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How acute and chronic physical disease may influence mental health - an analysis of neurotransmitter precursor amino acid levels

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Highlights

- Patients with somatic diseases are more likely to develop depression
- Changes in neurotransmitter precursor amino acids changes might be the pathophysiological link
- A prospective study was performed in 177 patients
- phenylalanine/tyrosine ratio was related to the factors acute physical disease and depression

- kynurenine/tryptophan was associated with chronic physical disease

Patients with somatic diseases are more likely to develop depression than physically healthy individuals, and comorbid depression has been shown to incrementally worsen patients´ health. Physical conditions are known to influence neurotransmitter precursor amino acids, changes in which are associated with depressive symptoms. In this prospective study we investigated neurotransmitter precursor amino acids levels in patients with acute and chronic physical disease and evaluated their association with depressive symptoms.

177 subjects with and without chronic medical comorbidity (factor: chronic physical disease) admitted to the trauma and orthopaedic surgery ward for a surgical intervention (factor: acute physical disease) were included in the analysis. Chronic medical comorbidity was scored using Charlson Index and depressive and anxiety symptoms using the Hospital Anxiety and Depression Scale (HADS, factor: mental health). The effect of covariates was also evaluated. C-reactive protein (CRP), neopterin, kynurenine/tryptophan (KYN/TRP) and phenylalanine/tyrosine (PHE/TYR) were analysed by HPLC or ELISA prior to surgery and at discharge. Mixed Model as well as correlation analyses were performed.

CRP and neopterin levels were influenced by the factors “acute physical disease” (both $p<0.001$) and “chronic physical disease” ($p=0.024$, $p=0.001$ respectively). PHE/TYR, an index of the catecholamine pathway) was related to the factors “acute physical disease” ($p<0.001$) and “mental health-depression” ($p=0.012$), while KYN/TRP (an index of the kynurenine pathway affecting also serotonin) was associated with “chronic physical disease” ($p=0.005$). No significant effect of “mental health-anxiety” was found. The effect of “mental health-depression” on PHE/TYR was more pronounced in females (gender $p=0.003$). Differences in HADS depression values correlated with changes in PHE/TYR and both correlated with CRP values.

In conclusion, inflammatory reactions related to acute or chronic physical conditions can influence the availability of neurotransmitter precursor amino acid levels and these changes are associated with mental health.

Key Words: kynurenine, tryptophan, phenylalanine, tyrosine, trauma surgery, depression, medical comorbidity, inflammation

1. Introduction

More than 300 million people of all ages around the world suffer from depression (Association). The risk for depression is increased in individuals with one or more chronic physical diseases: between 9.3% and 23.0% of participants with one or more chronic physical disease have comorbid depression which is higher than the numbers for physically healthy individuals. The combination of physical disease and depression incrementally worsens health compared with depression alone, with chronic diseases alone, or with a combination chronic physical diseases without depression (Moussavi et al., 2007). While it is now established that several pathogenetic mechanisms can lead to the phenotype of depression (Menard et al., 2016), accumulating evidence indicates a role for the immune system in the aetiology of depression as well as its common physical comorbidities (Khandaker et al., 2017). “Sickness behaviour” occurs in acute systemic inflammatory diseases and includes, in addition to physical symptoms, mental changes such as fatigue, cognitive disturbances, irritability and depressed mood (Dantzer et al., 2008). Additionally depressive symptoms are common in people with a chronic inflammatory illness such as rheumatoid arthritis (Dickens and Creed, 2001) or following treatment with interferons (Bonaccorso et al., 2002). Associations between circulating Interleukin (IL)-6 and C-reactive protein (CRP) concentrations and subsequent development and persistence of depressive symptoms have been found (Zalli et al., 2016). Inflammation often precedes depression and could thus be a causal risk factor for the illness (Khandaker et al., 2017). However, depression and stress can also cause an inflammatory reaction and thus immune activation and depressive mood may feed each other and enter a vicious cycle (Strasser et al., 2017).

The peripheral immune response could interact with the brain through several mechanisms such as afferent nerves, humoral pathways or inflammation-induced changes in neurotransmitter precursor monoamines (Dantzer et al., 2008). Cytokines can affect the catecholamine synthesis by altering the availability of the enzyme co-factor tetrahydrobiopterin (BH4) which is necessary for the conversion of phenylalanine (PHE) to tyrosine (TYR) by phenylalanine hydroxylase (PAH) (Sperner-Unterweger et al., 2014). PHE/TYR is a marker of PAH activity (Figure 1). Cytokines can also influence the serotonin pathway through activation of indoleamine 2,3-dioxygenase-1 (IDO) (Oxenkrug, 2010) which is involved in the breakdown of tryptophan (TRP) to kynurenine (KYN) but also accepts serotonin as a substrate (Capuron et al., 2003). KYN/TRP as a marker of TRP breakdown which, in case of an association with an immune activation background, is likely related to IDO activity (Widner et al., 2002) (Figure 1). Some downstream products of KYN are neurotoxic (Dantzer, 2017).

For mental disorders, monoamine depletion studies have shown that changes in mood can be induced in a vulnerable population (people who have a familial history of major depressive disorders or are drug-free in remission after an episode of major depression) within short periods of time (hours to days) (Ruhe et al., 2007). In acute physical disease, PHE/TYR was shown to be elevated in sepsis as compared to local cerebral infections (Conejero et al., 1987) and in patients following general anaesthesia compared to local anaesthesia (Hol et al., 2009), where it was associated with increased pain and prolonged hospital stay. Chronic physical diseases (many of which are associated with chronic low grade inflammation), e.g. HIV (Bipath et al., 2015), cancer (Hufner et al., 2015) or atherosclerosis (Baumgartner et al., 2017) have been shown to be associated with changes in the KYN pathway.

In the present study we investigate the pathophysiological basis behind the association of depressive symptoms with acute and chronic physical disease by analysing changes in CRP,

neopterin as well as KYN/TRP and PHE/TYR in patients with and without physical and mental comorbidities.

2. Methods

2.1. Ethics Statement

The study was approved by the ethics committee of Medical University Innsbruck, Austria. Written, informed consent was obtained from all participants prior to inclusion in the study.

2.2. Participants

In this study we prospectively analysed a sample of unselected patients admitted to the orthopaedic and trauma surgery ward for an intervention. 186 patients were included in the study of which 177 were included in the final analysis. Nine patients were excluded due to the following reasons: three excluded due to no blood drawn and six due to ISS (injury severity score) >= 10. ISS score <10 was used as a criterion so that only patients with minor traumatic injury such as ruptured ligaments were included. In 44 patients only one blood draw was available (either baseline (PRE) or discharge (POST)). Medical comorbidities were assessed using Charlson Index (Charlson et al., 1987). The Charlson comorbidity index is the most widely used comorbidity index which contains 19 factors. A continuous as well as a yes/no rating was used for our analyses since the original index was developed for oncology and in other settings a categorisation has been shown to be useful (Gregersen et al., 2017). Basic demographic and disease-related data were obtained from all participants (table 1, missing values below 10% in all cases).

2.2.1. Inclusion criteria:

- Age >18 years
- Sufficient command of the German language
- Informed written consent
- Admission to trauma and orthopedic surgery ward
- Inpatient therapy for a minimum of 1 night

2.2.3. Exclusion criteria:

- History of organ transplantation
- Patients with ISS score≥10
- Inpatient therapy >14 days

2.3. Psychometric assessment and pain assessment

Severity of depressive symptoms and anxiety at the time of the blood sampling were assessed using the Hospital Anxiety and Depression Scale (HADS) (Herrmann, 1997; Zigmond and Snaith, 1983). Dichotomization of the HADS values for the diagnosis of clinically relevant depressive symptoms was done using values retrieved from the literature (Bjelland et al., 2002).

2.4. Blood sampling and serum/plasma preparation

Blood was drawn between 7 am and 8:30 am prior to surgery (PRE) and following surgery prior to discharge (POST) during morning study visits (fasting) from an antecubital vein. Aliquots of serum were shock frozen in liquid nitrogen and stored at −80◦C until use.

2.5. Analysis of CRP, neopterin and neurotransmitter precursor monoamines

CRP was measured as measured in the central laboratory using Immunoturbidimetric assay (Roche MODULAR, Roche Diagnostics, Mannheim, Germany). Neopterin concentrations were measured by enzyme-linked immunosorbent assay (BRAHMS Diagnostics, Berlin, Germany). TRP and KYN serum concentrations as well as concentrations of PHE and TYR were determined by high-performance liquid chromatography, as described elsewhere (Neurauter et al., 2008a; Widner et al., 1997). The ratios of KYN/TRP were calculated as indices of tryptophan breakdown by IDO and PHE/TYR reflecting PAH activity respectively (Capuron et al., 2011).

2.6. Statistical analysis

Data are presented as percentages, means, standard deviation and confidence intervals. A mixed model analysis was performed for testing differences in immune parameter concentrations across conditions, i.e. the main effects of acute surgery “acute physical disease” (PRE vs POST), chronic medical comorbidity “chronic physical disease” (Yes vs No) and “mental health-depression” (Yes vs No). All factors were dichotomized for clarity reasons. The model investigated the main effects of acute and chronic physical disease as well as mental health-depression as well as their interaction effect on CRP, neopterin and neurotransmitter precursor amino acids. Variables with non-normal distribution were log-transformed or square root-transformed to approximate their distribution of normality. The following confounding variables were analysed for inclusion in the model: gender (male/female), interaction gender*depression, alcohol intake (none/occasionally/regularly), smoking (yes/no), SSRI treatment (yes/no), physical activity (yes/no), antidepressant/antipsychotic treatment (yes/no), anti-inflammatory medication (yes/no). Variables which showed a strong intercorrelation with one of the main factors were not included for statistical reasons. The variables where analysed in a univariate model and included into the multivariate model if the significance reached the inclusive threshold of p=0.10. Non-significant results were removed in a stepwise procedure. In case of a significant impact of any of the variables we adjusted for it. A power analysis for a two-way ANCOVA with main effects and interactions and two covariates resulted in detectable interaction effect sizes of at least f=0.25 for a sample size of 177 an alpha of 0.05, and a power of 0.80. Group comparisons were performed using Mann Whitney U test for continuous and Chi Square test for categorical variables with non-normal distribution. $P<0.05$ was considered significant in all analyses. Spearman rank correlation analyses were performed and $p<0.05$, was considered significant. All statistical analyses were performed using SPSS 20.0.

3. Results

3.1. Sample characteristics

In this study we prospectively analysed a sample of 177 unselected patients admitted to the orthopaedic and trauma surgery ward for an intervention. Important demographic and clinical data of the study population are given in table 1. Using HADS depression subscale 11.5 % of patients showed clinically significant depressive symptoms prior to surgery (9.3% of males and 14.7% of females Chi Square p=0.326) and 17.9 % (14.3% of males and 23% of females Chi Square test p=0.195) post surgery (Chi Square Test p=0.110). HADS anxiety subscale scores showed clinically significant anxiety in 18.2% prior to surgery and in 15.9 % post surgery (Chi Square test p=0.588).

3.2. Effect of acute and chronic physical disease on neurotransmitter precursor amino acids and their association with mental health-depression

The analysis of the effects of the factors acute physical disease, chronic physical disease and mental health-depression and their interaction was performed using a mixed model analysis. Additionally we evaluated the effect of potential confounders: gender, alcohol intake, smoking, physical activity, SSRI treatment, antidepressant/antipsychotic treatment, anti-inflammatory medication and in case of significant influence adjusted for these factors.

3.2.1. CRP and neopterin

CRP as a general marker of inflammation was influenced by “acute physical disease” ($p<0.001$; table 2), and “chronic physical disease” ($p=0.024$) while no effect of “mental health-depression” ($p=0.259$) was found. Neopterin as a marker of Th1-related inflammation was influenced by the factors “acute physical disease” ($p<0.001$; table 2) and “chronic physical disease” ($p=0.001$) but showed no association with “mental health-depression” ($p=0.296$). The interaction of acute physical disease*chronic physical disease was significant ($p=0.039$).

3.2.2. Phenylalanine/tyrosine pathway

A mixed model analysis showed that PHE/TYR was influenced by the factor “acute physical disease” ($p<0.001$; table 2) and showed an association with “mental health-depression”

(p=0.012) but not “chronic physical disease” (p=0.517). This was due to an increase in PHE for the condition “acute physical disease” and an increase in PHE and a decrease in TYR for the condition “mental health depression” (see supplementary table 1). The effect of “gender” was significant (p=0.003) with females showing higher PHE/TYR than males, this was mainly due to females with depression who showed the highest ratios (the interaction of “mental health-depression” and “gender” was significant p=0.001). The results of separate analysis of male and female data (supplemental table 2) underline this effect. When the analysis was performed by entering the HADS depression subscale score into the model (instead of a yes/no rating) the results remained unchanged (factor “acute physical disease” p<0.001, factor “mental health-depression” p=0.019).

3.2.3. Kynurenine/tryptophan pathway

KYN/TRP was found to be associated with the factor “chronic physical disease” (p=0.005; table 2) but not “acute physical disease” (p=0.156) or “mental health-depression” (p=0.828) using mixed model analysis. This effect was due to higher values of KYN and lower concentrations of TRP in the condition “chronic physical disease” (see supplementary table 1). No significant interactions of these factors were found. When the analysis was performed by entering the Charlson comorbidity index score into the model (instead of a yes/no rating) the results remained unchanged (KYN/TRP was found to be associated with the factor “chronic physical disease”; p<0.001).

3.3. Effect of acute and chronic physical disease on neurotransmitter precursor amino acids and their association with mental health-anxiety

When the mixed model analyses described above where performed with the factors “acute physical disease”, “chronic physical disease” and “mental health–anxiety” the results for “acute physical disease” and “chronic physical disease” were comparable to the main effects described above concerning CRP (significant effects of “acute physical disease” and “chronic physical disease”), neopterin (significant effects of “acute physical disease” and “chronic

physical disease"), PHE/TYR (significant effect of "acute physical disease"), KYN/TRP (significant effect "chronic physical disease"). However, the factor "mental health–anxiety" did not reach significance in any of the analyses. For PHE/TYR the interaction of mental health-anxiety*gender was significant (p=0.019).

3.4. Changes in Phenylalanine/tyrosine pathway correlate with changes in depressive symptoms

Changes in HADS depression subscale score prior to and following surgery correlated with the differences in PHE/TYR (r=0.197, p=0.015), and both differences correlated with the measured CRP (difference in HADS values r=0.195, p=0.010; difference in PHE/TYR: r=0.172, p=0.024; Figure 2). No correlation with KYN/TRP was found.

4. Discussion

In the present study we analyze the brain immune axis by looking at changes in neurotransmitter precursor amino acids concentrations and their relation to physical and mental disease. Higher PHE/TYR (catecholamine neurotransmitter pathway) were associated with acute physical disease and depressive symptoms while elevated KYN/TRP ratios (related to the serotonin neurotransmitter pathway) were associated with chronic physical conditions. The study provides evidence that changes in both monoamine neurotransmitter pathways could be important in linking physical morbidity and depressive symptoms.

4.1. The role of CRP and neopterin in depression associated with physical disease

Neopterin is a metabolite of guanosine triphosphate which is produced by human monocytes/macrophages through their activation by pro-inflammatory stimuli like T-cell-derived IFN-γ (Fuchs et al., 1992). The Th1-type immune response has been shown to induce IDO via IFN-γ which leads to degradation of TRP to KYN (Murr et al., 2000). In the current study we found associations of neopterin and CRP with acute and chronic physical disease, but no association with depressive symptoms. Earlier studies have shown an association of neopterin concentrations in patients with depression and/or anxiety (Muller, 2014). However, this is not true for all patients with depression (de Menezes et al., 2017). One should take into account that the patient population in our study was in general a psychiatrically healthy one: only 7.3% of the patients had previously been given a diagnosis of depression (not necessarily by a psychiatrist) which is in line with numbers in the general population (Kessler and Bromet, 2013). The point prevalence of depressive symptoms (11.5% PRE and 17.9% POST) resulted in a relatively low absolute number of patients with depression being evaluated in this study. This could explain why no association with inflammatory markers was found since inflammatory markers are influenced by such a multitude of factors and are thus maybe not a sensitive enough measure, especially since values below 0.05 mg/dl CRP were not quantified. However, CRP was associated with changes in the HADS depression subscale pre and post

surgery, i.e. larger changes in HADS depression subscale were associated with higher CRP values.

4.2. Increases in phenylalanine/tyrosine ratio are associated with acute physical disease and a diagnosis of depression

In the present study PHE/TYR was increased in patients following acute surgery and was associated with depressive symptoms. Differences in HADS values pre and post surgery even correlated with differences in PHE/TYR. The connection of PHE/TYR with depressive symptoms has been demonstrated previously: In a study examining inflammation and monoamine metabolism in aging, age was associated with increased immune markers and neuropsychiatric symptoms, and increased PHE/TYR correlated with neurovegetative symptoms (Capuron et al., 2011). Monoamine depletion studies (for serotonin and catecholamine pathway) have shown that changes in mood can be induced in a vulnerable population (people who have a familial history of major depressive disorders or are drug-free in remission after an episode of major depression) or following exposure to aversive psychological events PHE/TYR increased in acute physical disorders such sepsis as compared to local cerebral infections (Conejero et al., 1987) and in patients following general anaesthesia compared to local anaesthesia (Hol et al., 2009) were it was associated with increased pain and prolonged hospital stay.

4.3. Increases in kynurenine/tryptophan ratio and physical and mental disease

Changes in KYN/TRP have been shown to be associated with chronic physical low grade infections; e.g. HIV/herpesvirus infections where multiple herpesvirus co-infections were associated with increased KYN/TRP and poorer immune recovery following treatment (Yap et al., 2017). Changes in KYN pathway have also been shown for a multitude of other chronic physical diseases such as HIV-1 infection (Bipath et al., 2015), cancer (Hufner et al., 2015) or atherosclerosis (Baumgartner et al., 2017) and a bidirectional relationship between depression and obesity has been attributed to TRP metabolic pathway (Chaves Filho et al., 2018).

However, this is not an exclusive association as we and others found chronic physical conditions to be associated with changes in PHE/TYR (Hufner et al., 2015; Neurauter et al., 2008b). On the other hand also acute, not only chronic, changes in the serotonin pathway have been shown to induce depressive symptoms: acute TRP depletion has been found to produce transient relapse of depressive symptoms in some patients recovered from major depression, in particular those recovered on serotonergic agents, and those who have suffered from more than one major depressive episode (Bell et al., 2001; Booij et al., 2002).

TRP catabolites, like KYN and quinolinic acid, are depressiogenic and anxiogenic, activate oxidative pathways; and have neuroactive effects that may lead to neurodegeneration (Maes et al., 2011). In the present study we found no associations of KYN/TRP with depressive symptoms, neither with nor without physical disease. Results in the literature are heterogenous in that initially increased KYN/TRP ratios were proposed in depression due to IDO activation (Schroecksnadel et al., 2008; Swardfager et al., 2009) while other studies have described no association between KYN/TRP and depression (Bensimon et al., 2014; Quak et al., 2014). The inconsistent association of KYN/TRP with depression could be due to the fact that KYN is further degraded and more downstream metabolites such as kynurenic and quinolinic acid should better be measured. This is supported by the finding that inflammation might also activate enzymes downstream of IDO (Myint, 2012). However, the concentrations of KYN metabolites in the blood are probably only of little relevance, since these catabolites do not cross blood-brain barrier, and CSF specimens were not available. An alternative explanation for the missing association of KYN/TRP with depressive symptoms in the current study could be the fact the we analysed an unselected population of patients, of which only a small proportion (11.5% PRE and 17.9% POST) reached scores on the HADS which could be clinically relevant. These relatively low absolute numbers reflect the prevalence of depressive symptoms in the general population but might explain why no correlation of depressive symptoms with KYN/TRP was found (Leyton et al., 2000; Ruhe et al., 2007).

4.4. The role of gender and other variables in KYN/TRP and PHE/TYR analyses

The effect of increased PHE/TYR ratios for “mental health-depression” was mainly found in females, an effect that has been described before regarding an association of inflammation and depression (Morris et al., 2011). Compared to men, women may be more vulnerable to inflammation-induced mood and behavior changes since inflammation prompts greater feelings of loneliness and social disconnection for women than for men, which can contribute to the onset of depression (Derry et al., 2015). In an experimental laboratory study (randomized, placebo-controlled, double-blind trial) women appeared more vulnerable to the behavioral effects of inflammation compared to men: In response to an endotoxin challenge, women reported more depressive symptoms than men (Moieni et al., 2015). The direction of interaction of inflammation and depression (bidirectional vs unidirectional) may vary for men and women according to longitudinal data (Niles et al., 2018). Meta-analyes have shown inconsistent effects of inflammation, depressive symptoms and sex (Howren et al., 2009). Recently associations between the KYN/TRP pathway and cognitive deficits have been found in females with major depressive disorder but not in males (Zhou et al., 2018).

In the present study we did not find an effect of antidepressive or antipsychotic medication on PHE/TYR (nor KYN/TRP) probably due to the low absolute numbers of individuals receiving such treatment and the high variability in treatment regimen (table 1).

Lower KYN/TRP ratios were found in smokers compared to non-smokers in our study, an effect that has been observed previously and is thought to be related to the suppressive influence of smoking on the immune system and thus IDO activity (Pertovaara et al., 2006; Schennach et al., 2002).

4.5. Limitations

The current study has several limitations. Chronic and acute physical disease were different disease entities. The Charlson Comorbidity index assesses only certain disease, other comorbid diseases were not registered. The absolute number of individuals with depression was low, however, it was representative of an unselected clinical sample. Another limitation is the fact that we only analyzed peripheral values of the neurotransmitter precursor amino acids,

these have however been shown to correspond with the central situation (Felger et al., 2013; Schwarcz et al., 2012) but we cannot make any claims as to the specific association between central and peripheral values in the current study. We did not measure enzyme activity directly but indirectly infer it from the measured levels of neurotransmitter precursor monoamines. While there are an almost innumerable number of possible confounding factors we analyzed all that were available (some were unfortunately not recorded (e.g. menopause status) whilst acknowledging that this will always be a limitation. However, since our aim was to provide clinically relevant data by analyzing data from a real life clinical sample representing the everyday clinical sample making the analyzes relevant for clinical practice.

4.5. Conclusion

Immunological mechanisms related to inflammation induced by cell damage (surgery) or low grade inflammation in patients with chronic medical comorbidities (asthma or cardiovascular disease) could provide the basis for the connection between physical and mental disease. The influence of inflammation on neurotransmitter precursor amino acids could be the pathophysiological link.

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Tables

Table 1 Sample Characteristics

Variable	Number in % or Mean (SD)
Age in years	50.7 (17.8)
Female	40.9%
BMI in kg/cm	26.7 (4.5)
Smoking	29.5%
Alcohol consumption	
None	51.6%
Occasionally	31.2%
Regularly	7.5%
Antidepressive Medication (also for non-psychiatric reasons e.g. chronic pain) overall*	8.5%
SSRI (e.g. escitalopram)	4.5%
Trazodon	3.4%
Tricyclics (e.g. amitryptiline)	2.3%
SSNRI (e.g. venlafaxine)	1.1%
Buproprion	0.6%
Mirtazapine	0.6%
Antipsychotic medication overall	3.4%
Low potency FGA (e.g. prothipendyl)	1.7%
High potency SGA (e.g. quetiapine)	1.7%
Lifetime diagnosis of depression	7.3 %
Treatment with anti-inflammatory drugs	
None	22.6%
At discharge	33.9%
On admission and at discharge	43.5%
ISS score (only trauma cases)	3.18 (2.11)
Duration PRE-POST in days	3.51 (2.44)
Physical activity	None: 14.6% Light: 13.3% Active: 72.2%
Transfusion during surgery	1.7 %
Patients with chronic medical comorbidity	22.6 %
Charlson comorbidity index in points (only patients with comorbidity)	3.56 (1.74)

PRE: assessment before surgery, POST: assessment following surgery
SGA: second generation antipsychotic, FGA: first generation antipsychotic
*the percentages of the individual substance classes add up to more than 8.5% due to combination therapy in 7 patients
ISS score: injury severity score

Table 2 Analysis of mental and physical disease for CRP, neopterin and neurotransmitter precursor amino acids ratios

	Mental disease (Depression: N/Y)	Chronic physical disease (Medical comorbidity: N/Y)	Acute physical disease (Surgery: PRE/POST)
CRP (mg/dl)	N: 1.74 (1.27-2.21) Y: 1.24 (0.25-2.22)	N: 1.25 (0.670-1.82) Y: 1.74 (0.920-2.56)	PRE: 0.56 (-0.10-1.12) POST: 2.43 (1.76-3.10)

	P=0.259	P=0.024	P<0.001
NEOPTERIN (nmol/l)[§]	N: 6.77 (6.22 -7.32) Y: 7.35 (6.42-8.28) P=0.296	N: 5.94 (5.32-6.55) Y: 8.19 (7.22-9.15) P<0.001	PRE: 6.70 (6.06-7.34) POST: 7.42 (6.79-8.06) P< 0.001
PHE/TYR (µmol/µmol)[%]	N: 0.76 (0.72-0.81) Y: 0.88 (0.81-0.96) P= 0.012	N: 0.83 (0.79-0.88) Y: 0.81 (0.74-0.89) P= 0.517	PRE: 0.78 (0.73-0.83) POST: 0.87 (0.82-0.92) P<0.001
KYN/TRP[$] (µmol/mmol)	N: 34.12 (31.89-36.33) Y: 34.38 (30.42-38.34) P=0.828	N: 31.47 (28.92-34.01) Y: 37.02 (33.32-40.72) P=0.005	PRE: 33.73 (31.05-36.40) POST: 34.76 (32.01-37.42) P=0.156

[§]the interaction of acute physical disease*chronic physical disease was significant (p= 0.039), patients with chronic physical disease showed a higher rise in neopterin in the case of acute physical disease than those without

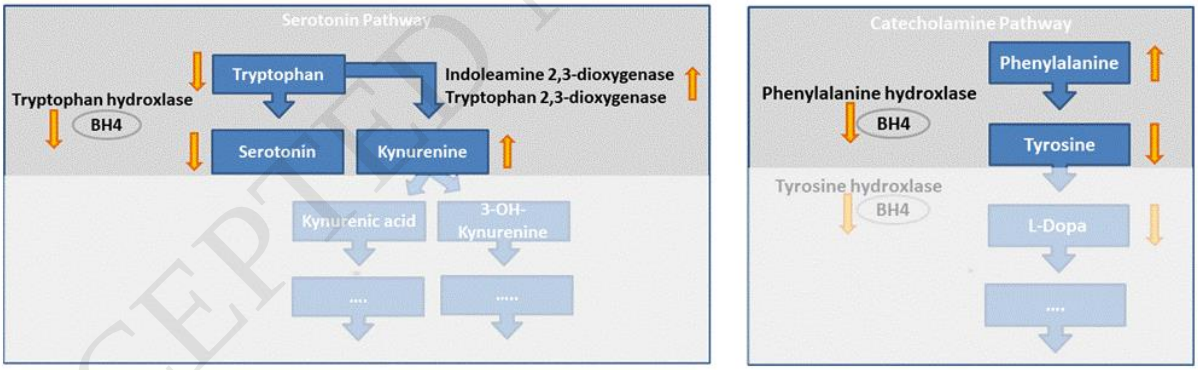
[%] corrected for gender (p= 0.003) and gender*depression (p=0.001) with depressed females showing higher values than females without depression, this was not evident in the males group

[$] corrected for smoking (p<0.001) with patients who smoked showing lower values.

Mean estimates and 95% confidence intervals are given. The model included the factors mental disease-depression, chronic physical disease and acute physical disease. Chronic physical disease was assessed using Charlson comorbidity index, acute physical disease was defined by the timepoints PRE and POST surgery. Depression was scored using HADS depression subscore. In case of a significant effect of co-factors we adjusted for these.

Figure 1

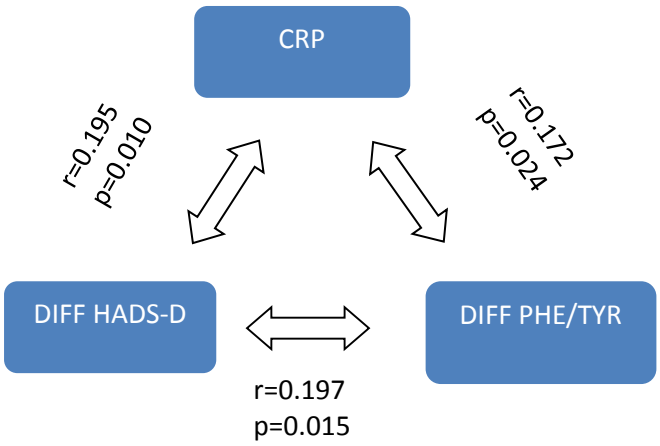
Neurotransmitter pathways analyzed in this study



Graphical depiction of the amino acids neurotransmitter pathways analyzed in the current study. Changes induced by inflammation are indicated with an arrow (modified from (Hufner et al., 2015). BH4: Tetrahydrobiopterin

Figure 2

Changes in PHE/TYR correlated with changes in depressive symptoms



Changes in HADS-D and phenylalanine/tyrosine pre and post surgery correlate with each other as well as with CRP concentrations. Absolute values were compared

Supplemental material

Table 1

	Mental disease (Depression: N/Y)	Chronic physical disease (Medical comorbidity: N/Y)	Acute physical disease (Surgery: PRE/POST)
KYN[?] (µmol/l)	N: 2.15 (2.02-2.27) Y: 2.13 (1.89-2.37) P=0.825	N: 1.98 (1.82-2.15) Y: 2.29 (2.06-2.52) P=0.029	PRE: 2.22 (2.06-2.37) POST: 2.06 (1.90-2.22) P=0.046
TRP[+] (µmol/l)	N: 60.45 (56.06-64.83) Y: 58.25 (52.06-64.45) P=0.452	N: 61.11 (55.95 66.28) Y: 57.59 (52.25-62.93) P = 0.068	PRE: 62.34 (57.45-67.24) POST: 58.36 (51.40-61.33) P<0.001
PHE[&] (µmol/l)	N: 102.80 (97.15-108.46) Y: 109.21 (98.48-119.94) P= 0.305	N: 101.15 (93.66-106.69) Y: 110.86 (101.00-120.72) P=0.148	PRE: 96.49 (89.62-103.37) POST: 115.52 (108.03-123.02) P<0.001
TYR[#] (µmol/l)	N: 134.96 (128.84-141.11) Y: 126.38 (114.24-138.53) P=0.180	N: 123.87 (116.53-131.21) Y: 137.49 (126.74-148.24) P=0.035	PRE: 127.12 (119.31-134.93) POST: 134.24 (126.32-142.16) P=0.041

[?]the interaction of mental health-depression* chronic physical disease was significant (p= 0.030), corrected for smoking (p<0.001)

[+]corrected for gender (p= 0.008) and SSRI treatment (p=0.013)

[&]corrected for anti-inflammatory medication (p=0.037)

[#]corrected for gender (p=0.001)

Table 2

	Mental disease (Depression: N/Y)	Chronic physical disease (Medical comorbidity: N/Y)	Acute physical disease (Surgery: PRE/POST)
FEMALE			
CRP (mg/dl)	N: 1.29 (0.82-1.76) Y: 1.55 (0.64-2.46) P=0.752	N: 1.39 (0.81-1.98) Y: 1.44 (0.70-2.18) P=0.483	PRE: 0.53 (-0.10-1.16) POST: 2.30 (1.68-2.93) P<0.001
NEOPTERIN[§] (nmol/l)	N: 7.37 (6.40 -8.35) Y: 7.46 (6.01-8.91) P=0.909	N: 5.89 (4.77-7.01) Y: 8.94 (7.38-10.50) P=0.002	PRE: 6.90 (5.84-7.96) POST: 7.93 (6.91-8.95) P=0.002
PHE/TYR (µmol/µmol)[%]	N: 0.78 (0.72-0.84) Y: 1.03 (0.93-1.13) P< 0.001	N: 0.89 (0.82-0.96) Y: 0.92 (0.82-1.01) P= 0.780	PRE: 0.85 (0.78-0.92) POST: 0.96 (0.89-1.03) P<0.001
KYN/TRP[$] (µmol/mmol)	N: 37.15 (34.01-40.29) Y: 39.13 (33.92-44.35) P=0.343	N: 34.97 (30.93-39.00) Y: 41.32 (36.36-46.27) P=0.034	PRE: 37.64 (34.11-41.16) POST: 38.65 (35.22-42.74) P=0.374
MALE			
CRP (mg/dl)	N: 2.13 (1.36-2.89) Y: 1.03 (0.70-2.76) P=0.135	N: 1.11 (0.17-2.05) Y: 2.05 (0.57-3.52) P=0.021	PRE: 0.60 (-0.53-1.72) POST: 2.56 (1.40-3.72) P<0.001
NEOPTERIN[§] (nmol/l)[§]	N: 6.34 (5.69 -6.99) Y: 7.25 (6.00-8.51) P=0.130	N: 5.96 (5.24-6.68) Y: 7.64 (6.37-8.90) P=0.011	PRE: 6.24 (5.42-7.06) POST: 7.35 (6.48-8.23) P< 0.001

PHE/TYR (µmol/µmol)	N: 0.76 (0.70-0.81) Y: 0.73 (0.61-0.84) P= 0.586	N: 0.76 (0.70-0.83) Y: 0.72 (0.61-0.83) P= 0.285	PRE: 0.70 (0.63-0.78) POST: 0.78 (0.71-0.86) P=0.005
KYN/TRP[$] (µmol/mmol)	N: 34.06 (31.18-36.95) Y: 33.55 (27.46-39.63) P=0.358	N: 29.79 (26.14-33.44) Y: 37.82 (31.35-44.30) P=0.175	PRE: 33.29 (29.49-37.08) POST: 34.33 (30.40-38.25) P=0.339

[§]the interaction of acute physical disease*chronic physical disease was significant in males (p=0.070 females, p=0.030 males)

[$]corrected for smoking males (p=0.001) females (p= 0.04)

Individual contributions

Design of study and data acquisition and analysis: D.F., M.B., B. S-U.

Data analysis and interpretation: K.H., D.F., B. S-U.

Writing of manuscript or revision for important content: K.H., D.F., M.B., B. S-U.

Final approval of the version to be submitted: K.H., D.F., M.B., B. S-U.

Conflicts of interest

The authors report no conflicts of interest.

All authors have approved the final version of the article.

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