relationship between coronary heart disease and depression in primary care patients. BMC Fam. Pract. 12, 38.
Ventriglia, M., Zanardini, R., Pedrini, L., Placentino, A., Nielsen, M.G., Gennarelli, M., Bocchio-Chiavetto, L., 2009. VEGF serum levels in depressed patients during SSRI antidepressant treatment. Prog. Neuropsychopharmacol. Biol. Psychiatry 33, 146–149.
Walters, P., Barley, E.A., Mann, A., Phillips, R., Tylee, A., 2014. Depression in primary care patients with coronary heart disease: baseline findings from the UPBEAT UK study. PLoS ONE 9, e98342.
Warner-Schmidt, J.L., Duman, R.S., 2008. VEGF as a potential target for therapeutic intervention in depression. Curr. Opin. Pharmacol. 8, 14–19.
Webster, J.C., Oakley, R.H., Jewell, C.M., Cidlowski, J.A., 2001. Proinflammatory cytokines regulate human glucocorticoid receptor gene expression and lead to the accumulation of the dominant negative beta isoform: a mechanism for the generation of glucocorticoid resistance. Proc. Natl. Acad. Sci. USA 98, 6865–6870.
Whooley, M.A., Wong, J.M., 2013. Depression and cardiovascular disorders. Annu. Rev. Clin. Psychol. 9, 327–354.
Wichers, M., Maes, M., 2002. The psychoneuroimmuno-pathophysiology of cytokine-induced depression in humans. Int. J. Neuropsychopharmacol. 5, 375–388.
Wichers, M.C., Maes, M., 2004. The role of indoleamine 2,3-dioxygenase (IDO) in the pathophysiology of interferon-alpha-induced depression. J. Psychiatry Neurosci. 29, 11–17.
Wirleitner, B., Rudzite, V., Neurauter, G., Murr, C., Kalnins, U., Erglis, A., Trusinskis, K., Fuchs, D., 2003. Immune activation and degradation of tryptophan in coronary heart disease. Eur. J. Clin. Invest. 33, 550–554.
Yehuda, R., 2001. Biology of posttraumatic stress disorder. J. Clin. Psychiatry 62 (Suppl. 17), 41–46.
Yoshimura, K., Yamada, M., Kajiwara, Y., Nishiguchi, S., Aoyama, T., 2013. Relationship between depression and risk of malnutrition among community-dwelling young–old and old–old elderly people. Aging Ment. Health 17, 456–460.