Supplementary Online Content

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This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods

Participants- Primary BRAINnet Sample

Participants who contributed data to this analyzed dataset were recruited from 5 medical research or clinical research sites. These sites agreed to collaborate to evaluate brain health in patients using a standardized set of assessments and contribute the data to a centralized library (the basis of the BRAINnet Foundation Database). The medical research sites were located in universities with teaching hospital outpatient clinics focused on the disorders of interest, and based in the same geographical communities as the controls (1). The recruitment of patients with MDD and PTSD was from the general community of the population center of Sydney, which is a diverse and representative area. The patients attended the Brain Dynamics Center for assessments (MDD) or the PTSD Unit for assessments (PTSD), both of which were physically based at Westmead Hospital, a teaching hospital for the Sydney Medical School. Thus, the patients were not recruited from academic center clinics but attended the academic center for their testing sessions. Panic Disorder participants were recruited from outpatient and community sources in the population center of Adelaide and tested at the Flinders University academic center, participating in the BRAINnet database. Healthy control subjects were recruited from the same geographical regions and socioeconomic backgrounds as the clinical subjects.

Inclusion criteria for all participants in regard to the capacity to undergo a computerized test were: reading at Year 5 level (equivalent to Year 6 in England and 5th grade in the United States), normal (or corrected to normal) vision, and ability to use a keyboard. Participants were additionally required to refrain from smoking and caffeinated beverages for at least 2 hours prior testing, and to refrain from alcohol for at least 12 hours prior to testing.

Exclusion criteria for healthy control participants included a personal or first degree family history of DSM-IV Axis I disorders, or a personal history of brain injury, neurological disorder or other serious medical condition, sleep or learning disorders, or drug or alcohol addiction (using the Alcohol Use Disorders Identification Test - AUDIT) of the World Health Organization (2) and the Fagerstrom Tobacco Dependency Questionnaire (3). Healthy controls were also screened for symptoms of mood, anxiety and trauma disorders using the Somatic and Psychological Health Report (SPHERE; (4)). The SPHERE is a 34-item scale that rates both the psychological and somatic symptoms of these disorders.

MDD participants were either medication naïve (70%) or washed out for at least five half-lives of the medication at the time of testing. Two PTSD patients were on SSRIs during the testing period, and 50% of PTSD patients had comorbid MDD. PTSD participants also had no history of brain injury, loss of consciousness, stroke, neurological disorder, or other serious medical conditions (e.g., CVD and diabetes). Patients were ruled out for current substance use disorder, psychosis, and personality disorders. Average time post trauma was 65 months (SD = 64 months). PTSD was related to trauma due to assault in 50% of patients and due to being in car accidents involving a fatality in the remaining 50%. Panic Disorder exclusion criteria included a personal history of neurological disorder, physical brain injury, or serious medical problems and substance use disorder (same as for the other groups). No patient had taken benzodiazepine medication within the 2 weeks prior to assessment. 32 patients had used no psychotropic medication for at least 6 months prior to testing. Of the remaining, 13 were taking SSRIs at the time of testing, and 7 were taking SNRI, tri-cyclic or MAO antidepressant medication. Based on patient diaries including self-reported DSM-IV defined symptoms over the two-week period following testing, Panic Disorder patients reported a mean Panic Disorder Severity Scale (PDSS) score of 12.12 and a total number of Panic Attacks (during this two week period) of 4.12.

Comorbidities were present in the sample (see eTable 1).

Participants- Independent Validation "RAD" Sample

Data for the independent validation sample was an expanded sample that incorporated participants from the Research domain criteria Anxiety and Depression ("RAD") project, an observational study focusing trans-diagnostically on the spectrum of depression and anxiety psychopathology. Consistent with the goal of RDoC, screening, and exclusion criteria were kept to a minimum. Inclusion criteria included: i) age (18+ years) to focus on the adult brain, ii) fluent and literate in English in order to understand task instructions, and iii) currently reporting mood and anxiety symptoms. Exclusion criteria included: i) current or lifetime experience of frank psychosis and/or mania, because the circuit dysfunctions associated with such phenomenology might obscure interpretation of anxiety and mood-related circuit dysfunctions, ii) presence of suicidal intent representing imminent risk as indicated during screening and on-site assessments, iii) medical condition or neurological disorder that could impact brain imaging

data and render images difficult to interpret, iv) history of physical brain injury or blow to the head resulting in loss of consciousness greater than five minutes and which in the judgment of investigators could interfere with interpretation of brain imaging assessments, and v) severe impediment to vision, hearing and/or hand movement, likely to interfere with the ability to complete the assessments, or follow the instructions.

90% of participants were free of antidepressant medications and other medications that could impact assessments. Participants were enrolled in the study from 2013 to 2017. Patients were enrolled from the Gronowski Center, a community mental health training clinic, and individuals from the immediate surrounding community. Comorbid mood and anxiety disorders were present (see eTable 2). Lifetime disorders were also assessed for Major Depressive Disorder (40.9%), Panic Disorder (21.5%), Bipolar II Disorder (4.7%). Bipolar I Disorder (7.3%), and Bipolar NOS (6.3%).

Behavioral Measure of Cognition: Integneuro

For data reduction, we first took into account those variations in measures attributable to age and sex. A ‘‘peer regression modeling’’ technique was used, based on well-established psychometric principles. Age was modeled using both linear and logarithmic terms, and sex was modeled using a linear term. The expected score for each measure on each task was subtracted from the participant’s actual score, and the resulting difference was divided by the standard error of the estimate of the regression equation.

For details on cognitive measures, see eTable 3.

Neurophysiological Measure of Brain Activation: LabNeuro

Participants were assessed in a controlled environment, seated in a comfortable chair in a dimly lit room. Data were recorded continuously with a sampling rate of 500Hz, with a virtual ground and an average reference. Horizontal eye movements were recorded with electrodes 1.5cm lateral to the outer canthus of each eye and vertical eye movements, with electrodes placed 3mm above the middle of the left eyebrow and 1.5cm below the middle of the left bottom eye-lid. Skin resistance was < 5 KOhms. Data were EOG corrected offline based on the established Gratton algorithm (5). For quantification of power spectra, we employed Fast Fourier Transformation (FFT).

Facial Emotion Paradigms

Emotion images were modified such that the eyes were presented in the central position of the image. 50% of the faces were female. The threshold for subliminal presentations, defined by a lack of sensory awareness, was established in an initial signal detection study (6).

For details on EEG measures, see eTable 3.

Self-Reported Functional Status: BRISC

Correlation analyses between the BRISC scales and the World Health Organization Quality of Life- BREF (WHOQOL-BREF; (7)), Satisfaction with Life Scale (SWLS; (8)), and Health Productivity Questionnaire (HPQ; (9)), at a corrected P-value of 0.01, demonstrated 1) positive correlations between higher emotional resilience and higher scores on the WHOQOL-BREF psychological component ($r = 0.52$, $P < 0.001$) and satisfaction with life on the SWLS ($r = 0.34$, $P = 0.01$), and 2) positive correlations between higher social skills and higher scores on the WHOQOL-BREF components of physical health ($r = 0.45$, $P = 0.001$) and environment ($r = 0.56$, $P < 0.001$), satisfaction with life ($r = 0.42$, $P = 0.001$) and presenteeism on the HPQ ($r = 0.37$, $P = 0.008$) (1).

For details on functioning measures, see eTable 3.

Data Analysis

Bonferroni Corrected Alphas

Multiple comparisons were addressed by using the Bonferroni Correction. For neurocognitive performance, ANOVAs were run on each of the nine tests, with a Bonferroni corrected alpha level of p=0.006. For neurophysiology measures, ANOVAs were run separately for EEG eyes open and closed and for conscious and nonconscious emotion conditions. In these ANOVAs, dependent variables were the four averaged regional power values for both alpha and beta bands; thus, the corrected alpha level was p=0.006. An ANOVA was run on the single measure of alpha asymmetry at p=.05. For self-reported daily function, ANOVAs were run on the two functioning domains of social skills and emotional resilience at the corrected alpha level of p= 0.03.

eTable 1. Comorbidities in Primary Sample

		Comorbid Diagnoses							
		MDD	PTSD	Panic	GAD	ADHD	Dys.	OCD	SAD
Primary Diagnosis	MDD	100 (100%)	12 (12.0%)	14 (14.0%)	36 (36%)	1 (1.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	PTSD	17 (36.2%)	47 (100%)	0 (0.0%)	4 (8.5%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	Panic	7 (13.2%)	15 (28.3%)	53 (100%)	5 (9.4%)	0 (0.0%)	3 (5.7%)	13 (24.5%)	31 (58.5%)

Notes: MDD: Major Depressive Disorder; PTSD: Post-traumatic Stress Disorder; Panic: Panic Disorder; ADHD: Attention Deficit/Hyperactivity Disorder; Dys.: Dysthymia; OCD: Obsessive-Compulsive Disorder; SAD: Social Anxiety Disorder. Percentages should be ready by row (i.e. of those with a primary MDD diagnosis, 12% had comorbid PTSD).

eTable 2. Comorbidities in Validation Sample

		Diagnosis 2						
		MDD	PTSD	Panic	GAD	Bipolar II	OCD	SAD
Diagnosis 1	MDD	77 (100%)	25 (32.5%)	12 (15.6%)	36 (46.8%)	6 (7.8%)	20 (26.0%)	24 (31.2%)
	PTSD	25 (53.2%)	47 (100%)	15 (31.9%)	26 (55.3%)	3 (6.4%)	17 (36.2%)	18 (38.3%)
	Panic	12 (33.3%)	15 (41.7%)	36 (100%)	17 (47.2%)	1 (2.8%)	12 (33.3%)	14 (38.9%)
	GAD	36 (30.8%)	26 (22.2%)	17 (14.5%)	117 (100%)	2 (1.7%)	23 (19.7%)	32 (27.4%)
	Bipolar II	6 (46.2%)	3 (23.1%)	1 (7.7%)	2 (15.4%)	13 (100%)	2 (15.4%)	0 (0.0%)
	OCD	20 (54.1%)	17 (45.9)	12 (32.4%)	23 (62.2%)	2 (5.4%)	37 (100%)	16 (43.2%)
	SAD	24 (43.6%)	18 (32.7%)	14 (25.5%)	32 (58.2)	0 (0.0%)	16 (29.1%)	55 (100%)

Notes: MDD: Major Depressive Disorder; PTSD: Post-traumatic Stress Disorder; Panic: Panic Disorder; GAD: Generalized Anxiety Disorder; OCD: Obsessive-Compulsive Disorder; SAD: Social Anxiety Disorder. In validation sample, participants were not coded by primary diagnosis. Table shows frequencies of participants with pairs of diagnoses. Percentages should be ready by row (i.e. of those with MDD, 32.5% had comorbid PTSD.

eTable 3. Study measures.

Table 3a: Primary Stratification

Unit of analysis	Construct	Measures	Paradigm
Self-Report	Potential threat ("anxious arousal")	Anxiety Scale questions	DASS-21 Questionnaire
	Sustained threat ("tension")	Stress Scale questions	DASS-21 Questionnaire
	Loss ("anhedonia")	Depression Scale questions	DASS-21 Questionnaire

Notes: Measures used for primary stratification of participants.

eTable 3b: Expression of Subtypes in Behavioral Domain

Unit of analysis	Construct	Measures	Paradigm
Behavior	Attention	RT	Choice Reaction Time
	Cognitive Control	Accuracy (switching errors), completion time, connection time	Switching of Attention
		Accuracy (errors), RT	Verbal Interference/Stroop
		Accuracy (total, false-positive, and false-negatives), RT, RT variability	Go-NoGo
		Accuracy (total, overrun errors), completion time	Maze
	Working Memory	Accuracy (total recall, and maximum recall span)	Digit Span
		Accuracy (total recall, and maximum recall span)	Span of Visual Memory
	Language fluency	Accuracy (number of words)	Word Generation for words starting with F,A,S
	Response Speed	Number and variability of taps	Motor Tapping

Notes: Measures used to evaluate the extent to which subtypes were expressed in the behavioral measure of neurocognition. To aid interpretation, scores on each of the paradigms were grouped conceptually according to RDoC-relevant Cognitive System domains of Attention (Choice Reaction Time), Cognitive Control (Switching of Attention, Verbal Interference/Stroop, Go-NoGo, Maze), Working Memory (Digit Span, Span of Visual Memory), Language fluency (Word Generation for words starting with F, A, S), and Response Speed (Motor Tapping).

eTable 3c: Expression of Subtypes in Physiological Domain

Unit of analysis	Construct	Measure	Paradigm
Physiology	Acute threat ("fear")	EEG Beta and Alpha power	Viewing of facial expressions of fear and anger under conscious and nonconscious conditions
	Loss ("sad")	EEG Beta and Alpha power	Viewing of facial expressions of sadness under conscious and nonconscious conditions
	Resting State	EEG Beta and Alpha power Alpha asymmetry	Resting eyes closed
		EEG Beta and Alpha power Alpha asymmetry	Resting eyes open

Notes: Measures used to evaluate the extent to which subtypes were expressed in the electrocortical measure of brain activation. EEG measures relevant to mood, anxiety and stress disorders: acute threat ("fear"), loss ("sad"), and resting state were quantified.

eTable 3d: Expression of Subtypes in Clinical Domain

Unit of analysis	Construct	Measure	Paradigm
Self-report	Social Processes	Resilience and social functions related to emotional intelligence	Brief Risk-resilience Index for Screening

Notes: evaluated the extent to which subtypes were expressed in daily functional capacity.

eTable 4. Component Loadings for three negative mood components.

	Anhedonia	Anxious Arousal	Tension
I found that I had nothing to look forward to *(DASS 10)*	*0.851*		
I felt that life was meaningless *(DASS 21)*	*0.821*		
I couldn't seem to experience any positive feeling at all *(DASS 3)*	*0.803*		
I felt downhearted and blue *(DASS 13)*	*0.799*		
I was unable to become enthusiastic about anything *(DASS 16)*	*0.796*		
I felt that I wasn't worth much as a person *(DASS 17)*	*0.792*		
I found it difficult to work up the initiative to do things *(DASS 5)*	*0.687*		*0.437*
I felt scared without any good reason *(DASS 20)*		*0.754*	
I was aware of action of heart in absence of physical exertion *(DASS 19)*		*0.746*	
I was worried about situations might panic make fool of self *(DASS 9)*		*0.739*	
I felt I was close to panic *(DASS 15)*		*0.725*	
I experienced breathing difficulty *(DASS 4)*		*0.674*	
I felt that I was using a lot of nervous energy *(DASS 8)*		*0.617*	*0.476*
I experienced trembling e.g. in the hands *(DASS 7)*		*0.615*	
I found myself getting agitated *(DASS 11)*	*0.418*		*0.693*
I was intolerant of anything kept me from what I was doing *(DASS 14)*			*0.677*
I felt that I was rather touchy *(DASS 18)*	*0.424*		*0.654*
I found it hard to wind down *(DASS 1)*			*0.648*
I found it difficult to relax *(DASS 12)*	*0.419*	*0.426*	*0.632*
I tended to overreact to situations *(DASS 6)*	*0.435*	*0.452*	*0.571*
I was aware of dryness of my mouth *(DASS 2)*		*0.435*	*0.455*

eTable 5. Component Loadings for three negative mood components in an independent validation sample.

	Anhedonia	Tension	Anxious Arousal
I found that I had nothing to look forward to *(DASS 10)*	*0.819*		
I was unable to become enthusiastic about anything *(DASS 16)*	*0.817*		
I felt that life was meaningless *(DASS 21)*	*0.787*		
I felt that I wasn't worth much as a person *(DASS 17)*	*0.771*		
I felt downhearted and blue *(DASS 13)*	*0.762*		
I couldn't seem to experience any positive feeling at all *(DASS 3)*	*0.753*		
I found it difficult to work up the initiative to do things *(DASS 5)*	*0.722*		
I found myself getting agitated *(DASS 11)*		*0.759*	
I felt that I was rather touchy *(DASS 18)*		*0.753*	
I tended to overreact to situations *(DASS 6)*		*0.709*	
I was intolerant of anything kept me from what I was doing *(DASS 14)*		*0.662*	
I found it hard to wind down *(DASS 1)*		*0.616*	*0.387*
I felt that I was using a lot of nervous energy *(DASS 8)*		*0.607*	*0.496*
I found it difficult to relax *(DASS 12)*		*0.586*	*0.398*
I experienced breathing difficulty *(DASS 4)*			*0.756*
I was aware of action of heart in absence of physical exertion *(DASS 19)*			*0.703*
I experienced trembling e.g. in the hands *(DASS 7)*			*0.603*
I felt I was close to panic *(DASS 15)*		*0.425*	*0.562*
I was worried about situations might panic make fool of self *(DASS 9)*		*0.347*	*0.551*
I felt scared without any good reason *(DASS 20)*		*0.398*	*0.536*
I was aware of dryness of my mouth *(DASS 2)*			*0.484*

eTable 6. Complete table of number and percentage of individuals with each diagnosis within subtypes.

	Normative Mood	Tension	Anxious Arousal	General Anxiety	Anhedonia	Melancholia
Diagnosis						
Control	155 (70.5%)	57 (25.9%)	2 (0.9%)	3 (1.4%)	1 (0.5%)	2 (0.9%)
MDD	6 (6.0%)	12 (12.0%)	23 (23.0%)	9 (9.0%)	23 (23.0%)	27 (27.0%)
PTSD	5 (10.6%)	5 (10.6%)	12 (25.5%)	14 (29.8%)	3 (6.4%)	8 (17.0%)
Panic	14 (26.4%)	7 (13.2%)	18 (34.0%)	12 (22.6%)	2 (3.8%)	0 (0.0%)

eTable 7. Sex Distribution by Cluster

	Normative Mood	Tension	Anxious Arousal	General Anxiety	Anhedonia	Melancholia	Total
Male	74	33	22	10	9	16	164
Female	106	48	33	28	20	21	256
Total	180	81	55	38	29	37	420

eTable 8. Partial eta squared values.

	6-Cluster Solution	DSM Diagnosis
Cognitive Control	0.063	0.044
Working Memory	0.039	0.022
Emotion EEG Beta	0.047	0.011
Frontal EEG Beta	0.051	0.041
Social Function	0.205	0.143
Resilience	0.171	0.170

eTable 9. R-squared for ANCOVAs including comorbidity covariates.

	6-Cluster Solution	DSM Diagnosis
Cognitive Control	0.108	0.070
Working Memory	0.053	0.033
Emotion EEG Beta	0.057	0.028
Frontal EEG Beta	0.101	0.067
Social Function	0.239	0.184
Resilience	0.184	0.213

eTable 10. Tukey HSD post-hoc tests to evaluate significant between-cluster differences. The first column indicates the measure, and the second and third columns indicate which two clusters are included in the pairwise test (e.g. for the neurocognitive Go-NoGo measure, there was a significant pairwise difference between the Low Symptom cluster and Anxious Arousal cluster.)

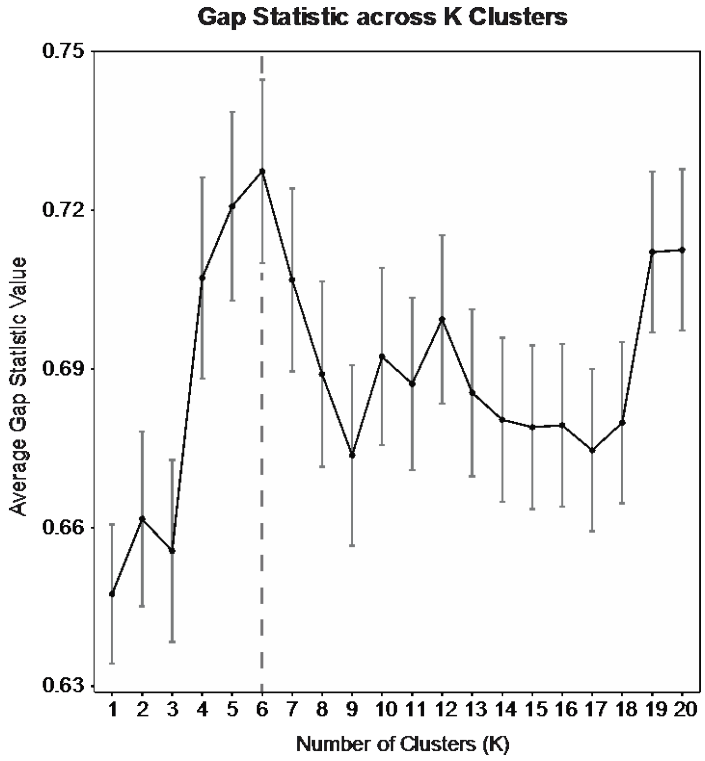
			Mean Difference	Std. Error	Sig.
Go-NoGo	Low Symptom	Tension	-0.03	0.08	1.00
		Anxious Arousal	**0.05**	**0.10**	**<0.001**
		General Anxiety	0.05	0.12	1.00
		Anhedonia	-0.09	0.13	0.98
		Melancholia	0.12	0.12	0.89
	Tension	Low Symptom	0.03	0.08	1.00
		Anxious Arousal	**0.48**	**0.11**	**<0.001**
		General Anxiety	0.08	0.13	0.99
		Anhedonia	-0.07	0.14	1.00
		Melancholia	0.15	0.13	0.84
	Anxious Arousal	**Low Symptom**	**-0.45**	**0.10**	**<0.001**
		Tension	**-0.48**	**0.11**	**<0.001**
		General Anxiety	**-0.40**	**0.14**	**0.05**
		Anhedonia	**-0.55**	**0.15**	**0.003**
		Melancholia	-0.33	0.14	0.16
	General Anxiety	Low Symptom	-0.05	0.12	1.00
		Tension	-0.08	0.13	0.99
		Anxious Arousal	**0.40**	**0.14**	**0.05**
		Anhedonia	-0.15	0.16	0.94
		Melancholia	0.07	0.15	1.00
	Anhedonia	Low Symptom	0.09	0.13	0.98
		Tension	0.07	0.14	1.00
		Anxious Arousal	**0.55**	**0.15**	**0.003**
		General Anxiety	0.15	0.16	0.94
		Melancholia	0.22	0.16	0.73
	Melancholia	Low Symptom	-0.12	0.12	0.89
		Tension	-0.15	0.13	0.84
		Anxious Arousal	0.33	0.14	0.16
		General Anxiety	-0.07	0.15	1.00
		Anhedonia	-0.22	0.16	0.73
Digit Span	Low Symptom	Tension	0.18	0.11	0.58
		Anxious Arousal	**0.42**	**0.13**	**0.02**
		General Anxiety	0.27	0.15	0.47
		Anhedonia	-0.10	0.16	0.99
		Melancholia	0.32	0.15	0.25
	Tension	Low Symptom	-0.18	0.11	0.58
		Anxious Arousal	0.24	0.15	0.57
		General Anxiety	0.09	0.17	0.99
		Anhedonia	-0.27	0.18	0.63
		Melancholia	0.14	0.16	0.95
	Anxious Arousal	**Low Symptom**	**-0.42**	**0.13**	**0.02**
		Tension	-0.24	0.15	0.57
		General Anxiety	-0.15	0.18	0.96
		Anhedonia	-0.51	0.19	0.08
		Melancholia	-0.10	0.18	0.99
	General		Mean	Std.	Sig.

	Anxiety		Difference	Error	
		Tension	-0.09	0.17	0.99
		Anxious Arousal	0.15	0.18	0.96
		Anhedonia	-0.37	0.20	0.47
		Melancholia	0.05	0.19	1.00
	Anhedonia	Low Symptom	0.10	0.16	0.99
		Tension	0.27	0.18	0.63
		Anxious Arousal	0.51	0.19	0.08
		General Anxiety	0.37	0.20	0.47
		Melancholia	0.42	0.20	0.31
	Melancholia	Low Symptom	-0.32	0.15	0.25
		Tension	-0.14	0.16	0.95
		Anxious Arousal	0.10	0.18	0.99
		General Anxiety	-0.05	0.19	1.00
		Anhedonia	-0.42	0.20	0.31
Social Function	Low Symptom	Tension	0.05	0.71	1.00
		Anxious Arousal	**5.37**	**0.82**	**<0.001**
		General Anxiety	0.55	0.95	0.99
		Anhedonia	**4.84**	**1.06**	**<0.001**
		Melancholia	**7.39**	**0.96**	**<0.001**
	Tension	Low Symptom	-0.05	0.71	1.00
		Anxious Arousal	**5.32**	**0.93**	**<0.001**
		General Anxiety	0.50	1.05	1.00
		Anhedonia	**4.80**	**1.15**	**0.001**
		Melancholia	**7.35**	**1.06**	**<0.001**
	Anxious Arousal	**Low Symptom**	**-5.37**	**0.82**	**<0.001**
		Tension	**-5.32**	**0.93**	**<0.001**
		General Anxiety	**-4.82**	**1.12**	**<0.001**
		Anhedonia	-0.52	1.22	1.00
		Melancholia	2.03	1.13	0.47
	General Anxiety	Low Symptom	-0.55	0.95	0.99
		Tension	-0.50	1.05	1.00
		Anxious Arousal	**4.82**	**1.12**	**<0.001**
		Anhedonia	**4.30**	**1.31**	**0.01**
		Melancholia	**6.85**	**1.23**	**<0.001**
	Anhedonia	**Low Symptom**	**-4.84**	**1.06**	**<0.001**
		Tension	**-4.80**	**1.15**	**0.001**
		Anxious Arousal	0.52	1.22	1.00
		General Anxiety	**-4.30**	**1.31**	**0.01**
		Melancholia	2.55	1.32	0.38
	Melancholia	**Low Symptom**	**-7.39**	**0.96**	**<0.001**
		Tension	**-7.35**	**1.06**	**<0.001**
		Anxious Arousal	-2.03	1.13	0.47
		General Anxiety	**-6.85**	**1.23**	**<0.001**
		Anhedonia	-2.55	1.32	0.38
Resilience	Low Symptom	Tension	1.23	0.96	0.79
		Anxious Arousal	**7.82**	**1.10**	**<0.001**
		General Anxiety	2.44	1.25	0.37
		Anhedonia	**5.25**	**1.41**	**0.003**
		Melancholia	**7.27**	**1.29**	**<0.001**
	Tension	Low Symptom	-1.23	0.96	0.79
		Anxious Arousal	**6.59**	**1.25**	**<0.001**
		General Anxiety	1.21	1.38	0.95

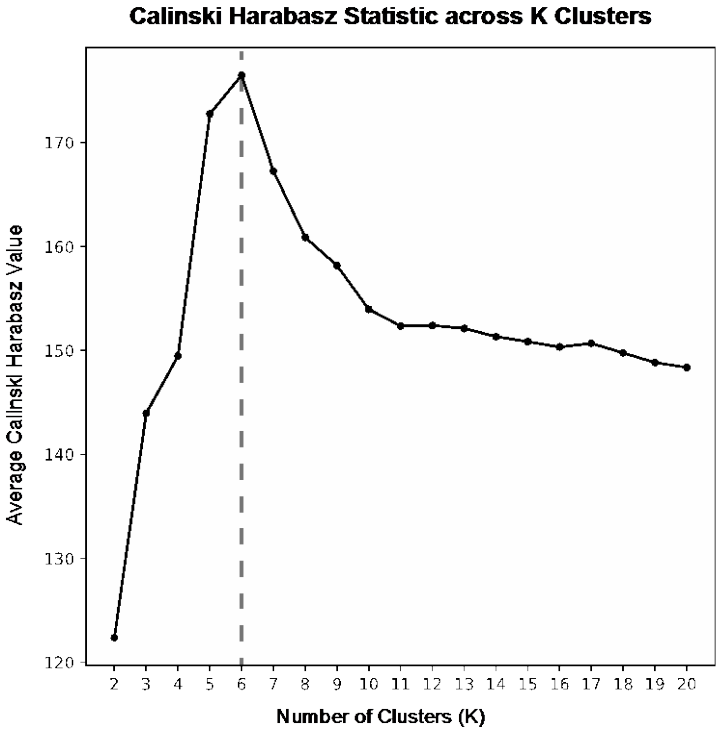
			Mean Difference	Std. Error	Sig.
		Anhedonia	4.01	1.52	0.09
		Melancholia	**6.03**	**1.42**	**<0.001**
	Anxious Arousal	**Low Symptom**	**-7.82**	**1.10**	**<0.001**
		Tension	**-6.59**	**1.25**	**<0.001**
		General Anxiety	**-5.38**	**1.48**	**0.004**
		Anhedonia	-2.57	1.62	0.60
		Melancholia	-0.55	1.52	1.00
	General Anxiety	Low Symptom	-2.44	1.25	0.37
		Tension	-1.21	1.38	0.95
		Anxious Arousal	**5.38**	**1.48**	**0.004**
		Anhedonia	2.81	1.72	0.58
		Melancholia	**4.83**	**1.63**	**0.04**
	Anhedonia	**Low Symptom**	**-5.25**	**1.41**	**0.003**
		Tension	-4.01	1.52	0.09
		Anxious Arousal	2.57	1.62	0.60
		General Anxiety	-2.81	1.72	0.58
		Melancholia	2.02	1.75	0.86
	Melancholia	**Low Symptom**	**-7.27**	**1.29**	**<0.001**
		Tension	**-6.03**	**1.42**	**<0.001**
		Anxious Arousal	0.55	1.52	1.00
		General Anxiety	**-4.83**	**1.63**	**0.04**
		Anhedonia	-2.02	1.75	0.86
Emotion EEG	Low Symptom	Tension	-1.11	0.79	0.73
		Anxious Arousal	-0.23	0.97	1.00
		General Anxiety	**-4.18**	**1.11**	**0.002**
		Anhedonia	-0.08	1.25	1.00
		Melancholia	-2.37	1.11	0.27
	Tension	Low Symptom	1.11	0.79	0.73
		Anxious Arousal	0.88	1.08	0.97
		General Anxiety	-3.08	1.20	0.11
		Anhedonia	1.02	1.34	0.97
		Melancholia	-1.26	1.20	0.90
	Anxious Arousal	Low Symptom	0.23	0.97	1.00
		Tension	-0.88	1.08	0.97
		General Anxiety	**-3.95**	**1.33**	**0.04**
		Anhedonia	0.15	1.45	1.00
		Melancholia	-2.14	1.33	0.59
	General Anxiety	**Low Symptom**	**4.18**	**1.11**	**0.002**
		Tension	3.08	1.20	0.11
		Anxious Arousal	**3.95**	**1.33**	**0.04**
		Anhedonia	4.10	1.54	0.09
		Melancholia	1.82	1.43	0.80
	Anhedonia	Low Symptom	0.08	1.25	1.00
		Tension	-1.02	1.34	0.97
		Anxious Arousal	-0.15	1.45	1.00
		General Anxiety	-4.10	1.54	0.09
		Melancholia	-2.28	1.54	0.68
	Melancholia	Low Symptom	2.37	1.11	0.27
		Tension	1.26	1.20	0.90
		Anxious Arousal	2.14	1.33	0.59
		General Anxiety	-1.82	1.43	0.80
		Anhedonia	2.28	1.54	0.68

			Mean Difference	Std. Error	Sig.
Resting EEG	Low Symptom	Tension	-1.21	1.01	0.84
		Anxious Arousal	-1.46	1.16	0.81
		General Anxiety	-1.22	1.37	0.95
		Anhedonia	**-6.26**	**1.48**	**<0.001**
		Melancholia	-2.30	1.35	0.53
	Tension	Low Symptom	1.21	1.01	0.84
		Anxious Arousal	-0.25	1.33	1.00
		General Anxiety	-0.01	1.51	1.00
		Anhedonia	**-5.05**	**1.61**	**0.02**
		Melancholia	-1.09	1.50	0.98
	Anxious Arousal	Low Symptom	1.46	1.16	0.81
		Tension	0.25	1.33	1.00
		General Anxiety	0.24	1.62	1.00
		Anhedonia	-4.80	1.71	0.06
		Melancholia	-0.84	1.60	1.00
	General Anxiety	Low Symptom	1.22	1.37	0.95
		Tension	0.01	1.51	1.00
		Anxious Arousal	-0.24	1.62	1.00
		Anhedonia	-5.04	1.86	0.08
		Melancholia	-1.08	1.76	0.99
	Anhedonia	**Low Symptom**	**6.26**	**1.48**	**<0.001**
		Tension	**5.05**	**1.61**	**0.02**
		Anxious Arousal	4.80	1.71	0.06
		General Anxiety	5.04	1.86	0.08
		Melancholia	3.96	1.84	0.27
	Melancholia	Low Symptom	2.30	1.35	0.53
		Tension	1.09	1.50	0.98
		Anxious Arousal	0.84	1.60	1.00
		General Anxiety	1.08	1.76	0.99
		Anhedonia	-3.96	1.84	0.27

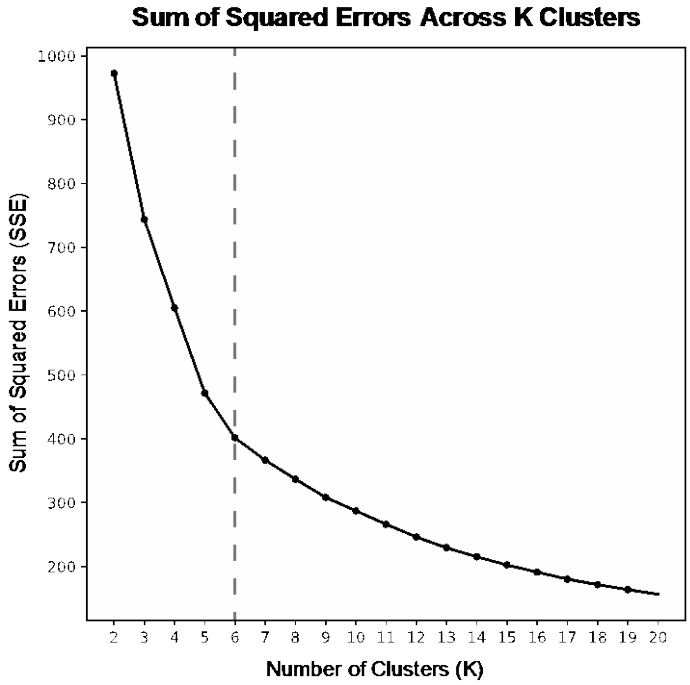
eFigure 1. The gap statistic, calculated for clusters (k) 1 to 20.



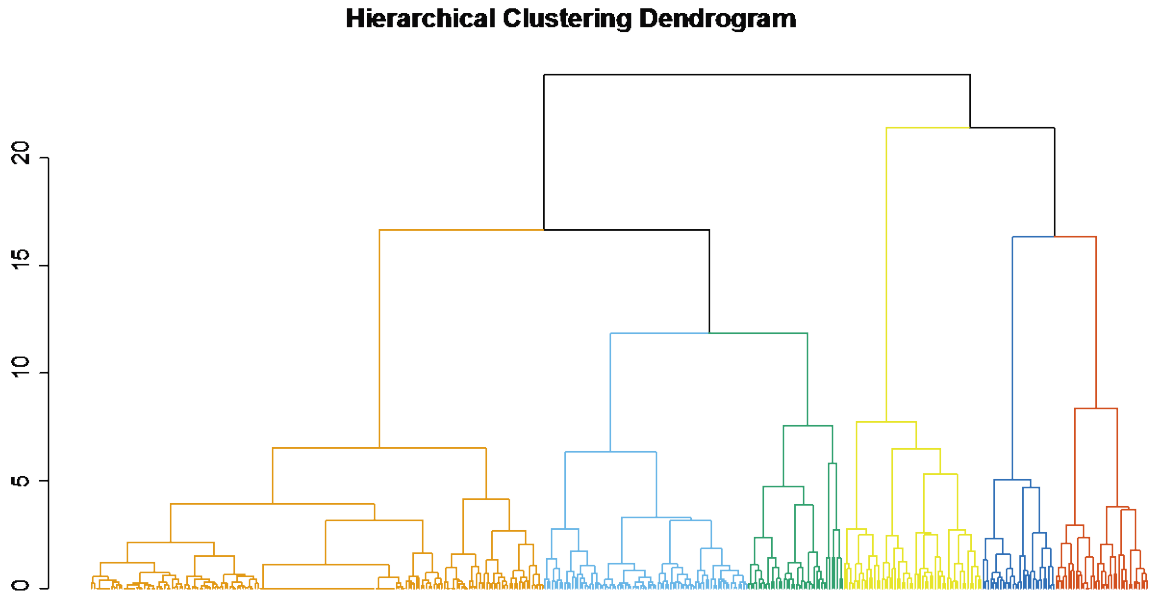
eFigure2. The Calinski-Harabasz index, calculated for clusters (k) 1 to 20. The maximum achieved index value indicates the best clustering of the data.



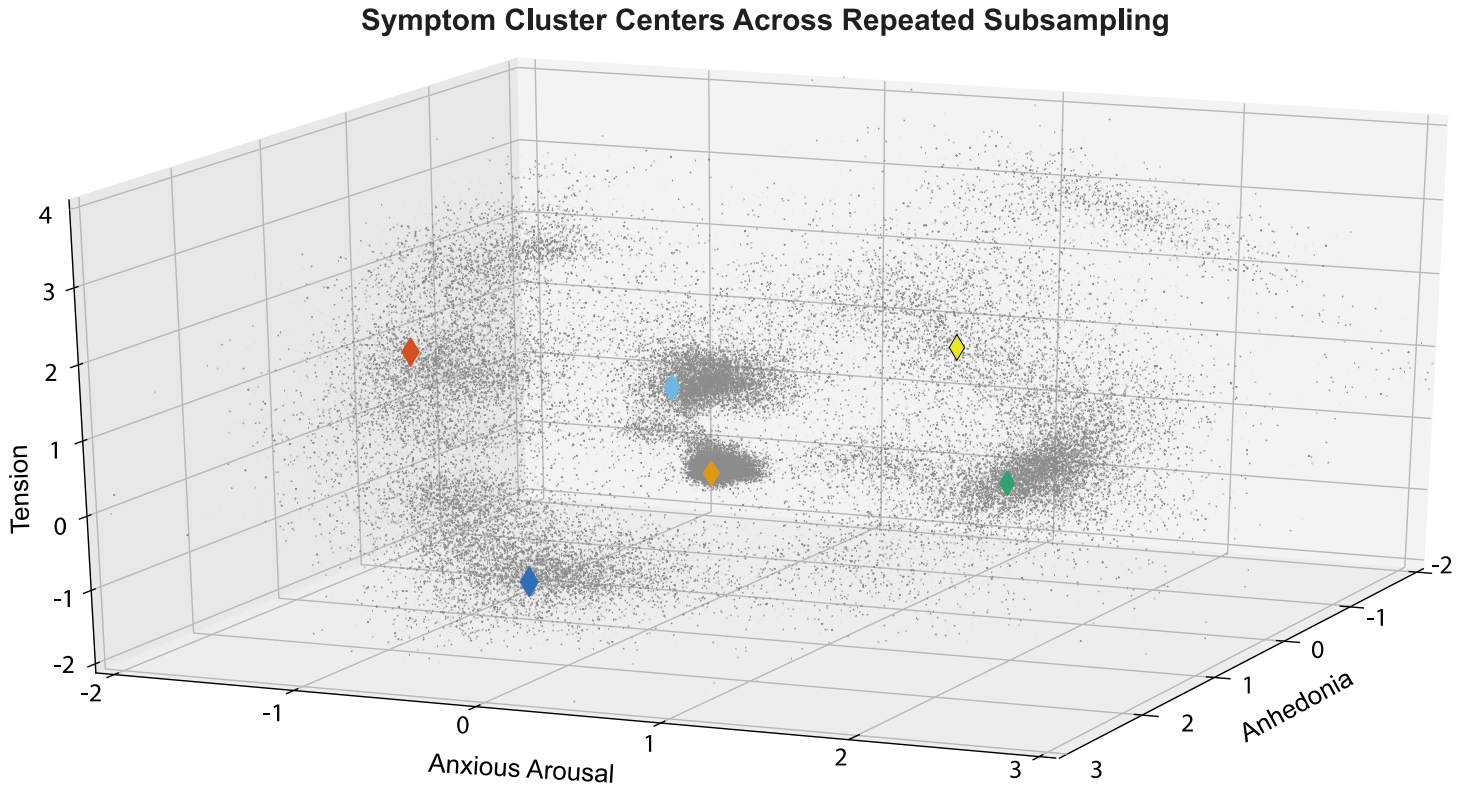
eFigure 3. The total within-cluster sum of squares for clusters (k) 1 to 20. This metric measures the compactness of the clustering solution. The location of a bend (knee) in the plot can be used as an indicator for the appropriate number of clusters.



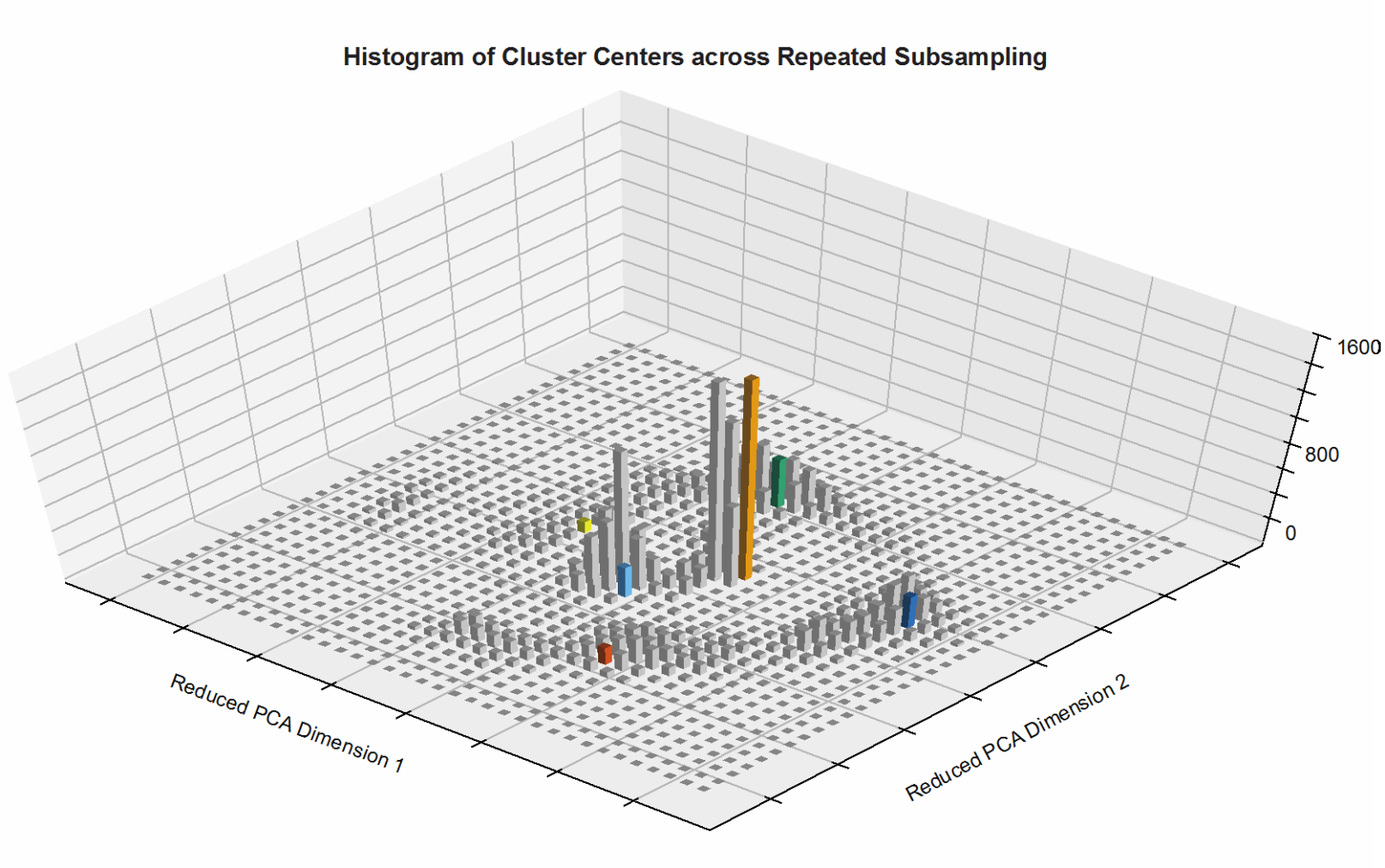
eFigure 4. The dendrogram for hierarchical clustering. The y-axis represents the distance between clusters. Colors represent the six-cluster solution chosen.



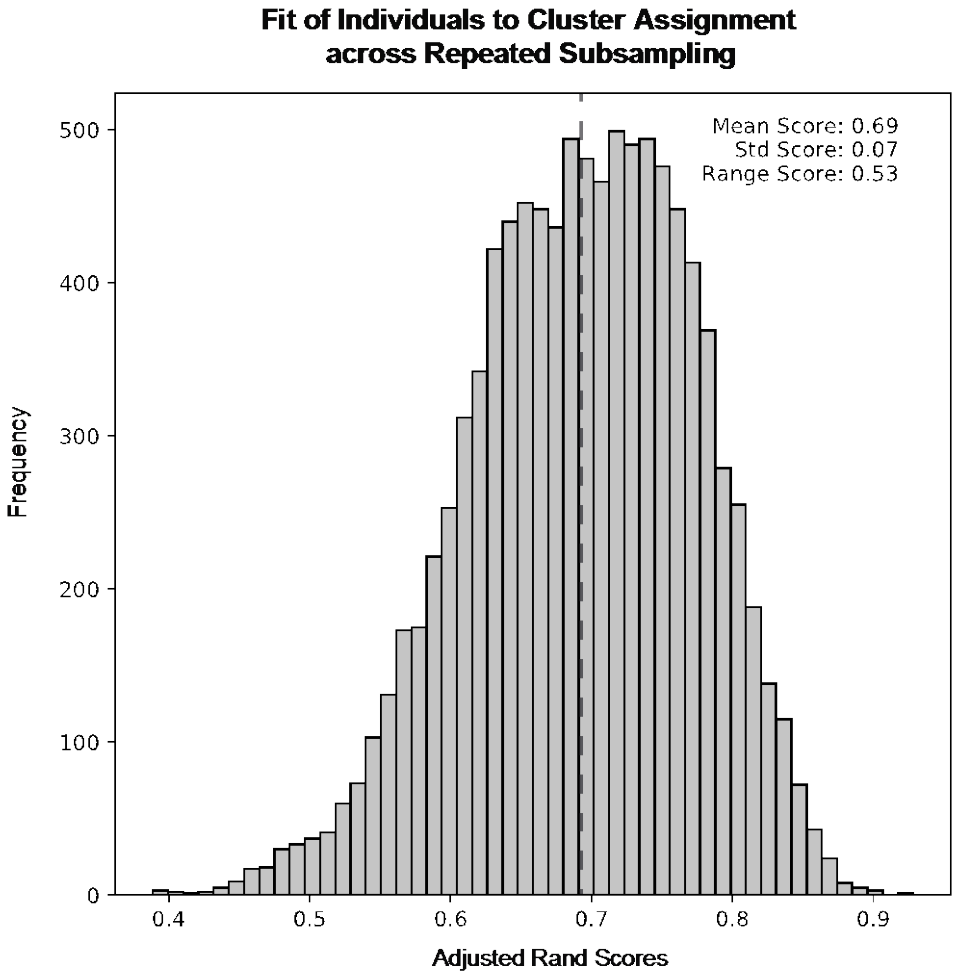
eFigure 5. Cluster centers plotted from 10,000 repeated subsamples. Labels on the x, y and z axis refer to the three symptom components. These labels are based on the assumption that the PCA performed across the 10,000 subsamples yielded the same principal components as the whole sample. Diamonds indicate the cluster centers of the original clustering solution. Colors correspond to each of the six clusters.



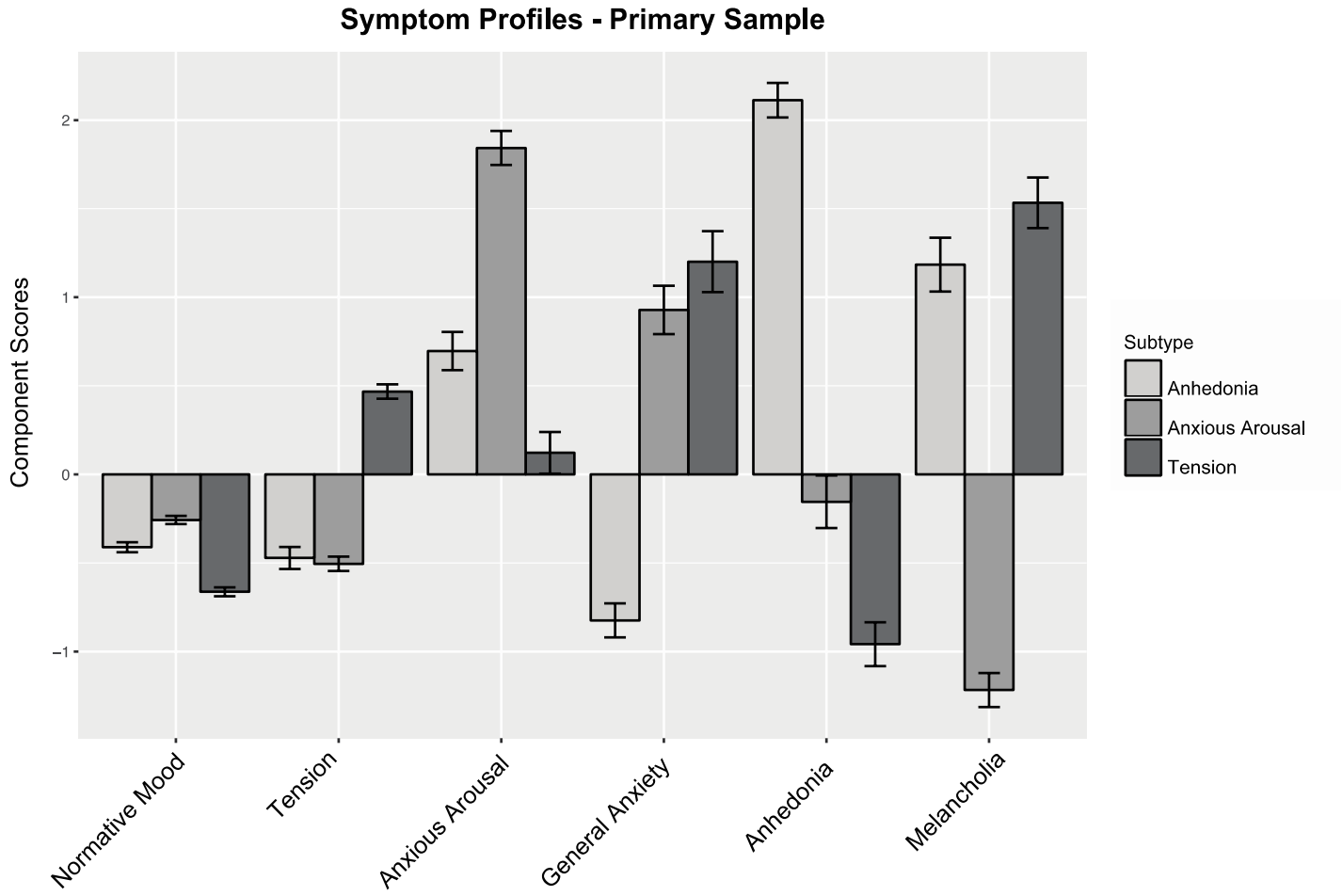
eFigure 6. Histogram of cluster centers calculated from repeated subsampling. For visualization purposes, we embedded our three-dimensional PCA space to two dimensions using nonmetric multidimensional scaling (MDS) in Python's scikit-learn package. The resulting two dimensions are on the x and y axis. Results for 6,000 of the 10,000 permutations are displayed, due to the computational constraints on visualization. The histogram bars that are colored contain the cluster centers of the original clustering solution. Colors correspond to each of the six clusters.

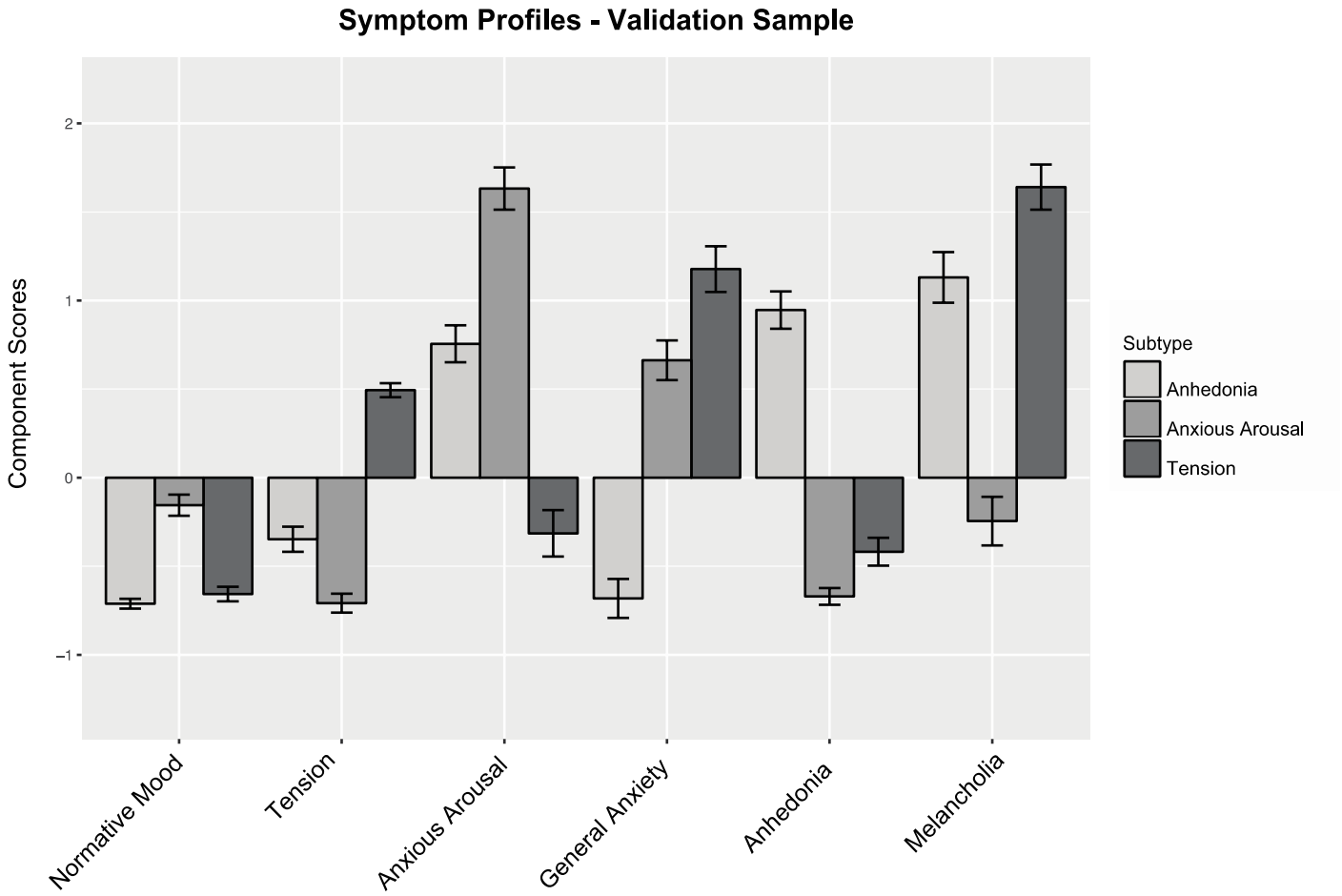


eFigure 7. The distribution of adjusted Rand scores visualized in a histogram. The mean Rand score was 0.69.

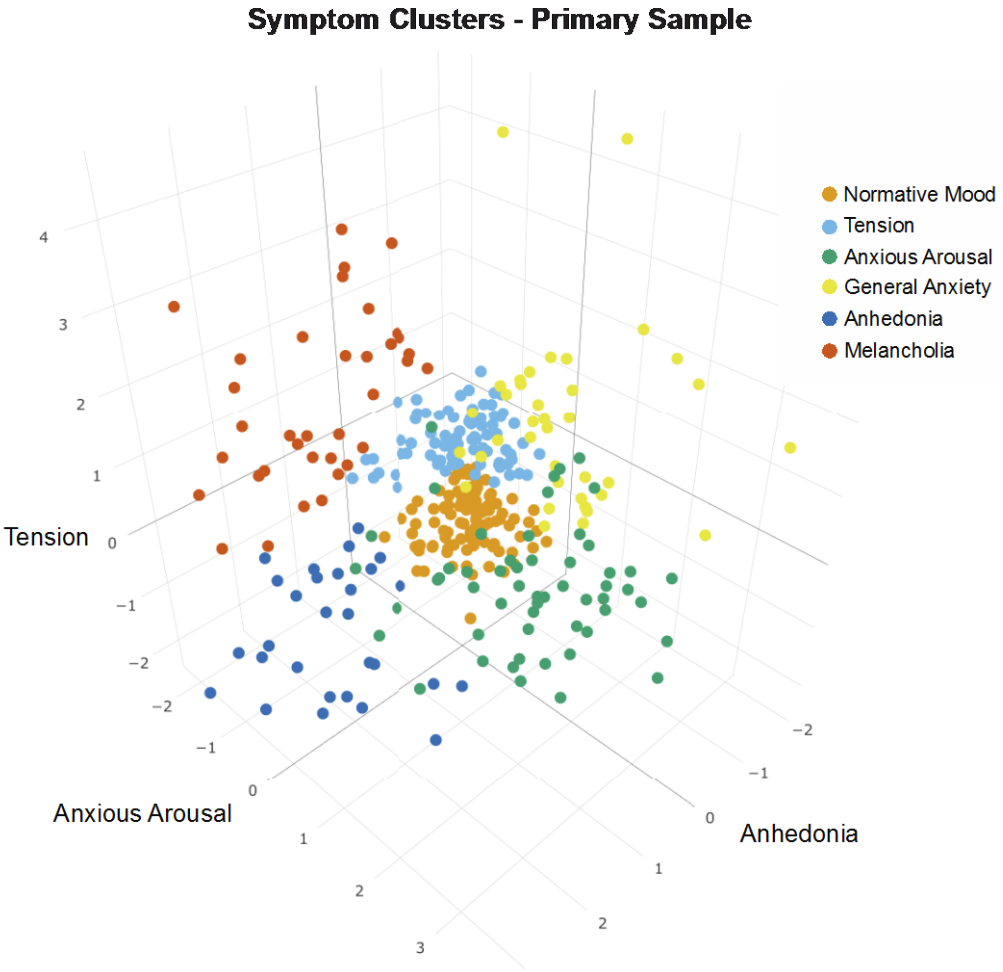


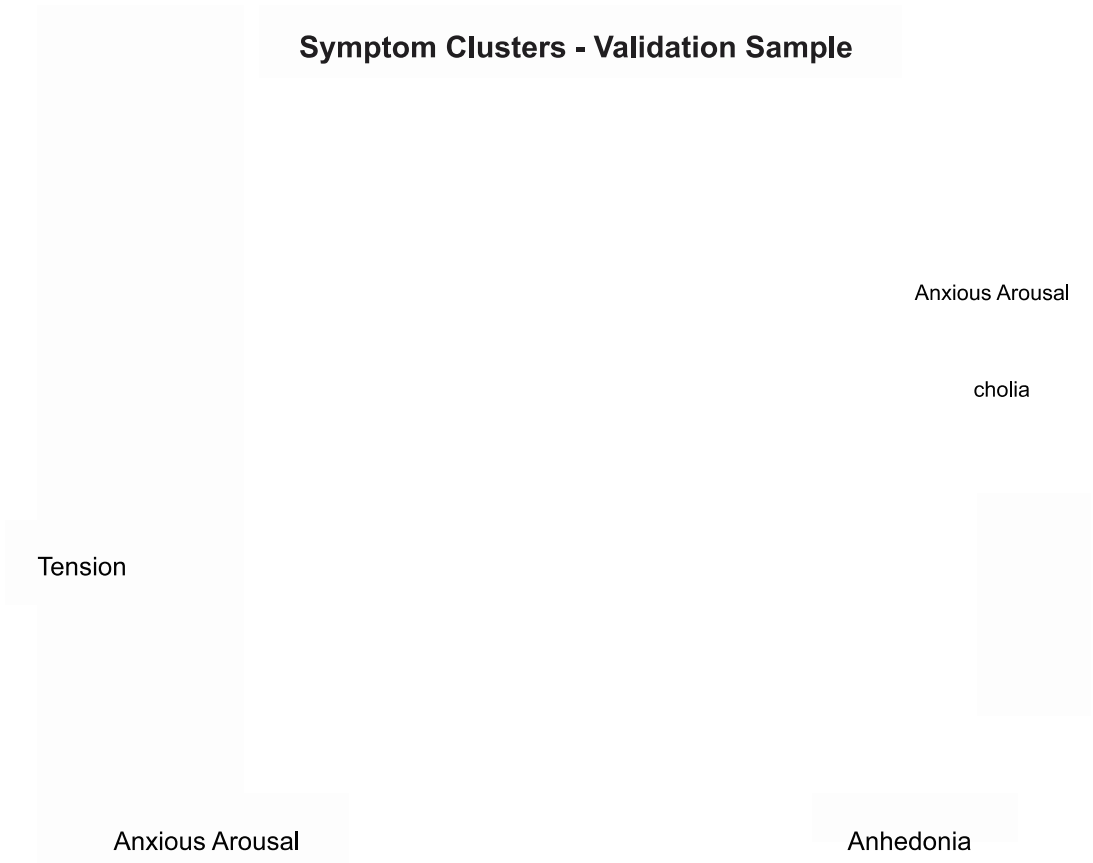
eFigure 8. Symptom profiles in the primary BRAINnet (top) and validation RAD (bottom) samples. Subtypes are on the x axis and average component score on the y axis. Error bars represent +/- 1 standard error (SE) from the mean.



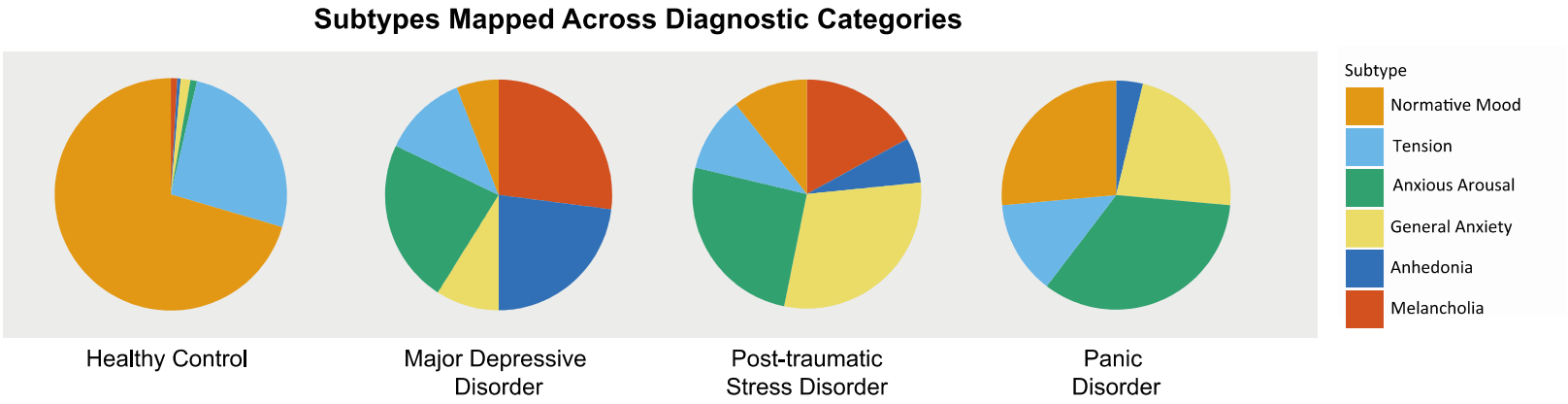


eFigure 9. Scatter plots showing the clusters in the primary BRAINnet (top) and validation RAD (bottom) samples in 3-dimensional PCA space. Each symptom component is a spatial dimension on the X, Y and Z axes. Colors represent each of the six subtypes.

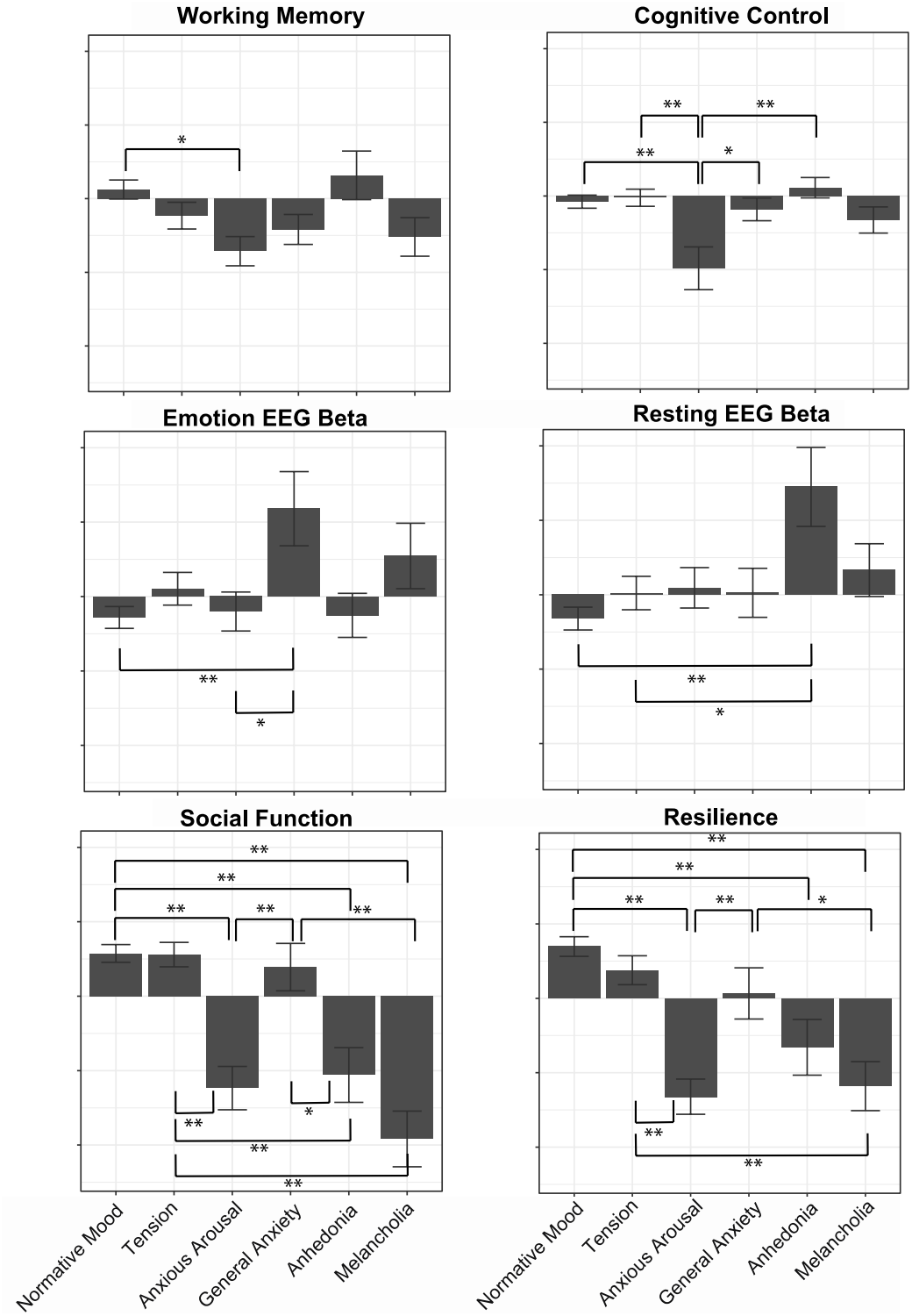



Symptom Clusters - Validation Sample
Anxious Arousal
cholia
Tension
Anxious Arousal
Anhedonia

eFigure 10. Distribution of each diagnostic category by subtype. Each pie chart shows participants from each of the four diagnostic groups. The colors represent each of six subtypes. Each slice of the pie chart shows what percentage of individuals with that diagnosis fell into a specific subtype. For example, out of individuals diagnosed with Major Depressive Disorder, approximately a quarter fell into the Anxious Arousal subtype.



eFigure 11. Tukey HSD post-hoc tests to evaluate significant between-cluster differences on measures of working memory, cognitive control, emotion EEG beta, frontal EEG beta, social function, and resilience. One asterisk refers to a post-hoc test significant at the 0.05 level; two asterisks refer to significance at the 0.01 level.



eResults

Principal Component Analysis (PCA)

Statistical analyses were conducted using the stats, psych, cluster and factoextra packages in R, and NumPy, SciPy, IPython, Jupyter, matplotlib and scikit-learn packages in Python.

The PCA was run with a varimax rotation to ensure the independence of components. The three-component PCA solution aligned with the three DASS-21 subscales (Depression, Anxiety, and Stress), with the exception of two items. The item, "I was aware of dryness in my mouth," falls under the DASS Anxiety subscale, but loaded more highly onto our third "tension" component, and the item "I felt that I was using a lot of nervous energy" is included in the DASS Stress subscale, but loaded more highly onto our second "anxious arousal" component. Because component scores are "weighted" based on the importance of each item in the component, and therefore provide more information than subscale totals alone, component scores rather than DASS subscales were used in subsequent analyses. PCA loadings can be found in eTable 4.

Optimal Cluster Number Determination

The optimal number of clusters was determined using 1) the gap statistic a metric that compares the change in within-cluster dispersion with that expected under a reference null distribution (a distribution with no obvious clustering) (10) (eFigure 1) 2) the Calinski-Harabasz method, which identifies the number of clusters based on the ratio of between-cluster variance to within-cluster variance (11) (eFigure 2), 3) the elbow method, a graphical method that shows the percentage of variance explained as a function of number of clusters (eFigure 3), and 4) the dendrogram, a tree diagram that shows relative similarity between cases (eFigure 4).

The gap statistic compares the difference between within-cluster dispersion with what is expected under a null distribution as a function of cluster number (10). The optimal number of clusters indicated by this metric is the solution that yields the largest gap statistic, signifying the clustering solution is far from a null distribution (i.e. a uniform distribution of points). We calculated the gap statistic for 2 to 20 clusters. The number of Monte Carlo bootstrapping samples ("B" copies of the reference data sets) used was B=500. This metric was maximized at a six-cluster solution (eFigure 1).

The Calinski-Harabasz method identifies the number of clusters based on the ratio of inter-cluster variance to intra-cluster variance (11). Larger scores denote more optimal clustering solutions since it indicates both a large separation between clusters and low separation within clusters. We calculated the Calinski-Harabasz index for 2 to 20 clusters and found the index was maximized at a six-cluster solution (eFigure 2).

The elbow method is a graphical method that shows the percentage of variance explained as a function of cluster number. Based on this method, the number of clusters should be chosen in such a way that adding an additional cluster doesn't significantly improve the modeling of the data (or percentage of variance explained). When plotted, this point can be identified by locating the "elbow" in the plot. We calculated the sum of squared errors (SSE) for 2-20 clusters and located an elbow at 6 clusters (eFigure 3). Because the elbow method is based on visual interpretation, we used this as a secondary method to confirm the gap statistic and Calinski-Harabasz values.

The dendrogram is a tree diagram that shows the relative similarity between cases, and are organized into branches that represent the clusters. Visual analysis of the diagram confirmed a six-cluster solution fits the data (eFigure 4). These four metrics combined strongly indicate a six-cluster solution is optimal in our dataset.

K-Means Clustering Method as Comparison Clustering Method

To ensure our clustering solution was not a specific result of our clustering method, we used scikit-learn's K-means algorithm as a validating clustering method. Specifically, we 1) ran the K-means algorithm seeded with centroids using the K-means++ method, and 2) performed K-means clustering using the resulting cluster centroids of the hierarchical cluster solution. When using the K-means++ algorithm, the cluster centers are initialized in a way that probabilistically favors larger distances between them. When repeated, this method will tend to result in the globally optimal solution, whereas the solution obtained from seeding K-means with the hierarchical centroids can only be treated as a local optimum. Using the solutions found by both methods, we were able to quantify the difference in performance between the global solution and our local solution. In both cases, the K-means algorithm was run ten times (maximum number of iterations for each run: 300), and the solution with the lowest inertia (sum of squared distances for each point to its closest centroid) was used. K=6 was selected as the number of clusters, as

this was reliably determined to be the most optimal. In the resulting cluster solutions, individuals were largely assigned to the same cluster group for both K-means++ (Adjusted Rand Index: 0.79) and seeded K-means (Adjusted Rand Index: 0.80) when compared to the initial hierarchical clustering solution. Additionally, the similarity in performance indicates that the solution found using the hierarchical centroids was the global optimum.

Fit of Individuals to Cluster Assignment

To evaluate how well each individual fit their cluster assignment, we calculated the silhouette scores for individuals in our clustering solution. A silhouette analysis is a commonly used metric of cluster cohesion (how similar an object is to its own cluster) compared to separation (how far it is to neighboring clusters) (12). The average silhouette score for our clustering solution was 0.34.

Stability of Clustering Solution

To evaluate the stability of the clustering solution, we repeated our clustering analysis in 10,000 randomly selected subsamples, each containing 70% of the subjects. In each subsample, we re-ran the same PCA and hierarchical clustering methods as in the original analysis. We chose a six-cluster solution for all subsamples given 1) six was strongly supported to be the optimal number of clusters in the full sample, and 2) in a sample reduced by 30% and original cluster sizes of as small as n=29, allowing cluster number to vary would potentially result in entire clusters being missed. We evaluated the stability of the resulting 10,000 cluster centers by plotting the resulting center locations (eFigure 5 and 6).

We also evaluated the stability of cluster assignments at the individual subject level. In each of the 10,000 subsamples, subjects left out of the cluster identification process (the remaining 30%), were assigned to clusters using linear discriminant analysis classifiers. This left out sample was combined with the left-in sample to form a complete cluster solution. We then tested whether the individual cluster assignments were stable over the 10,000 subsamples by calculating an adjusted Rand score to test the similarity between each clustering solution compared to the original clustering solution (eFigure 7). The average adjusted Rand score was 0.69 (min 0.34, max 0.92).

Subtypes are Replicated in Independent Validation Sample

The three components revealed from the PCA in the independent validation sample closely matched the component loadings in the primary BRAINnet sample (eTable 5). Pearson correlations were run to compare the loadings between components in the validation sample compared to the primary sample (Anhedonia: $r = 0.966$, $p < 0.001$; Anxious Arousal: $r = 0.779$, $p < 0.001$; Tension: $r = 0.866$, $p < 0.001$).

These three components were the inputs to the same hierarchical clustering algorithm used in the primary analysis; agglomerative hierarchical clustering with Ward error sum of squares algorithm was run using the R cluster package. A six-cluster solution was chosen for this validation for the specific aim of evaluating whether the six-cluster solution in the primary analysis replicated in an independent sample. This analysis yielded the same six-cluster solution: Normative Mood, Tension, Anxious Arousal, General Anxiety, Anhedonia, and Melancholia. The symptom profiles of each subtype were replicated in the validation sample (eFigures 8 and 9).

There was a strong equivalence in the structure of the cluster solution across the BRAINnet and the independent RAD validation sample. The profile of scores characterizing each cluster showed the same structure across samples (eFigure 8). This equivalence is especially striking given that the cluster algorithm was run entirely independently for the validation sample, and because the validation sample was acquired at a different time, location, and with a different participant population. Within the context of this overall reproducibility of the cluster structure, there were two small differences across samples in the severity of specific symptom component features that characterized specific clusters. First, for the Anhedonia subtype cluster, participants in the BRAINnet sample had slightly higher severity on the Anhedonia component and slightly lower severity on the Tension component than did participants in the RAD sample. This variation in severity did not impact the equivalence of the overall structure of this cluster, which followed the same U-shaped profile across samples (eFigure 8). Second, for the Melancholia subtype, participants in the BRAINnet sample had slightly lower severity on the Anxious Arousal component than did participants in the RAD sample. Again, however, this variation in severity occurred in the context of a reproducible profile structure for this subtype. It is possible that participant population differences account for these variations in severity within the context of consistent overall cluster structures.

Types Transcend Diagnostic Boundaries

An assumption of our clustering approach was that clusters would not represent diagnostic groups (i.e. Cluster 1 = MDD, Cluster 2 = Panic Disorder, Cluster 3 = PTSD). To test this assumption, we examined the percentage of individuals within each diagnosis that were assigned to each of the six clusters (eTable 6). Additionally, chi-squared tests revealed differences in the frequencies of individuals across cluster, that are important to assess (controls: χ^2=524.33, p<.001; MDD: χ^2=22.88, p<.001; PTSD: χ^2=12.11, p=.03; Panic: χ^2=14.64, p=.006).

Each diagnostic group had a unique pattern. Controls, while assigned predominantly to the normative mood cluster, had 25% of individuals assigned to the Tension cluster, with a couple individuals spread between the remaining four clusters. The MDD participants were almost equally split between Anxious Arousal, Anhedonia, and Melancholia cluster. The Panic Disorder participants, in contrast, were represented mainly in the Normative Mood, Anxious Arousal, and General Anxiety clusters. The PTSD participants also had a unique distribution, being more evenly split than the other diagnostic groups across all six clusters, with comparatively more representation in the Anxious Arousal and General Anxiety clusters. These results give strong evidence that the clustering solution does not represent diagnosis, and that diagnosis splits between the clusters in different patterns.

Consideration of Sex Differences

It is important to consider sex differences in the clusters, as sex differences may affect domain structure. A chi-square test revealed there was no statistically significant association between sex and cluster (χ^2=4.09, p =0.54). eTable 7 shows the sex distribution by cluster.

Comparison of Variance Explained by Cluster Versus Diagnosis

We examined the variance explained by both the 6-cluster solution and conventional DSM groups (eTable 8). Partial eta squared values we calculated as variance measures. The 6-cluster solution explained more variance than DSM diagnosis on the working memory, cognitive control, emotion EEG beta, resting EEG beta, social skills, and resilience measures.

Because the clustering solution explained more variance than diagnosis on each of these external measures, we are confident in claiming that our clustering approach can, at minimum, offer additional information about individuals at the symptom, neurocognitive, neurophysiological, and functioning domain levels, that can be used as a complementary approach alongside diagnostic groupings.

Consideration of Comorbidity: Comparison of Clustering Solution vs. Diagnosis

We further examined the variance explained by the 6-cluster solution compared to DSM groups by including comorbid disorders as covariates (eTable 9). Comorbid disorders of Panic Disorder, Generalized Anxiety Disorder, Post-traumatic Stress Disorder, Attention-Deficit/Hyperactivity Disorder, Major Depressive Disorder, Dysthymia, Obsessive-Compulsive Disorder, and Social Anxiety Disorder were included as covariates in an Analysis of Covariance (ANCOVA) test. The results for the cluster versus diagnosis comparison, without comorbidity, outlined above, held for this ANCOVA. That is, the 6-cluster solution explained more variance than diagnosis, with comorbid disorders as covariates, on working memory, cognitive control, resting EEG beta, emotion EEG beta, and social skills. The only exception was that diagnosis with these covariates explained more variance on the resilience measure of daily functioning. This result suggests that resilience may be a functional capacity that is to a large extend independent of transdiagnostic clusters defined by neurocognitive and brain-based measures.

Post-Hoc Tests using Tukey Honest Significant Difference (HSD)

To evaluate which individual groups differed from each other in our significant one-way ANOVA tests, we ran the Tukey HSD post-hoc test to compare the neurocognitive, EEG, and psychosocial function measures among subtype groups (eTable 10, eFigure 11).

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