

This is an Open Access document downloaded from ORCA, Cardiff University's institutional repository:<https://orca.cardiff.ac.uk/id/eprint/171613/>

This is the author's version of a work that was submitted to / accepted for publication.

Citation for final published version:

Hilbert, Kevin, Boeken, Ole Jonas, Langhammer, Till, Groenewold, Nynke A., Bas-Hoogendam, Janna Marie, Aghajani, Moji, Zugman, André, Åhs, Fredrik, Arolt, Volker, Beesdo-Baum, Katja, Björkstrand, Johannes, Blackford, Jennifer U., Blanco-Hinojo, Laura, Böhnlein, Joscha, Bülow, Robin, Cano, Marta, Cardoner, Narcis, Caseras, Xavier, Dannowski, Udo, Domschke, Katharina, Fehm, Lydia, Feola, Brandee, Fredrikson, Mats, Goossens, Liesbet, Grabe, Hans J., Grotegerd, Dominik, Gur, Raquel E., Hamm, Alfons O., Harrewijn, Anita, Heinig, Ingmar, Herrmann, Martin J., Hofmann, David, Jackowski, Andrea P., Jansen, Andreas, Kaczkurkin, Antonia N., Kindt, Merel, Kingsley, Ellen N., Kircher, Tilo, Klahn, Anna L., Koelkebeck, Katja, Krug, Axel, Kugel, Harald, Larsen, Bart, Leehr, Elisabeth J., Leonhardt, Lieselotte, Lotze, Martin, Margraf, Jürgen, Michalowski, Jarosław, Muehlhan, Markus, Nenadic, Igor, Pan, Pedro M., Pauli, Paul, Peñate, Wenceslao, Pittig, Andre, Plag, Jens, Pujol, Jesus, Richter, Jan, Rivero, Francisco L., Salum, Giovanni A., Satterthwaite, Theodore D., Schäfer, Axel, Schäfer, Judith, Schienle, Anne, Schneider, Silvia, Schrammen, Elisabeth, Schruers, Koen, Schulz, Stefan M., Seidl, Esther, Stark, Rudolf M., Stein, Frederike, Straube, Benjamin, Straube, Thomas, Ströhle, Andreas, Suchan, Boris, Thomopoulos, Sophia I., Ventura-Bort, Carlos, Visser, Renee, Völzke, Henry, Wabnegger, Albert, Wannemüller, André, Wendt, Julia, Wiemer, Julian, Wittchen, Hans-Ulrich, Wittfeld, Katharina, Wright, Barry, Yang, Yunbo, Zilverstand, Anna, Zwanzger, Peter, Veltman, Dick J., Winkler, Anderson M., Pine, Daniel S., Jahanshad, Neda, Thompson, Paul M., Stein, Dan J., Van der Wee, Nic J.A. and Lueken, Ulrike 2024. Cortical and subcortical brain alterations in specific phobia and its animal and blood-injection-injury subtypes: a mega-analysis from the ENIGMA Anxiety Working Group. *The American Journal of Psychiatry* 181 (8), 728–740. [10.1176/appi.ajp.20230032](https://doi.org/10.1176/appi.ajp.20230032)

Publishers page: <http://dx.doi.org/10.1176/appi.ajp.20230032>

Please note:

Changes made as a result of publishing processes such as copy-editing, formatting and page numbers may not be reflected in this version. For the definitive version of this publication, please refer to the published source. You are advised to consult the publisher's version if you wish to cite this paper.

This version is being made available in accordance with publisher policies. See <http://orca.cf.ac.uk/policies.html> for usage policies. Copyright and moral rights for publications made available in ORCA are retained by the copyright holders.



number of words: 5293

number of tables: 2

number of figures: 3

Cortical and subcortical brain alterations in specific phobia and its animal and blood-injection-injury subtypes: a mega-analysis from the ENIGMA-Anxiety Working Group

Running Title: ENIGMA mega-analysis in specific phobia

Authors

Kevin Hilbert¹, PhD, Ole Jonas Boeken¹, MSc, Till Langhammer¹, MSc, Nynke A. Groenewold^{2,3}, PhD, Janna Marie Bas-Hoogendam^{4,5,6}, PhD, Moji Aghajani^{7,8}, PhD, André Zugman⁹, MD, PhD, Fredrik Åhs¹⁰, PhD, Volker Arolt¹¹, MD, Katja Beesdo-Baum¹², PhD, Johannes Björkstrand¹³, PhD, Jennifer U. Blackford¹⁴, PhD, Laura Blanco-Hinojo^{15,16}, PhD, Joscha Böhnlein¹⁷, PhD, Robin Bülow¹⁸, MD, MSc, Marta Cano¹⁹⁻²², PhD, Narcis Cardoner^{20,22,23}, MD, PhD, Xavier Caseras²⁴, PhD, Udo Dannlowski¹¹, PhD, MD, Katharina Domschke²⁵, MD, Lydia Fehm¹, PhD, Brandee Feola²⁶, PhD, Mats Fredrikson²⁷, PhD, Liesbet Goossens²⁸, PhD, Hans J. Grabe^{29,30}, MD, Dominik Grotegerd¹¹, PhD, Raquel E. Gur³¹, MD, PhD, Alfons O. Hamm³², PhD, Anita Harrewijn^{9,33}, PhD, Ingmar Heinig³⁴, PhD, Martin J. Herrmann³⁵, PhD, David Hofmann³⁶, PhD, Andrea P. Jackowski³⁷, PhD, Andreas Jansen³⁸, PhD, Antonia N. Kaczurkin³⁹, PhD, Merel Kindt⁴⁰, PhD, Ellen N. Kingsley⁴¹, MSc, Tilo Kircher⁴², PhD, MD, Anna L. Klahn⁴³, PhD, Katja Kölkebeck⁴⁴, MD, Axel Krug^{42,45}, PhD, Harald Kugel⁴⁶, PhD, Bart Larsen³¹, PhD, Elisabeth J. Leehr¹¹, PhD, Lieselotte Leonhardt³⁴, MSc, Martin Lotze⁴⁷, MD, Jürgen Margraf⁴⁸, PhD, Jarosław Michałowski⁴⁹, PhD, Markus Muehlhan^{50,51}, PhD, Igor Nenadić⁴², MD, Pedro M. Pan³⁷, MD, PhD, Paul Pauli⁵², PhD, Wenceslao Peñate⁵³, PhD, Andre Pittig⁵⁴, PhD, Jens Plag^{55,56}, MD, Jesus Pujol^{16,57}, MD, PhD, Jan Richter^{32,58}, PhD, Francisco L. Rivero^{53,59}, PhD, Giovanni A. Salum⁶⁰, MD, PhD, Theodore D. Satterthwaite³¹, MD, Axel Schäfer⁶¹, PhD, Judith Schäfer³⁴, PhD, Anne Schienle⁶², PhD, Silvia Schneider⁶³, PhD, Elisabeth Schrammen¹¹, MSc, Koen Schruers²⁸, MD, PhD, Stefan Schulz^{52,64}, PhD, Esther Seidl⁶⁵, PhD, Rudolf M. Stark⁶⁶, PhD, Frederike Stein⁴², PhD, Benjamin Straube⁶⁷, PhD, Thomas Straube³⁶, PhD, Andreas Ströhle⁶⁸, MD, Boris Suchan⁶⁹, PhD, Sophia I. Thomopoulos⁷⁰, BA, Carlos Ventura-Bort⁷¹, PhD, Renee Visser⁴⁰, PhD, Henry Völzke⁷², MD, Albert Wabnegger⁶², PhD, André Wannemüller⁶⁹, PhD, Julia Wendt⁷¹, PhD, Julian Wiemer⁵², PhD, Hans-Ulrich Wittchen, PhD, Katharina Wittfeld^{29,30}, PhD, Barry Wright⁷³, MD, MBBS, FRCpsych, Yunbo Yang⁶⁷, PhD, Anna Zilverstand⁷⁴, PhD, Peter Zwanzger^{75,76}, MD, Dick J. Veltman⁸, MD, PhD, Anderson M. Winkler⁹, MD, DPhil, Daniel S. Pine⁹, MD, Neda Jahanshad⁷⁰, PhD, Paul M. Thompson⁷⁰, PhD, Dan J. Stein⁷⁷, MD, PhD, Nic J.A. Van der Wee^{4,6}, MD, PhD, Ulrike Lueken¹, PhD

Corresponding author

Kevin Hilbert

Humboldt-Universität zu Berlin, Faculty of Life Sciences, Department of Psychology

Unter der Linden 6, 10099 Berlin, Germany

Phone: 0049-30 2093 9337

E-Mail: Kevin.Hilbert@hu-berlin.de

Affiliations

- 45 1. Department of Psychology, Humboldt-Universität zu Berlin, Berlin, Germany
- 46 2. Neuroscience Institute, Department of Psychiatry and Mental Health, University of Cape Town
- 47 3. South African Medical Research Council (SA-MRC) Unit on Child and Adolescent Health,
- 48 Department of Paediatrics and Child Health, Red Cross War Memorial Children's Hospital, University
- 49 of Cape Town, Cape Town, South Africa
- 50 4. Department of Psychiatry, Leiden University Medical Center, Leiden, The Netherlands
- 51 5. Department of Developmental and Educational Psychology, Institute of Psychology, Leiden
- 52 University, Leiden, The Netherlands
- 53 6. Leiden Institute for Brain and Cognition, Leiden, The Netherlands
- 54 7. Leiden University, Institute of Education & Child Studies, Section Forensic Family & Youth Care,
- 55 Leiden, The Netherlands
- 56 8. Department of Psychiatry, Amsterdam UMC location VUMC, Amsterdam, The Netherlands
- 57 9. Emotion and Development Branch, National Institute of Mental Health, National Institutes of
- 58 Health, Bethesda, MD, USA
- 59 10. Department of Psychology and Social Work, Mid Sweden University, Östersund, Sweden
- 60 11. Institute for Translational Psychiatry, University of Münster, Münster, Germany
- 61 12. Institute for Clinical Psychology und Psychotherapy, Behavioral Epidemiology, Technische
- 62 Universität Dresden, Dresden, Germany
- 63 13. Department of Psychology, Lund University, Lund, Sweden
- 64 14. Munroe-Meyer Institute, University of Nebraska Medical Center, Omaha, NE, USA
- 65 15. MRI Research Unit, Department of Radiology, Hospital del Mar, Barcelona, Spain
- 66 16. IMIM-CIBER de Salud Mental, Instituto de Salud Carlos III., Barcelona, Spain
- 67 17. Institute for Translational Psychiatry, University of Münster, Münster, Germany
- 68 18. Institute of Diagnostic Radiology and Neuroradiology, University Medicine Greifswald,
- 69 Greifswald, Germany
- 70 19. Mental Health Department, Unitat de Neurociència Traslacional, Parc Tauli University Hospital,
- 71 Institut d'Investigació i Innovació Sanitària Parc Taulí (I3PT), Barcelona, Spain
- 72 20. Institut d'Investigació Biomèdica Sant Pau (IIB SANT PAU), Hospital de la Santa Creu i Sant Pau,
- 73 Barcelona, Spain
- 74 21. Department of Psychobiology and Methodology of Health Sciences, Universitat Autònoma de
- 75 Barcelona, Barcelona, Spain
- 76 22. Centro de Investigación Biomédica en Red de Salud Mental, Carlos III Health Institute, Madrid,
- 77 Spain
- 78 23. Department of Psychiatry and Forensic Medicine, Universitat Autònoma de Barcelona,
- 79 Barcelona, Spain
- 80 24. Department of Psychological Medicine and Clinical Neurosciences, Cardiff University Cardiff
- 81 United Kingdom
- 82 25. Department of Psychiatry and Psychotherapy, Medical Center - University of Freiburg, Faculty of
- 83 Medicine, University of Freiburg, Freiburg, Germany
- 84 26. Department of Psychiatry and Behavioral Sciences, Vanderbilt University Medical Center,
- 85 Nashville, TN, USA
- 86 27. Department of Psychology, Uppsala University, Uppsala, Sweden
- 87 28. Department of Psychiatry and Neuropsychology, Maastricht University Medical Center,
- 88 Maastricht, The Netherlands

- 89 29. Department of Psychiatry and Psychotherapy, University Medicine Greifswald, Greifswald,
90 Germany
- 91 30. German Center for Neurodegenerative Diseases (DZNE), Site Rostock/Greifswald, Greifswald,
92 Germany
- 93 31. Department of Psychiatry, University of Pennsylvania, Philadelphia, PA, USA
- 94 32. Department of Biological and Clinical Psychology, University of Greifswald, Greifswald, Germany
- 95 33. Department of Psychology, Education and Child Studies, Erasmus University Rotterdam
96 Bethesda, MD, USA
- 97 34. Institute of Clinical Psychology & Psychotherapy, Technische Universität Dresden, Dresden,
98 Germany
- 99 35. Department of Psychiatry, Psychosomatics and Psychotherapy, Center of Mental Health,
100 University Hospital Würzburg, Würzburg, Germany
- 101 36. Institute of Medical Psychology and Systems Neuroscience, University of Münster, Münster,
102 Germany
- 103 37. LiNC, Department of Psychiatry, Federal University of São Paulo, São Paulo, Brazil
- 104 38. Core-Facility Brainimaging, Faculty of Medicine, University of Marburg, Marburg, Germany
- 105 39. Department of Psychology, Vanderbilt University, Nashville, TN, USA
- 106 40. Department of Psychology, University of Amsterdam, Amsterdam, The Netherlands
- 107 41. COMIC Research, Leeds and York Partnership NHS Foundation Trust, Leeds, United Kingdom
- 108 42. Department of Psychiatry, University of Marburg, Marburg, Germany
- 109 43. Department of Psychiatry and Neurochemistry, Institute of Neuroscience and Physiology, The
110 Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
- 111 44. LVR-Hospital Essen, Department of Psychiatry and Psychotherapy, Institute and Hospital of the
112 University of Duisburg-Essen, Essen, Germany
- 113 45. Department of Psychiatry, University Hospital of Bonn, Bonn, Germany
- 114 46. University Clinic for Radiology, University of Münster, Münster, Germany
- 115 47. Functional Imaging Unit. Diagnostic Radiology and Neuroradiology, University Medicine
116 Greifswald, Greifswald, Germany
- 117 48. Mental Health Research and Treatment Center, Faculty of Psychology, Ruhr-Universität Bochum,
118 Bochum, Germany
- 119 49. Laboratory of Affective Neuroscience in Poznan, SWPS University of Social Sciences and
120 Humanities, Warszawa, Poland
- 121 50. Department of Psychology, Faculty of Human Sciences, Medical School Hamburg, Hamburg,
122 Germany
- 123 51. ICAN Institute for Cognitive and Affective Neuroscience, Medical School Hamburg, Hamburg,
124 Germany
- 125 52. Department of Psychology, University of Würzburg, Würzburg, Germany
- 126 53. Department of Clinical Psychology, Psychobiology and Methodology, Universidad de La Laguna,
127 San Cristóbal de La Laguna, Spain
- 128 54. Translational Psychotherapy, Department of Psychology, Friedrich-Alexander University
129 Erlangen-Nürnberg (FAU), Erlangen, Germany Erlangen Germany
- 130 55. Department of Psychiatry and Psychotherapy, Campus Mitte, Charité-Universitätsmedizin Berlin,
131 Berlin, Germany
- 132 56. Department of Psychiatry, Psychotherapy and Psychosomatics, Alexianer Krankenhaus
133 Hedwigshoehe, St. Hedwig Kliniken, Berlin, Germany

- 134 57. MRI Research Unit, Department of Radiology, Hospital del Mar, Barcelona, Spain
135 58. Department of Experimental Psychopathology, University of Hildesheim, Hildesheim, Germany
136 59. Department of Psychology, Faculty of Health Science, Universidad Europea de Canarias, La
137 Orotava, Spain
138 60. Section on Negative Affect and Social Processes, Hospital de Clínicas de Porto Alegre,
139 Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil
140 61. Bender Institute of Neuroimaging (BION), Justus-Liebig University Gießen, Center of Mind, Brain
141 and Behavior, Universities of Marburg and Gießen, Gießen, Germany
142 62. Department of Psychology, University of Graz, Graz, Austria
143 63. Faculty of Psychology, Clinical Child and Adolescent Psychology, Mental Health Research and
144 Treatment Center, Ruhr-Universität Bochum, Bochum, Germany
145 64. Department of Behavioural Medicine and Principles of Human Biology for the Health Sciences,
146 Universität Trier, Trier, Germany
147 65. Research Group Security and Privacy, Faculty of Computer Science, University of Vienna, Vienna,
148 Austria
149 66. Department of Psychotherapy and Systems Neuroscience, Bender Institute of Neuroimaging
150 (BION), Justus-Liebig University Gießen, Center of Mind, Brain and Behavior, Universities of Marburg
151 and Gießen, Gießen, Germany
152 67. Department of Psychiatry and Psychotherapy, Philipps-University Marburg, Marburg, Germany
153 68. Department of Psychiatry and Neuroscience, Campus Charité Mitte, Charité-Universitätsmedizin
154 Berlin, Berlin, Germany
155 69. Department of Psychology, Ruhr-Universität Bochum, Bochum, Germany
156 70. Imaging Genetics Center, Mark and Mary Stevens Neuroimaging and Informatics Institute, Keck
157 School of Medicine, University of Southern California Marina del Rey, CA, USA
158 71. Department of Biological Psychology and Affective Science, Faculty of Human Sciences,
159 University of Potsdam, Potsdam, Germany
160 72. Institute for Community Medicine, University Medicine Greifswald, Greifswald, Germany
161 73. Health Sciences, University of York, York, United Kingdom
162 74. Department of Psychiatry and Behavioral Sciences, University of Minnesota Medical School,
163 Minneapolis, MN, USA
164 75. KBO-Inn-Salzach-Klinikum, Wasserburg am Inn, Germany
165 76. Department of Psychiatry and Psychotherapy, Ludwig-Maximilians-University of Munich, Munich,
166 Germany
167 77. South African Medical Research Council Unit on Risk & Resilience in Mental Disorders,
168 Neuroscience Institute, Department of Psychiatry and Mental Health, University of Cape Town, Cape
169 Town, South Africa
170
171
172
173
174

175 **Disclosures**

176 KD is a member of the Steering Committee Neurosciences. HJG has received travel grants and
177 speakers honoraria from Fresenius Medical Care, Neuraxpharm, Servier and Janssen Cilag as well as
178 research funding from Fresenius Medical Care. PMP received payment or honoraria for lectures and
179 presentations in educational events for Sandoz, Daiichi Sankyo, Eurofarma, Abbot, Libbs, Instituto
180 Israelita de Pesquisa e Ensino Albert Einstein, Instituto D'Or de Pesquisa e Ensino. PT received a
181 research grant from Biogen, Inc., for research unrelated to this manuscript. All other authors report
182 no financial relationships with commercial interests.

183

184 **Acknowledgements**

185 The authors would like to thank Sophia Seemann, Laurenz Endl and Cassandra Friebe for their help in
186 organizing the data and supporting the quality control process.

187 The ENIGMA Anxiety working group gratefully acknowledges support from the NIH Big Data to
188 Knowledge (BD2K) award (U54 EB020403 to PT). For a complete list of ENIGMArelated
189 grant support please see here: <http://enigma.ini.usc.edu/about-2/funding/>.

190 NAG was supported by a Developing Emerging Academic Leaders Fellowship. Her work was
191 supported by a grant from Carnegie Corporation of New York. The statements made and views
192 expressed are solely the responsibility of the author. JMBH was supported by a Rubicon grant from
193 Dutch Research Council NWO (019.201SG.022). MC was supported by a Instituto de Salud Carlos III
194 Sara Borrell grant CD20/00189 (MC). UD was funded by the Interdisciplinary Center for Clinical
195 Research (IZKF) of the medical faculty of Münster (grant Dan3/012/17 to UD). LG and KS were
196 supported by the Weijerhorst Foundation. JM was funded by the National Science Centre (Narodowe
197 Centrum Nauki, DEC-2011/03/D/HS6/05951 and DEC-2011/03/D/HS6/05578). RMV receives support
198 from the Dutch Research Council (NWO Veni 016.195.246). YY is supported by “The Adaptive Mind”,
199 funded by the Excellence Program of the Hessian Ministry of Higher Education, Science, Research
200 and Art (PIs: TK, BS).

201 This work was supported by multiple grants from the National Institutes of Health (NIH): SIT and PT
202 were supported by R01MH116147, P41EB015922, and R01MH121246. JUB was supported by NIMH
203 K01MH083052. DSP is supported by NIMH Intramural Research Program project ZIA-MH002781.

204 This work was further supported by multiple grants from the German Research Foundation (DFG): to
205 the CRC940/2 (project C5 to KBB and MM), to the FOR2107 (JA1890/7-1 and JA1890/7-2 to AJ;
206 KI588/14-1, KI588/14-2 and KI 588/22-1 to TK; NE2254/1-2, NE2254/3-1, NE2254/4-1 to IN;
207 DA1151/5-1 and DA1151/5-2 to UD; STR 1146/18-1 to BS), to the SFB-TRR58 (projects C09 and Z02
208 to UD and UL; project B01 and project 378414384 to JW). RMS was also funded by the German
209 Research Foundation (DFG grant STA 475/10-1, DFG graduate school "Neuroact - Brain and
210 Behavior).

211 The German research consortia PROTECT-AD ("Providing Tools for Effective Care and Treatment of
212 Anxiety Disorders: Outcome, Mediators and Moderators of Enhanced Extinction Learning " ; PI:
213 HUW), is one out of nine research consortia in the German federal research program "research
214 network on mental disorders ", funded by the German Federal Ministry of Education and Research
215 (BMBF; www.fzpe.de; project no. 01EE1402) and comprises six projects. MRI data were collected as
216 part of project P4 (PIs: TK and BS, University of Marburg; project no. 01EE1402E). SHIP is part of the
217 Community Medicine Research Net of the University Medicine Greifswald, which is supported by the
218 German Federal State of Mecklenburg- West Pomerania. The BHRCS study was supported with
219 grants from the National Institute of Development Psychiatric for Children and Adolescent (INPD;
220 Grant: Fapesp 2014/50917-0 - CNPq 465550/2014-2).

221

222

223

224

225

226

227 **Abstract**

228 **Objective:** Specific phobia is a common anxiety disorder, but the literature on associated brain
229 structure alterations exhibits substantial gaps. The ENIGMA-Anxiety Working Group examined brain
230 structure differences between subjects with specific phobias and healthy controls as well as
231 between animal and blood-injection-injury (BII) subtypes. Additionally, the authors investigated
232 associations of brain structure with symptom severity and age (youth vs. adults).

233 **Methods:** Datasets from 31 original studies were combined to create a final sample with $n_{\text{phobia}}=1452$
234 phobia subjects and $n_{\text{control}}=2991$ healthy participants (62.7% female, 5-90yrs). Imaging processing
235 and quality control were conducted using established ENIGMA-protocols. Subcortical volumes as
236 well as cortical surface area and thickness were examined in a preregistered analysis (osf.io/n6bhz).

237 **Results:** Phobia subjects compared to healthy controls showed mostly smaller subcortical volumes,
238 mixed surface differences and larger cortical thickness across a substantial number of regions.
239 Phobia subgroups also showed differences, including, as hypothesized, larger medial orbitofrontal
240 cortex thickness in BII phobia compared to animal phobia ($n_{\text{BII}}=182$; $n_{\text{animal}}=739$). All findings were
241 driven by adult subjects; the authors found no significant results in children and adolescents.

242 **Conclusions:** Brain alterations associated with specific phobia exceeded those of other anxiety
243 disorders in comparable analyses in extent and effect size and were not limited to reductions in
244 brain structure. Moreover, phenomenological differences between phobia subgroups were reflected
245 in diverging neural underpinnings, including brain areas related to fear processing and higher
246 cognitive processes. The findings implicate brain structure alterations in specific phobia, although
247 subcortical alterations in particular may also relate to broader internalizing psychopathology.

248

249 **Keywords**

250 Specific Phobia, Animal phobia, Blood-Injection-Injury phobia, structural neuroimaging, mega-
251 analysis, ENIGMA

252

253
254
255
256
257
258
259
260
261
262
263
264
265
266

267
268
269
270
271
272
273
274
275
276
277

Introduction

Specific phobia is the most prevalent anxiety disorder (1, 2), with global lifetime prevalence ranging between 2.6-12.5% (3). According to the DSM-IV-TR and DSM-5, it involves marked and disproportionate fear and anxiety or frequent avoidance of particular objects or situations. Its onset is often early in childhood (3) and many cases develop into future internalizing disorders (4). Given its prototypical fear reaction and early onset, specific phobia has been used as a model disorder to investigate the neural processing of fear and fear circuitry dysfunctions (5, 6). Functional neuroimaging studies of the disorder implicate the anterior to mid-cingulate gyrus, the amygdala, insula, thalamus and inferior frontal gyrus (7, 8). These alterations have been related to the rapid processing of external threat stimuli (thalamus (5)), stimulus saliency (amygdala (5); particularly interoception: insula (7); particularly exteroception: anterior cingulate (7)), fear conditioning (amygdala (5)), emotion regulation (anterior cingulate (5)) and impaired emotion appraisal (inferior frontal gyrus (8)). The current paper complements these functional correlates by reporting findings from a large, multi-site investigation examining neuroanatomical correlates of specific phobia.

In contrast to functional MRI investigations, few studies examined differences in brain structure associated with specific phobia, and those were generally conducted in small samples and targeted isolated regions of interest (e.g. 9-11). As structural alterations may underlie the disorder-related functional differences, deeper knowledge of structural correlates is needed. The literature possesses three major gaps. First, the animal phobia subtype exhibits a prototypical, sympathetically mediated fear response (12), whereas the blood-injection-injury (BII) subtype shows a less clear-cut response, with some evidence of a unique diphasic fear response (13, 14). The corresponding neural activation patterns seem to implicate fear-related components such as the amygdala, insula, dorsal anterior cingulate cortex (ACC) and thalamus for the animal subtype, but are less clear-cut for the BII subtype (15-17). In contrast, the orbitofrontal cortex (OFC) has been implicated for BII phobia (15, 17). Given the paucity of research on brain structure associated with specific phobia, it remains unclear if these

278 subtypes indeed manifest unique neurostructural correlates corresponding to functional activation
279 patterns.

280 The few available preliminary findings indicate this might only be partially the case, particularly for
281 the ACC being associated with specific phobia in general and the OFC being associated with the BII
282 subtype specifically (18, 19). Second, despite their early onset during childhood, few studies examine
283 brain structure related to specific phobia before adulthood. Third, previous research on anxiety
284 disorders demonstrated that the presence of depressive comorbidity altered gray matter volumes
285 (20). However, it is unclear whether comorbid depressive symptoms also influence brain structure
286 associated with specific phobia.

287 This investigation aims to address these gaps by comparing brain structure in subjects with specific
288 phobia and healthy subjects. Within the Enhancing NeuroImaging Genetics through Meta-Analysis
289 (ENIGMA) collaboration (21), the ENIGMA-Anxiety Working Group (22) obtained 33 datasets with
290 information on neurostructural correlates of specific phobia and its animal and BII subtypes of which
291 31 datasets (age range 5-90 years) were included. We examine the following hypotheses: 1)
292 Compared to healthy controls, specific phobia subjects across all subtypes would show altered
293 cortical thickness and surface area in the dorsal ACC and the insula, and altered subcortical volumes
294 in the amygdala and thalamus. Additionally, 2) animal phobia subjects would show altered amygdala
295 and thalamus volumes when compared to healthy controls or BII phobia subjects, while BII phobia
296 subjects will show altered cortical thickness and surface area in OFC areas when compared to
297 healthy controls or animal phobia subjects. Furthermore, we expected 3) a linear association of
298 these metrics with symptom severity and 4) a linear association with depression severity for insula,
299 dorsal ACC and amygdala metrics, both within the specific phobia group. This work is also the first
300 investigation of brain structure associated with specific phobia in children and adolescents, but given
301 the paucity of available studies, we refrained from a hypothesis on the relationship with age.

302

303
304
305
306
307
308
309
310
311
312
313
314
315
316
317
318
319
320
321
322
323
324
325
326
327
328

Materials and Methods

Samples

We collected datasets from 33 original datasets acquired on 43 distinct MRI scanners. We included datasets with initially at least 10 subjects with SPH, while datasets with fewer subjects were excluded in total, leading to the inclusion of 31 of the collected datasets. Subjects were included with current or past specific phobia, whether or not specific phobia was the primary diagnosis. Past studies used different criteria for determining specific phobia, from formal diagnoses using standardized clinical interviews to diagnostics based on established cut-off scores in questionnaires. We included both types of studies in order to maximize sample size. Subjects were excluded for a current or lifetime diagnosis of bipolar disorder, psychosis, or schizophrenia. No current or past diagnoses of any mental disorder were allowed for healthy controls. All participants provided written informed consent when participating in the original studies, and these original studies acquired positive evaluation by institutional review boards and ethic committees. The current study was preregistered at the Open Science Framework (osf.io/n6bhz).

This project depended on datasets from original studies. Most of these original studies have not been analyzed for neurostructural correlates of specific phobia, with some exceptions (18, 19, 23, 24). The current analysis provides unprecedented statistical power and heterogeneity regarding the number of participants included with specific phobia.

Imaging processing and quality control

Original studies contributed their data to our mega-analysis either by processing their data on site and sending the resulting subject-level data plus demographic and clinical variables or by sending us raw brain imaging data (structural T1-weighted MR images) so that we performed the processing centrally. Imaging processing and quality control were in both cases conducted using FreeSurfer (25) with established ENIGMA-protocols and instructions for quality control (available at <https://enigma.ini.usc.edu/protocols/imaging-protocols/>). In short, structural images were

329 segmented and processed to calculate volume data for eight subcortical regions per laterality and to
330 calculate surface area and cortical thickness data for thirty-four cortical regions per laterality and the
331 total intracranial volume. Cortical region segmentation was performed according to the Desikan-
332 Killiany cortical atlas (26). Resulting segmentations were checked visually for substantial over- or
333 underestimation; this process was supported by summary statistics, boxplots, and outlier
334 histograms. Individuals were excluded from the cortical or subcortical analysis, respectively, if the
335 FreeSurfer segmentation failed altogether, and if there were over- or underestimations in at least
336 25% of the cortical or subcortical regions. Otherwise, only the data from the affected regions was
337 excluded.

338

339 **Statistical Analysis**

340 FreeSurfer-derived data for cortical and subcortical regions was used as input in a linear mixed
341 model on R version 4.0.4 including disorder state (specific phobia, healthy controls) as variable of
342 interest and age, sex and intracranial volume (ICV) as fixed factors and scanner as random intercepts
343 (supplemental table 1 provides an overview on scanner characteristics and a description of
344 procedures for grouping subjects across studies for this covariate). Here, we deviated from the
345 preregistration as the model was overparameterized for many brain regions in the fundamental
346 group comparison and we thus reduced model complexity by eliminating random slopes. There were
347 rare instances where models for individual areas were still overparameterized for phobia subtype
348 comparisons and dimensional analyses, where we further reduced model complexity. This affected
349 only non-significant areas. To limit multiple testing against the background of the large number of
350 regions, left and right side cortical thickness, surface area and subcortical volumes were averaged.
351 Additionally, p-values from all regions were corrected using the false-discovery rate (FDR) as
352 proposed by Benjamini and Hochberg (27), with FDR corrections run separately for subcortical
353 volumes (eight regions), cortical surface area (34 regions) and cortical thickness (34 regions).
354 Standard errors and effect sizes were calculated according to Nakagawa and Cuthill (2007; 28).

355 A second preregistered analysis was conducted to test for structural correlates specifically for the
356 subtypes (hypothesis 2). This approach was limited to animal and BII subtypes (including dental
357 phobia) for whom sufficient data for subtype analysis was available ($n_{\text{BII}}=182$; $n_{\text{animal}}=739$;
358 supplemental figure 1). For the subtype analysis, disorder subtype (animal subtype, BII subtype) was
359 used as variable of interest on specific phobia subjects only. Here, we included only specific phobia
360 subjects with a single subtype, not with multiple subtypes. As this analysis yielded interesting results,
361 we conducted two additional post-hoc analyses with animal phobia subjects vs healthy controls and
362 BII phobia subjects vs healthy controls, which were not included in the preregistration.

363 Three further preregistered analyses examined dimensional associations by using phobia severity,
364 trait anxiety, and depression severity as variables of interest (hypotheses 3 and 4). As phobia
365 severity was assessed using a broad range of questionnaires across original studies, we classified
366 participants into ten ordinal categories according to their questionnaire score within their original
367 study. These ten ordinal were used in linear mixed models (deviating from the preregistration which
368 mistakenly specified ordinal regressions that would require ordinal outcomes rather than ordinal
369 predictors). For trait anxiety and depression severity, we used the State-Trait Anxiety Inventory –
370 Trait version (STAI-T; 29) and the Beck Depression Inventory-II (BDI-II; 30) scores, respectively.

371 Accompanying these main analyses, we conducted further exploratory analyses on robustness of the
372 results by testing whether areas still showed significant differences between groups when using
373 specific phobia subjects with formal diagnosis only, using subjects with current specific phobia only,
374 using specific phobia and healthy controls subjects with and without medication only, examining
375 adults (>21 years) only and children and adolescents (≤ 21 years) only, excluding subjects from
376 scanners with less than ten participants, excluding subjects additional comorbidities, and examining
377 the impact of education, re-including outliers, and unilateral vs bilateral regions (for details see
378 supplemental methods). Given the diverging findings for the age groups in the adults only and
379 children and adolescents only analyses, we added further exploratory analyses on an age-by-
380 diagnosis interaction (details also in the supplemental methods).

381

382

Results

383

We received data from n=5330 individuals. Table 1 provides detailed information on the amount and

384

reason of excluded subjects. The final sample consisted of n=4443 participants, with $n_{\text{phobia}}=1452$

385

specific phobia subjects and $n_{\text{control}}=2991$ healthy controls. Sociodemographic information can be

386

found in table 2, and supplemental table 2 shows current and lifetime comorbidities within specific

387

phobia subjects. Compared regarding sociodemographic variables, specific phobia subjects included

388

significantly less males, were significantly younger, and had significantly fewer years of education

389

compared to healthy controls (all $ps < 0.001$).

390

391

[Tables 1-2]

392

393

Specific phobia subjects versus healthy controls

394

The main group comparison showed significantly smaller subcortical volumes for specific phobia

395

(n=1452) vs healthy controls (n=2991) in several regions including the caudate, putamen and

396

hippocampus, significantly larger thickness in several cortical regions and mixed alterations in

397

surface area (figure 1 for effect sizes and graphical overview; supplemental table 3 for detailed

398

results on all available regions including sample sizes per region). These findings remained robust for

399

most exploratory analyses (supplemental table 4). However, when including education as an

400

additional covariate, only subcortical volume differences in the caudate nucleus, putamen, and

401

accumbens remained significant.

402

Crucially, when splitting the sample into adults (>21 years; n=2650) and children to adolescents

403

(=<21 years; n=1793), the majority of findings remained significant in adults, and additional group

404

differences emerged for the insula, banks superior temporal sulcus, the entorhinal cortex and the

405

temporal pole (supplemental table 5). Conversely, no group differences emerged for any regions for

406

the comparison of specific phobia vs healthy controls in children to adolescents. The age-by-

407 diagnosis analysis across the whole range of age found no significant interactions between age and
408 diagnosis.

409

410 [Figure 1]

411

412 **Direct comparison of animal and BII subtypes**

413 The comparison of animal (n=739) versus BII phobia (n=182) subjects showed a significant difference
414 in one area included in our hypotheses, with BII phobia subjects showing larger cortical thickness in
415 the medial OFC. Additionally, there were further group differences in areas not included in the
416 hypotheses for cortical thickness, namely within the lateral occipital cortex, pars orbitalis, pars
417 triangularis, pericalcarine, posterior cingulate, rostral middle frontal, superior frontal cortex and
418 frontal pole (figure 2; supplemental table 6 for detailed results on all available regions including
419 sample sizes per region). Again, these findings overall remained robust when re-including outliers,
420 excluding scanners with <10 participants, excluding additional comorbidities, excluding subjects with
421 psychotropic medication, using unilateral instead of bilateral data, and adding education as an
422 additional covariate (supplemental table 7). Results were less robust, when allowing only for
423 subjects with a formal diagnosis of specific phobia, restricting specific phobia subjects to those
424 taking medication, and when allowing only for subjects with current specific phobia (supplemental
425 table 7). However, these follow-up examinations had to use considerably reduced animal and BII
426 phobia sample sizes.

427 Again, splitting the sample of subtypes in adults (>21 years; n=605) and children and adolescents
428 (= <21 years; n=316) had a considerable effect. For adults, group differences in the medial OFC and
429 most other regions remained significant, and additional thickness differences in the transverse
430 temporal gyrus emerged (supplemental table 8). Similar to the main analysis, no group differences
431 emerged for any regions for the comparison of specific phobia vs healthy controls for the children to

432 adolescents. The age-by-subtype analysis across the whole range of age did not find any significant
433 interactions.

434

435 [Figure 2]

436

437 **Comparison of animal and BII subtypes versus healthy controls**

438 Given the considerable number of significant differences between the animal and BII subgroups in
439 the previous analysis, we performed an additional, exploratory comparison of both subtypes
440 ($n_{\text{animal}}=739$; $n_{\text{BII}}=182$) with healthy controls ($n=2991$) which was not specified in the preregistration.

441 These analyses found significant differences for animal phobia compared to healthy controls in a
442 large number of subcortical and cortical areas including smaller volume in the caudate, putamen and
443 hippocampus and larger medial OFC cortical surface consistent with effects in the main analysis of
444 specific phobia vs healthy controls and further areas (figure 3, yellow; supplemental table 9 for
445 detailed results on all available regions including sample sizes per region). Conversely, only relatively
446 few group differences emerged for BII phobia subjects against healthy controls. This included larger
447 medial OFC cortical surface (figure 3, blue; supplemental table 10 for detailed results on all available
448 regions including sample sizes per region).

449

450 [Figure 3]

451

452 **Dimensional effects of phobia severity, trait anxiety and depression severity**

453 No significant associations with phobia severity, trait anxiety or depression severity emerged for any
454 area - neither across all phobia subjects nor in animal or BII phobia subjects separately (phobia
455 severity: $n_{\text{all}} = 825$, $n_{\text{animal}} = 614$, $n_{\text{bii}} = 164$; trait anxiety: $n_{\text{all}} = 809$, $n_{\text{animal}} = 451$, $n_{\text{bii}} = 50$; depression
456 severity: $n_{\text{all}} = 622$, $n_{\text{animal}} = 399$, $n_{\text{bii}} = 69$). As there was also sufficient variability in trait anxiety within

457 the healthy controls, we conducted an additional analysis in this group to examine the impact of trait
458 variability in a normative group (n=1755), which also yielded no significant results.

459

460

Discussion

461 We here present a preregistered analysis from the ENIGMA-Anxiety Working Group that examined
462 brain structure differences between subjects with specific phobia and healthy controls, as well as
463 between two phobia subtypes, between different age groups, and in relation to anxiety and
464 depression severity. We found group differences between specific phobia and healthy controls in
465 most subcortical areas including the hippocampus, caudate, putamen (smaller volume in specific
466 phobia) and pallidum (larger volume in specific phobia), and multiple cortical areas. These group
467 differences were largely driven by animal phobia but not BII phobia subjects. Comparing these two
468 subgroups directly, we found larger cortical thickness in the medial OFC in BII phobia subjects in line
469 with a-priori hypotheses, and in further cortical areas. We did not find associations between brain
470 structure and symptom severity. Finally, all findings occurred exclusively in adult subjects but not in
471 children and adolescents.

472 Group differences between specific phobia and healthy controls, which were largely driven by
473 animal phobia subjects, exceeded those reported for Generalized Anxiety Disorder and Social
474 Anxiety Disorder in comparable analyses in extent and effect size (31, 32). Notably, these group
475 differences were not limited to smaller volume, surface area and thickness, but also included
476 enlarged areas, contrary to other ENIGMA studies within the internalizing spectrum, such as in
477 Obsessive-Compulsive Disorder or Major Depression (33, 34). While these findings implicate notable
478 brain structure alterations in specific phobia, they appeared minimally related to our a-priori
479 hypotheses. Furthermore, they showed no overlap with major regions emphasized in functional
480 activation maps for specific phobia (7, 8), no overlap with structural alterations commonly
481 associated with general psychopathology (35) and no overlap with the regions commonly selected as
482 ROIs in prior studies of specific phobia (9-11). Similar to our results, a previous whole-brain

483 investigation also failed to detect specific-phobia-related differences in regions such as the
484 amygdala, thalamus and insula (18). This suggests that specific-phobia-related alterations in brain
485 structure may not match the amygdalocentric perspective that prevailed in functional research for
486 some time. Further, it raises the question to what degree specific-phobia-related alterations in brain
487 structure are related to alterations in neural activation. While the relationship between structural
488 and functional brain alterations is not fully understood yet, initial evidence suggests that structural
489 alterations first occur in central hub regions of the brain and then propagate along functional (and,
490 less clearly, anatomical and genetic) connectivity patterns (41). A promising candidate for explaining
491 this pattern is nodal stress (41). Nodal stress suggests that brain hub regions are particularly strained
492 due to strong network activity and may first show disorder-associated alterations (42). This potential
493 mechanism suggests that functional alterations precede structural changes in the same regions, thus
494 disorder-associated functional and structural maps should show considerable overlap. This is only
495 partially evident in the comparison of structural changes from our study to functional changes from
496 meta-analyses to date (e.g. altered activation in the hippocampus, putamen, caudate and lingual
497 gyrus in (7), but not with major regions such as the dorsal ACC or anterior insula as discussed above).
498 However, comparing these structural and functional alterations is hindered by the fact that meta-
499 analyses of functional changes in specific phobia to date are based on very limited sample sizes, well
500 below the sample size of our current study. The impression that the relationship of structural to
501 functional changes in specific phobia is not yet fully understood is additionally strengthened by the
502 lack of any significant correlations between brain structure and phobia or trait anxiety severity in our
503 study, as opposed to previous functional studies which reported such associations (16, 36). At the
504 same time, our findings do implicate various new subcortical brain structures in the neuroanatomy
505 of specific phobia, with most subcortical regions showing significantly different, and mostly reduced,
506 volumes in specific phobia compared to healthy controls. Interestingly, similar subcortical
507 differences in the putamen and pallidum have been found in a related ENIGMA-Anxiety Working
508 Group study on Social Anxiety Disorder (32). Additionally, there was a nonsignificant trend for

509 subcortical volume in the pallidum to be inversely associated with depression severity in our current
510 analysis. Together, these results suggest that the reported subcortical differences may at least partly
511 be related to broader internalizing psychopathology instead of being a specific neural substrate of
512 specific phobia. The current results also underscore the need to complement analyses with pre-
513 selected ROIs with more whole-brain examinations of specific phobia brain structure in future
514 studies, and the importance of being sufficiently powered for these kinds of analysis.

515 Direct comparisons between phobia subgroups showed significant differences between animal
516 ($n_{\text{animal}}=739$) and BII phobia ($n_{\text{BII}}=182$) in a variety of cortical regions including the medial OFC, where
517 animal phobia subjects showed lower cortical thickness. These results fit with previous results of
518 increased volumes (18) in BII compared to animal phobia in orbitofrontal regions, and align with the
519 idea of fear-processing in BII phobia involving impairment during cognitive processes such as
520 stimulus appraisal and evaluation (17, 37) and emotion regulation (37, 38) to a larger extent. For
521 other areas implicated in our a-prior hypotheses, particularly the amygdala and thalamus, functional
522 differences between phobia groups were also common in earlier studies (15, 16, 36, 38), but both
523 areas exhibited only non-significant trends in our analysis. Additionally, in our analysis, volume and
524 cortical thickness in these areas were not related to phobia severity in or across subgroups. In
525 conclusion, the current results further provide evidence that phenomenological differences between
526 subgroups also relate to diverging neural underpinnings, but more research is needed to understand
527 the exact functional implications of this finding, particularly regarding the less sympathetically
528 mediated, sometimes even diphasic fear response in the BII subtype.

529 The current study examined data on phobia-related differences in brain structure in children and
530 adolescents. However, all group differences were exclusively found in the adult subsamples.
531 Although this is in line with ENIGMA-Anxiety study on Social Anxiety Disorder (32), this was a
532 surprising finding given that disorder onset early during childhood is so common (4) and given that
533 other anxiety disorders and even youth at risk for anxiety disorders appear to be accompanied by

534 neurofunctional and –structural correlates (39). However, adults may have substantially stronger
535 levels of disorder persistence compared to children and adolescents, as specific phobia cases
536 typically begin in childhood (4) but most will remit before adulthood (40). Alternatively, the finding
537 could be associated with increased overall psychopathology load during adulthood, or with subtle
538 neuroanatomical correlates of specific phobia during youth that disappear against the predominant
539 age-related changes and brain variability. Future research on the trajectory of phobia associated
540 alterations over the developmental span and taking into account disorder duration and persistence
541 is needed to elucidate this null finding. Finally, this null finding might also be influenced by smaller
542 power for children and adolescents in the disorder subtype analyses. For the main comparison of
543 specific phobia vs healthy controls however, we did not find indications of substantially lower power
544 in children and adolescents compared to adults.

545 We here report an examination of brain structure alterations associated with specific phobia
546 substantially exceeding previous sample sizes. Still, sample sizes remained moderate for individual
547 analyses, particularly regarding the phobia subgroups. Additionally, despite using established
548 ENIGMA protocols and procedures, harmonization of this wealth of data is only possible to a limited
549 degree. Particularly site-specific scanners and scan sequences, FreeSurfer versions, raters for quality
550 control and differences in phobia severity questionnaires may induce systematic variation in the
551 data unrelated to group membership. We aimed to model site-specific scanners and scan sequences
552 within our analytic approach, but residual effects may remain, particularly as sample sizes per
553 scanner were considerably imbalanced. This might have influenced parameter estimates particularly
554 for scanners with only few participants. Sites also used a variety of different phobia severity
555 questionnaires, which we aimed to ameliorate by transforming data into site-specific centiles, but
556 this procedure naturally leads to information loss.

557 In conclusion, we here present a preregistered analysis by the ENIGMA-Anxiety Working Group on
558 brain structure associated with specific phobia. Our findings implicate brain structure alterations in

559 specific phobia, although subcortical alterations in particular may also relate to broader internalizing
560 psychopathology. Subgroup specific analyses support the idea that phenomenological differences
561 between subgroups also relate to diverging neural underpinnings, with brain areas related to higher
562 cognitive processes being particularly implicated in BII phobia. Interestingly, specific phobia-related
563 differences emerged only for adults but not for children or adolescents. This may be due to stronger
564 levels of disorder persistence, increasing overall psychopathology load in adult patients, or to age-
565 related developmental changes in the brain. Examining and disentangling the age- and disorder
566 course-related trajectories of specific phobia in the brain may be promising avenues for further
567 research. Additionally, future analyses of resting-state data may provide valuable insights on the role
568 of large-scale brain circuits. Overall, brain structure in specific phobia is understudied and its role in
569 the etiopathogenesis of the disorder is not well understood. This work is a starting point for further
570 investigations on the role of brain morphometric alterations for our understanding and treatment of
571 specific phobia.

- 573 1. Kessler RC, Petukhova M, Sampson NA, et al: Twelve-month and lifetime prevalence and
574 lifetime morbid risk of anxiety and mood disorders in the United States. *Int J Methods Psychiatr*
575 *Res* 2012; 21(3):169-184
- 576 2. Wittchen HU, Jacobi F, Rehm J, et al: The size and burden of mental disorders and other
577 disorders of the brain in Europe 2010. *Eur Neuropsychopharmacol* 2011; 21(9):655-679
- 578 3. Wardenaar KJ, Lim CCW, Al-Hamzawi AO, et al: The cross-national epidemiology of specific
579 phobia in the World Mental Health Surveys. *Psychol Med* 2017; 47(10):1744-1760
- 580 4. Beesdo-Baum K, Knappe S: Epidemiology and natural course. In: Emmelkamp P, Ehring T,
581 editors. *The Wiley Handbook of Anxiety Disorders*, Wiley & Sons, 2014 pp. 26 - 46
- 582 5. Ipser JC, Singh L, Stein DJ: Meta-analysis of functional brain imaging in specific phobia.
583 *Psychiatry Clin Neurosci* 2013; 67(5):311-322
- 584 6. Linares IM, Trzesniak C, Chagas MH, et al: Neuroimaging in specific phobia disorder: a
585 systematic review of the literature. *Rev Bras Psiquiatr* 2012; 34(1):101-111
- 586 7. Chavanne AV, Robinson OJ: The Overlapping Neurobiology of Induced and Pathological Anxiety:
587 A Meta-Analysis of Functional Neural Activation. *Am J Psychiatry* 2021; 178(2):156-164
- 588 8. Gentili C, Messerotti Benvenuti S, Lettieri G, et al: ROI and phobias: The effect of ROI approach
589 on an ALE meta-analysis of specific phobias. *Hum Brain Mapp* 2019; 40(6):1814-1828
- 590 9. Fisler MS, Federspiel A, Horn H, et al: Spider phobia is associated with decreased left amygdala
591 volume: A cross-sectional study. *Bmc Psychiatry* 2013; 13:70
- 592 10. Fisler MS, Federspiel A, Horn H, et al: Pinpointing regional surface distortions of the amygdala in
593 patients with spider phobia. *J Psychiatry Brain Sci* 2014; 1(2):1-7
- 594 11. Rosso IM, Makris N, Britton JC, et al: Anxiety sensitivity correlates with two indices of right
595 anterior insula structure in specific animal phobia. *Depress Anxiety* 2010; 27(12):1104-1110
- 596 12. McTeague LM, Lang PJ, Wangelin BC, et al: Defensive mobilization in specific phobia: fear
597 specificity, negative affectivity, and diagnostic prominence. *Biol Psychiat* 2012; 72(1):8-18
- 598 13. Ritz T, Meuret AE, Simon E: Cardiovascular activity in blood-injection-injury phobia during
599 exposure: evidence for diphasic response patterns? *Behaviour research and therapy* 2013;
600 51(8):460-468
- 601 14. Thyer BA, Curtis GC: On the diphasic nature of vasovagal fainting associated with blood-injury-
602 illness phobia. *Pavlov J Biol Sci* 1985; 20(2):84-87
- 603 15. Hilbert K, Evens R, Maslowski NI, et al: Fear processing in dental phobia during crossmodal
604 symptom provocation: An fMRI study. *Biomed Res Int* 2014:1-9
- 605 16. Lueken U, Hilbert K, Stolyar V, et al: Neural substrates of defensive reactivity in two subtypes of
606 specific phobia. *Soc Cogn Affect Neurosci* 2014; 9(11):1668-1675
- 607 17. Lueken U, Kruschwitz JD, Muehlhan M, et al: How specific is specific phobia? Different neural
608 response patterns in two subtypes of specific phobia. *Neuroimage* 2011; 56(1):363-372
- 609 18. Hilbert K, Evens R, Maslowski NI, et al: Neurostructural correlates of two subtypes of specific
610 phobia: A voxel-based morphometry study. *Psychiatry Res Neuroimaging* 2015; 231(2):168-175
- 611 19. Lueken U, Hilbert K, Wittchen HU, et al: Diagnostic classification of specific phobia subtypes
612 using structural MRI data: A machine-learning approach. *J Neural Transm* 2015; 122(1):123-134
- 613 20. van Tol MJ, van der Wee NJ, van den Heuvel OA, et al: Regional brain volume in depression and
614 anxiety disorders. *Arch Gen Psychiatry* 2010; 67(10):1002-1011
- 615 21. Thompson PM, Jahanshad N, Ching CRK, et al: ENIGMA and global neuroscience: A decade of
616 large-scale studies of the brain in health and disease across more than 40 countries.
617 *Translational psychiatry* 2020; 10(1):100
- 618 22. Bas-Hoogendam JM, Groenewold NA, Aghajani M, et al: ENIGMA-anxiety working group:
619 Rationale for and organization of large-scale neuroimaging studies of anxiety disorders. *Hum*
620 *Brain Mapp* 2022; 43(1):83-112

- 621 23. Schienle A, Scharmüller W, Leutgeb V, et al: Sex differences in the functional and structural
622 neuroanatomy of dental phobia. *Brain Struct Funct* 2013; 218(3):779-787
- 623 24. Wabnegger A, Scharmüller W, Schienle A: Sex-specific associations between grey matter volume
624 and phobic symptoms in dental phobia. *Neurosci Lett* 2014; 580:83-87
- 625 25. Fischl B, Salat DH, Busa E, et al: Whole brain segmentation: automated labeling of
626 neuroanatomical structures in the human brain. *Neuron* 2002; 33(3):341-355
- 627 26. Desikan RS, Segonne F, Fischl B, et al: An automated labeling system for subdividing the human
628 cerebral cortex on MRI scans into gyral based regions of interest. *Neuroimage* 2006; 31(3):968-
629 980
- 630 27. Benjamini Y, Hochberg Y: Controlling the False Discovery Rate: A Practical and Powerful
631 Approach to Multiple Testing. *J R Stat Soc Series B* 1995; 57(1):289-300
- 632 28. Nakagawa S, Cuthill IC: Effect size, confidence interval and statistical significance: a practical
633 guide for biologists. *Biol Rev Camb Philos Soc* 2007; 82(4):591-605
- 634 29. Spielberger CD, Gorsuch RL, Lushene PR, et al: Manual for the State-Trait Anxiety Inventory.
635 Palo Alto, CA, Consulting Psychologists Press, Inc, 1983.
- 636 30. Beck AT, Steer RA, Brown GK: Manual for the Beck Depression Inventory, 2nd ed. San Antonio,
637 TX, The Psychological Corporation, 1996.
- 638 31. Harrewijn A, Cardinale EM, Groenewold NA, et al: Cortical and subcortical brain structure in
639 generalized anxiety disorder: findings from 28 research sites in the ENIGMA-Anxiety Working
640 Group. 2021; 11(1):502
- 641 32. Groenewold NA, Bas-Hoogendam JM, Amod AR, et al: Volume of subcortical brain regions in
642 social anxiety disorder: mega-analytic results from 37 samples in the ENIGMA-Anxiety working
643 group. *Mol Psychiatry* accepted;
- 644 33. Boedhoe PSW, Schmaal L, Abe Y, et al: Cortical Abnormalities Associated With Pediatric and
645 Adult Obsessive-Compulsive Disorder: Findings From the ENIGMA Obsessive-Compulsive
646 Disorder Working Group. 2018; 175(5):453-462
- 647 34. Schmaal L, Hibar DP, Samann PG, et al: Cortical abnormalities in adults and adolescents with
648 major depression based on brain scans from 20 cohorts worldwide in the ENIGMA Major
649 Depressive Disorder Working Group. 2017; 22(6):900-909
- 650 35. Goodkind M, Eickhoff SB, Oathes DJ, et al: Identification of a common neurobiological substrate
651 for mental illness. *Jama Psychiat* 2015; 72(4):305-315
- 652 36. Caseras X, Mataix-Cols D, Trasovares MV, et al: Dynamics of brain responses to phobic-related
653 stimulation in specific phobia subtypes. *Eur J Neurosci* 2010; 32(8):1414-1422
- 654 37. Brinkmann L, Poller H, Herrmann MJ, et al: Initial and sustained brain responses to threat
655 anticipation in blood-injection-injury phobia. *Neuroimage Clin* 2017; 13:320-329
- 656 38. Michalowski JM, Matuszewski J, Drozdziel D, et al: Neural response patterns in spider, blood-
657 injection-injury and social fearful individuals: new insights from a simultaneous EEG/ECG-fMRI
658 study. *Brain Imaging Behav* 2017; 11(3):829-845
- 659 39. Strawn JR, Dominick KC, Patino LR, et al: Neurobiology of Pediatric Anxiety Disorders. *Curr*
660 *Behav Neurosci Rep* 2014; 1(3):154-160
- 661 40. Albor YC, Benjet C, Mendez E, et al: Persistence of Specific Phobia From Adolescence to Early
662 Adulthood: Longitudinal Follow-Up of the Mexican Adolescent Mental Health Survey. *J Clin*
663 *Psychiatry* 2017; 78(3):340-346
- 664 41. Cauda F, Nani A, Manuella J, et al: Brain structural alterations are distributed following
665 functional, anatomic and genetic connectivity. *Brain* 2018; 141(11):3211-3232
- 666 42. Zhou J, Gennatas ED, Kramer JH, et al: Predicting Regional Neurodegeneration from the Healthy
667 Brain Functional Connectome. *Neuron* 2012; 73:1216-1227

669
670

671

Figure legends

672 **FIGURE 1.** Significant differences between specific phobia subjects and healthy controls^a

673

674 ^aThe bar chart (left) provides effect sizes between groups of individuals with specific phobia and
675 healthy controls, error bars are standard errors. Positive effect sizes signify larger volume, surface
676 area and thickness in specific phobia compared to healthy controls. The graphical depiction (right)
677 shows the significant differences in the brain. Panel (A) shows subcortical volumes, panel (B) cortical
678 thickness and panel (C) cortical surface area.

679

680 **FIGURE 2.** Significant differences between animal phobia subjects and BII phobia subjects^a

681

682 ^aThe bar chart (above) provides effect sizes between groups of individuals with animal phobia and BII
683 phobia, error bars are standard errors. Positive effect sizes signify larger volume, surface area and
684 thickness in animal phobia subjects compared to BII phobia subjects. The graphical depiction (below)
685 shows the significant differences in cortical thickness between animal phobia and BII phobia
686 subjects.

687

688 **FIGURE 3.** Significant differences between animal phobia subjects and healthy controls and BII
689 phobia subjects and healthy controls^a

690

691 ^aThe bar chart (left) provides effect sizes between groups of individuals with animal phobia subjects
692 and healthy controls in green and between BII phobia subjects and healthy controls in orange. Error
693 bars are standard errors. Positive effect sizes signify larger volume, surface area and thickness in the
694 respective phobia subgroup compared to healthy controls. The graphical depiction (right) shows the
695 significant differences between animal phobia subjects and HC (upper right) and BII phobia subjects

696 and HC (lower right) in the brain. Panel (A) shows subcortical volumes, panel (B) cortical thickness

697 and panel (C) cortical surface area.

698

Table 1. Number of initial images, number of included images and reasons for exclusion by site.

Study	initial data				Freesurfer fail		quality control exclusion		Comorbidity		Other exclusion		
	initial datasets cortical	initial datasets subcortical	initial datasets covariates	initial datasets all complete	cortical	sub-cortical	cortical	sub-cortical	psychosis	bipolar	healthy controls with disorder	number of included images	percent of included images
Barcelona (38, 39)	52	52	52	52	0	0	0	0	0	0	0	52	100.0
BHRCS (40)	605	605	2511	596	0	0	0	0	0	2	96	498	83.6
BION-SP (41)	29	29	29	29	0	0	3	2	0	0	0	25	86.2
Bochum	18	18	18	18	1	2	0	0	0	0	0	15	83.3
COMIC (42)	12	12	12	12	0	0	0	0	0	0	0	12	100.0
Czuwaj (43)	46	46	48	46	0	0	0	0	0	0	10 ^a	36	78.3
Dresden CRC940C5	187	187	182	182	2	0	1	1	0	0	0	178	97.8
Dresden Phobia Subtypes (23)	95	95	126	95	0	0	0	0	0	0	6	89	93.7
Graz (30)	86	86	86	86	0	1	2	4	0	0	0	80	93.0
Graz II (29)	72	72	72	72	0	0	7	13	0	0	0	52	72.2
Greifswald (44)	44	44	45	44	0	0	0	0	0	0	0	44	100.0
Jena (45)	29	29	30	29	0	0	0	0	0	0	0	29	100.0
Marburg FOR2107 MR (46, 47)	532	532	532	532	0	0	0	1	0	0	0	531	99.8
Muenster Dental (48)	38	38	38	38	0	0	0	1	0	0	0	37	97.4

Muenster FOR2107 MS (49)	275	275	275	275	0	0	2	3	0	0	0	270	98.2
Muenster SFBTRR-58 C09 (50)	96	96	215	96	0	1	1	4	0	0	0	90	93.8
Muenster Spider (51)	507	507	507	507	3	1	0	1	0	0	0	502	99.0
PHOBIA EXPOSURE (52)	20	20	20	20	1	1	0	0	0	0	0	19	95.0
PNC (53, 54)	945	854	854	717	4	5	4	6	43	4	0	651	90.8
Protect-AD (55)	57	57	57	57	1	0	0	0	0	0	0	56	98.2
RepSpi (56)	38	38	38	38	0	0	0	0	0	0	0	38	100.0
SDAN (57, 58)	119	119	119	119	0	0	0	0	0	0	0	119	100.0
SHIP (59)	995	977	995	977	80	0	0	55	0	3	238	649	66.4
SMARTSCAN (60)	93	93	95	93	0	0	0	0	0	0	0	93	100.0
SPIN	14	14	14	14^b	0	2	0	0	0	0	0	0^b	0.0
SPIN NF (61)	19	19	19	19	11	11	0	1	0	0	0	7	36.8
Teneriffa (62)	77	77	78	77	0	0	0	4	0	0	0	73	94.8
Uppsala (63)	47	47	47	47	0	0	0	1	0	0	0	46	97.8
Vanderbilt (64, 65)	19	19	18	18	0	0	0	0	0	0	0	18	100.0
Wuerzburg SFBTRR-58 C09 (50)	87	87	87	87	0	0	0	0	0	0	0	87	100.0
Wuerzburg Spider (66)	36	36	36	36	4	5	3	3	0	0	0	28	77.8
Wuerzburg Spider II	25	25	25	25	2	1	0	0	0	0	0	23	92.0
Wuerzburg Spider III	16	16	0	0	0	0	0	0	0	0	0	0	0.0

Total	5 330	5 221	7 280	5 053	109	28	23	99	43	9	343	4 443	87.9
--------------	--------------	--------------	--------------	--------------	------------	-----------	-----------	-----------	-----------	----------	------------	--------------	-------------

initial datasets cortical / subcortical / covariate file: Datasets (subjects) were counted regardless of whether raw MRI data or the results of the Freesurfer preprocessing done on site were contributed.

^aIncludes a group of social phobia subjects without specific phobia that were thus not included in any group here.

^bNot considered further due to <10 initial specific phobia datasets.

BHRCS: Brazilian High Risk Cohort Study; BION-SP: Bender Institute of Neuroimaging; COMIC: COMIC Research / Leeds and York Partnership NHS Foundation Trust; Dresden CRC940C5: DFG Collaborative Research Centre 940, project C5; Marburg FOR2107 MR: DFG-Research Group 2107 Marburg site; Muenster FOR2107 MS: DFG-Research Group 2107 Muenster site; Muenster SFBTRR-58 C09: DFG Collaborative Research Centre Transregio 58, project C09, Muenster site; PNC: Philadelphia Neurodevelopmental Cohort; Protect-AD: Providing Tools for Effective Care and Treatment of Anxiety Disorders consortium, specific phobia sample; SDAN: Section on Development and Affective Neuroscience; SHIP: Study of Health in Pomerania; Wuerzburg SFBTRR-58 C09: DFG Collaborative Research Centre Transregio 58, project C09, Wuerzburg site.

699

700

701

702

703

704

705

706

Table 2. Sociodemographic characteristics of the sample used for the main analysis.

Study	specific phobia (formal diagnosis)	subtype																						
		all						specific phobia						healthy controls										
		n	sex		age		education		sex	age		education		sex	age		education							
total	% female	m	sd	range	m	sd	% female	m	sd	range	m	sd	% female	m	sd	range	m	sd	range	m	sd			
Barcelona	34 (34)	16	18	0	18	52	88.5	22.1	2.7	18-29	NA	NA	88.2	22.2	2.6	18-29	NA	NA	88.9	21.7	2.8	18-29	NA	NA
BHRCS	76 (28)	76	0	0	422	498	45.2	9.5	1.9	5-14	4.0	1.7	43.4	9.2	1.7	6-13	3.7	1.5	45.5	9.5	1.9	5-14	4.1	1.7
BION-SP	15 (15)	15	0	0	10	25	92.0	23.6	3.1	18-31	NA	NA	86.7	23.5	3.3	18-31	NA	NA	100.0	23.8	3.0	19-28	NA	NA
Bochum	15 (15)	0	15	0	0	15	46.7	39.3	10.7	27-60	NA	NA	46.7	39.3	10.7	27-60	NA	NA	NA	NA	NA	NA	NA	NA
COMIC	12 (12)	12	0	0	0	12	83.3	28.2	9.6	17-42	NA	NA	83.3	28.2	9.6	17-42	NA	NA	NA	NA	NA	NA	NA	NA
Czuwaj	25 (0)	12	13	0	11	36	77.8	23.0	4.2	19-38	NA	NA	92.0	23.0	4.8	19-38	NA	NA	45.5	23.2	2.3	20-27	NA	NA
Dresden CRC940C5	97 (97)	96	0	0	81	178	91.6	24.7	6.1	17-48	12.5	1.1	92.8	25.4	6.8	17-48	12.3	1.3	90.1	23.8	5.1	18-44	12.9	0.6
Dresden Phobia Subtypes	59 (22)	33	26	0	30	89	77.5	23.7	4.7	18-46	12.4	0.7	78.0	23.9	5.0	18-46	12.3	0.8	76.7	23.1	4.1	18-38	12.5	0.5
Graz	41 (41)	0	41	0	39	80	60.0	29.7	9.9	19-62	12.6	1.0	61.0	30.1	10.7	20-62	12.6	1.1	59.0	29.2	9.0	19-53	12.7	0.9
Graz II	25 (25)	0	25	0	27	55	55.8	29.6	10.2	20-56	NA	NA	56.0	33.6	11.5	23-56	NA	NA	55.6	25.9	7.1	20-48	NA	NA
Greifswald	20 (0)	20	0	0	24	44	100.0	22.3	3.0	18-29	13.0	0.0	100.0	21.9	2.9	18-28	13.0	0.0	100.0	22.7	3.1	19-29	13.0	0.0
Jena	14 (14)	14	0	0	15	29	100.0	24.8	6.1	19-49	NA	NA	100.0	24.4	4.1	21-35	NA	NA	100.0	25.2	7.7	19-49	NA	NA
Marburg FOR2107 MR	16 (16)	8	0	8	515	531	59.5	34.8	12.7	18-65	13.7	2.6	62.5	32.6	14.3	18-59	11.4	2.1	59.4	34.9	12.7	18-65	13.8	2.6

Muenster FOR2107 MS	27 (27)	5	3	19	243	270	64.8	29.4	11.2	18-65	14.3	2.3	74.1	36.1	13.5	19-64	14.8	2.7	63.8	28.6	10. 7	18-65	14.2	2.2
Muenster SFBTRR-58 C09	90 (90)	85	0	5	0	90	83.3	28.3	9.3	18-56	14.7	2.8	83.3	28.3	9.3	18-56	14.7	2.8	NA	NA	NA	NA	NA	NA
Muenster Dental	18 (0)	0	18	0	19	37	81.1	28.0	10.3	18-60	12.8	1.0	88.9	29.6	11.0	19-53	12.7	1.2	73.7	26.5	9.6	18-60	12.9	0.9
Muenster Spider	29 (29)	29	0	0	473	502	54.6	37.3	11.8	18-59	15.3	2.4	86.2	25.1	5.5	18-39	NA	NA	52.6	38.1	11. 7	18-59	15.3	2.4
PHOBIA EXPOSURE	19 (19)	19	0	0	0	19	100.0	23.3	3.1	19-29	NA	NA	100.0	23.3	3.1	19-29	NA	NA	NA	NA	NA	NA	NA	NA
PNC	319 (319)	0	0	319	332	650	56.7	14.6	3.8	8-23	7.8	3.6	64.9	14.3	3.6	8-21	7.4	3.4	48.6	14.9	3.9	8-23	8.2	3.8
Protect-AD	56 (56)	6	8	42	0	56	57.1	34.6	13.3	18-67	NA	NA	57.1	34.6	13.3	18-67	NA	NA	NA	NA	NA	NA	NA	NA
RepSpi	18 (0)	18	0	0	20	38	84.2	23.4	4.2	18-43	NA	NA	100.0	24.1	5.9	19-43	NA	NA	70.0	22.9	1.8	18-26	NA	NA
SDAN	47 (47)	0	0	47	72	119	61.3	13.1	2.9	8-18	7.2	2.9	66.0	11.9	2.9	8-18	6.3	2.9	58.3	13.7	2.8	8-18	7.9	2.8
SHIP	130 (125)	29	15	81	519	649	56.6	55.5	12.5	31-90	10.5	1.4	79.2	50.8	10.1	31-76	10.5	1.4	50.9	56.7	12. 8	31-90	10.5	1.4
SMARTSCAN	46 (46)	46	0	0	47	93	86.0	20.7	2.1	16-25	12.9	0.4	91.3	20.5	2.3	16-25	12.9	0.4	80.9	20.9	1.9	16-25	12.9	0.4
SPIN NF	7 (7)	7	0	0	0	7	100.0	21.7	2.4	19-26	14.7	0.8	100.0	21.7	2.4	19-26	14.7	0.8	NA	NA	NA	NA	NA	NA
Teneriffa	34 (34)	34	0	0	39	73	71.2	27.9	11.0	18-56	NA	NA	82.4	35.1	11.9	19-56	NA	NA	61.5	21.7	4.6	18-41	NA	NA
Uppsala	46 (0)	46	0	0	0	47	73.9	26.0	7.5	20-55	NA	NA	73.9	26.0	7.5	20-55	NA	NA	NA	NA	NA	NA	NA	NA
Vanderbilt	9 (9)	2	0	7	9	18	77.8	20.4	3.9	10-25	16.0	1.3	77.8	20.3	3.6	12-25	16.1	1.5	77.8	20.4	4.3	10-25	15.9	1.2
Wuerzburg SFBTRR-58 C09	87 (87)	87	0	0	0	87	85.1	28.6	8.5	18-60	12.0	1.5	85.1	28.6	8.5	18-60	12.0	1.5	NA	NA	NA	NA	NA	NA
Wuerzburg Spider	11 (0)	11	0	0	16	27	100.0	22.0	3.7	18-37	NA	NA	100.0	21.7	5.3	18-37	NA	NA	100.0	22.2	2.3	18-26	NA	NA

Wuerzburg Spider II	13 (0)	13	0	0	10	23	78.3	26.7	6.8	19-42	11.6	1.2	76.9	29.5	7.5	21-42	11.2	1.2	80.0	23.0	3.1	19-29	12.1	0.9
Total	1 452(1 213)	73 9	18 2	528	2 991	4 443	62.7	29.0	16.9	5-90	10.8	4.3	75.3	25.2	13.2	6-76	10.1	3.9	56.7	30.8	18. 1	5-90	11.1	4.4

BHRCS: Brazilian High Risk Cohort Study; BION-SP: Bender Institute of Neuroimaging; COMIC: COMIC Research / Leeds and York Partnership NHS Foundation Trust; Dresden CRC940C5: DFG Collaborative Research Centre 940, project C5; Marburg FOR2107 MR: DFG-Research Group 2107 Marburg site; Muenster FOR2107 MS: DFG-Research Group 2107 Muenster site; Muenster SFBTRR-58 C09: DFG Collaborative Research Centre Transregio 58, project C09, Muenster site; PNC: Philadelphia Neurodevelopmental Cohort; Protect-AD: Providing Tools for Effective Care and Treatment of Anxiety Disorders consortium, specific phobia sample; SDAN: Section on Development and Affective Neuroscience; SHIP: Study of Health in Pomerania; Wuerzburg SFBTRR-58 C09: DFG Collaborative Research Centre Transregio 58, project C09, Wuerzburg site.

707

708

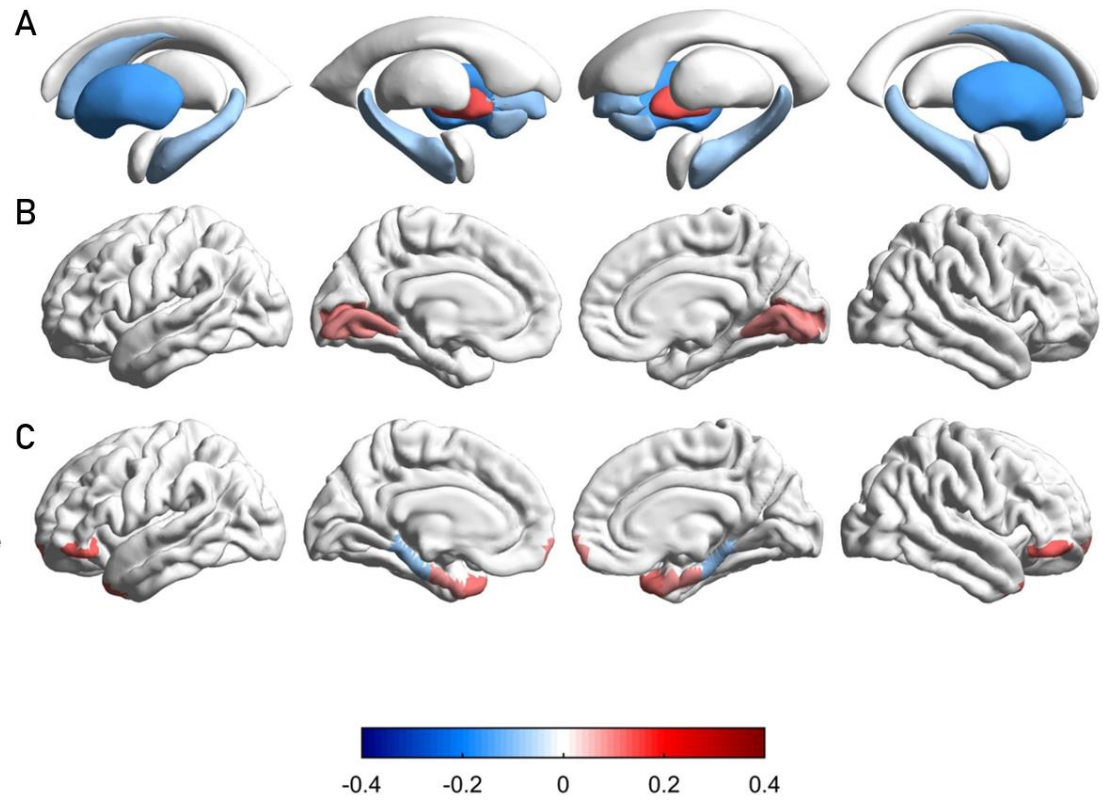
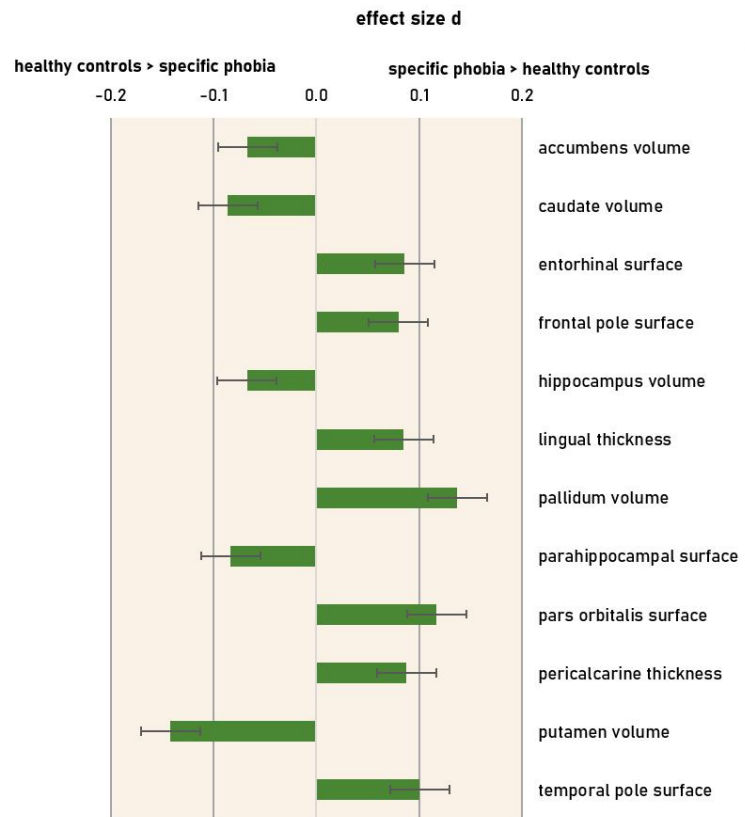
709

710

711

712

713



714

715

716

717

