ELECTRONIC SUPPLEMENTARY FILE (ESF)

Adverse childhood experiences are strongly associated with activation of M1 macrophage, T helper cell, neurotoxic and growth factor profiles and predict reoccurrence of illness and the phenome of affective disorders.

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ESF Table S1. The cytokines, chemokines and growth factors examined in the current study

Protein abbreviations	Gene Symbol	Protein name
IL-1β	IL1B	Interleukin-1β
IL-1RA	IL1RN	Interleukin-1 receptor antagonist
IL-2	IL2	Interleukin-2
IL-4	IL4	Interleukin-4
IL-5	IL5	Interleukin-5
IL-6	IL6	Interleukin-6
IL-7	IL7	Interleukin-7
CXCL8	CXCL8	C-X-C motif chemokine ligand 8 (IL-8)
IL-9	IL9	Interleukin-9
IL-10	IL10	Interleukin-10
IL-12	IL12	Interleukin-12
IL-13	IL13	Interleukin-13
IL-15	IL15	Interleukin-15
IL-17	IL17	Interleukin-17
CCL11	CCL11	Eotaxin
FGF2	FGF2	Fibroblast growth factor 2, Basic fibroblast growth factor
G-CSF	CSF3	Granulocyte Colony Stimulating Factor, Colony Stimulating Factor 3 (Granulocyte)
GM-CSF	CSF2	Granulocyte-macrophage colony-stimulating factor, Colony-stimulating factor 2
IFN-γ	IFNG	Interferon-γ
CXCL10	CXCL10	C-X-C motif chemokine ligand 8, Interferon gamma-induced protein 10 (IP10)
CCL2	CCL2	C-C Motif Chemokine Ligand 2 (MCP1)
CCL3 / MIP-1α	CCL3	Macrophage inflammatory protein-1 alpha, C-C Motif Chemokine Ligand 3
PDGF	PDGFA	Platelet Derived Growth Factor Subunit A
CCL4 / MIP-1β	CCL4	C-C Motif Chemokine Ligand 4, Macrophage Inflammatory Protein 1-Beta, Lymphocyte Activation Gene 1 Protein
CCL5 /RANTES	CCL5	C-C Motif Chemokine Ligand 5, Regulated Upon Activation, Normally T-Expressed, And Presumably Secreted
TNF-α	TNF	Tumor Necrosis Factor-Alpha
VEGF	VEGFA	Vascular Endothelial Growth Factor

Adapted from: Rachayon, M.; Jirakran, K; Sodsai, P; Klinchanhom, S.; Sughondhabirom, A.; Plaimas, K.; Suratanee, A.; Maes, M. Effects of cannabidiol on activated immune-inflammatory pathways in major depressive patients and healthy controls. Pharmaceuticals, 2022, in press.

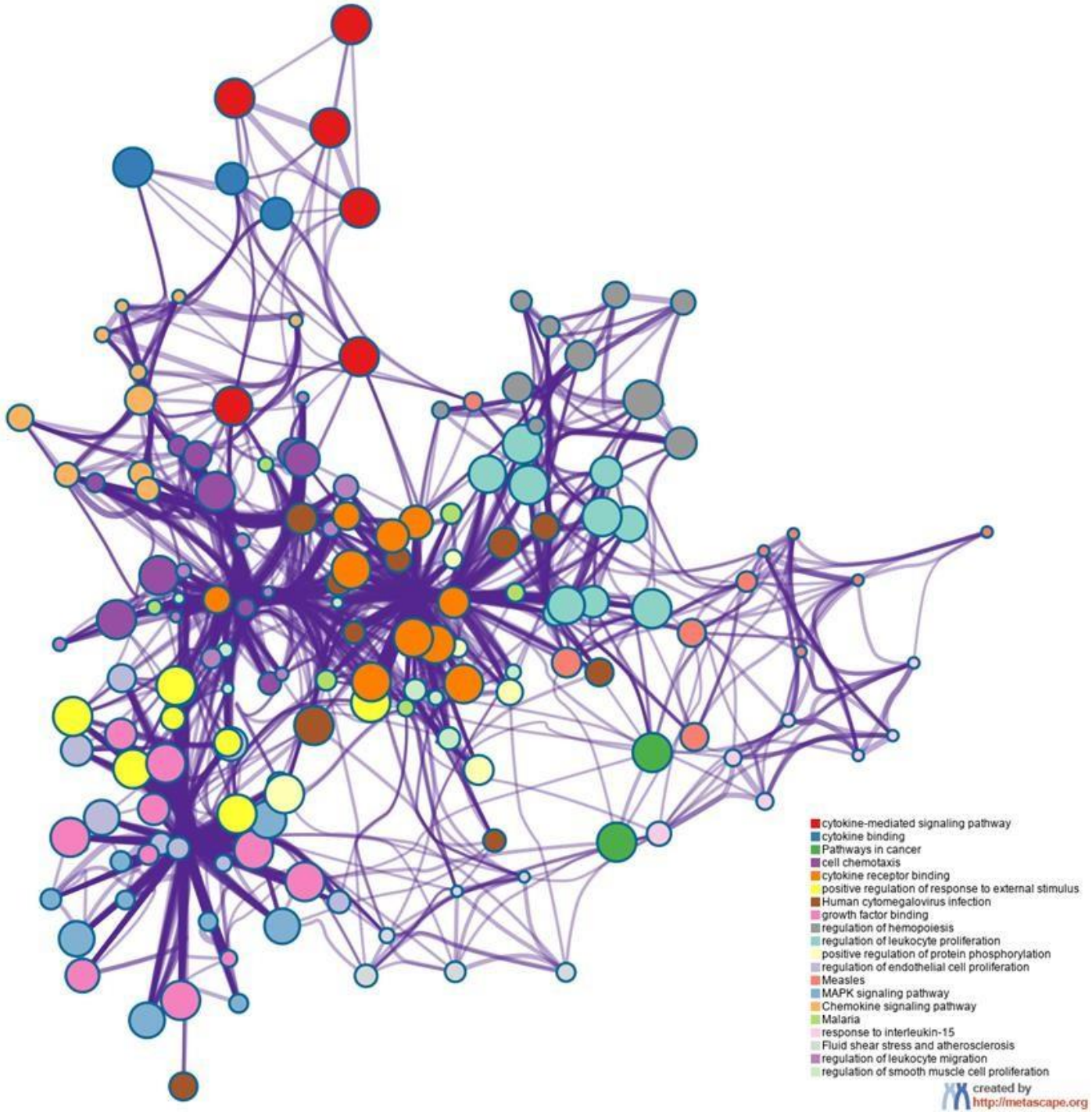
ESF Table S2. The 9 immune and growth factor profiles employed in this study

Immune Profile	Members
M1 macrophage	IL-1β, sIL-1RA, IL-6, TNF-α, CXCL8, CCL3
T helper-1	IL-2, IFN-γ, IL-12
T helper-2	IL-4, IL-5, IL-9, IL-13
T helper-17	IL-6, IL-17
IRS	IL-1β, IL-6, TNF-α, CXCL8, CCL3, IL-2, IFN-γ, IL-12, IL-17, IL-15, G-CSF, GM-CSF, CXCL10, CCL5, CCL2
CIRS	IL-4, IL-10, sIL-1RA
Neuroimmunotoxic	IL-1β, IL-6, TNF-α, CXCL8, CCL3, IL-2, IFN-γ, IL-12, IL-17, CXCL10, CCL11, CCL5, CCL2
Growth factors	FGF, PDGF, VEGF
T cell growth	IL4, IL9, IL12, GM-CSF, IL-15

These profiles were computed as z unit based composite scores

IRS: immune-inflammatory response system; CIRS: compensatory immunoregulatory system

Adapted from: Rachayon, M.; Jirakran, K; Sodsai, P; Klinchanhom, S.; Sughondhabirom, A.; Plaimas, K.; Suratanee, A.; Maes, M. Effects of cannabidiol on activated immune-inflammatory pathways in major depressive patients and healthy controls. Pharmaceuticals, 2022, in press.


cytokine-mediated signaling pathway
cytokine binding
Pathways in cancer
cell chemotaxis
cytokine receptor binding
positive regulation of response to external stimulus
Human cytomegalovirus infection
growth factor binding
regulation of hemopoiesis
regulation of leukocyte proliferation
positive regulation of protein phosphorylation
regulation of endothelial cell proliferation
Measles
MAPK signaling pathway
Chemokine signaling pathway
Malaria
response to interleukin-15
Fluid shear stress and atherosclerosis
regulation of leukocyte migration
regulation of smooth muscle cell proliferation
created by
http://metascape.org

ESF Figure S1. Enriched ontology term clusters in the protein-protein-interaction network of Adverse Childhood Experiences. Circular nodes represent the terms that are overrepresented and their size reflects the amount of input genes. Colors identify clusters of terms and the colored labels describe the terms employed to characterize the clusters. The thickness of the bundled edges represents the similarity score with a threshold of 0,3. The network is generated using MetaScape.

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Vessle- ediaediranspou

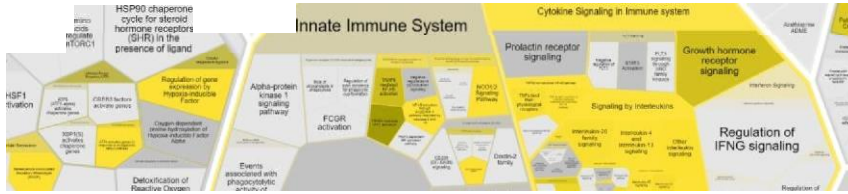
Response of

to heme

deficiency

Hems signaling

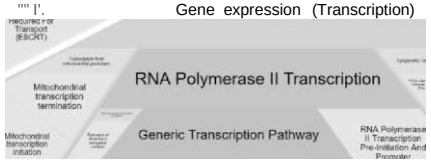
Immune System



Neutrophil degranulation

Advanced glycosylation endproduct

Gene expression (Transcription)



Disease

Cell Cycle

Sensory Perception

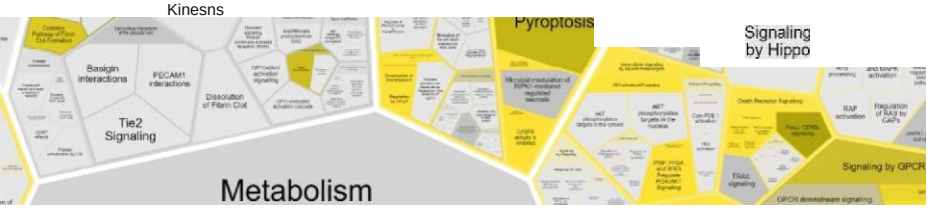
Events Events

Hemos ass

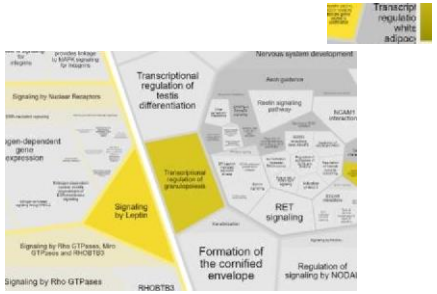
Programmed Ceel Death

Signal Transduction

Kinesns



Signaling by Hippo



C5F3 (G-CSF)

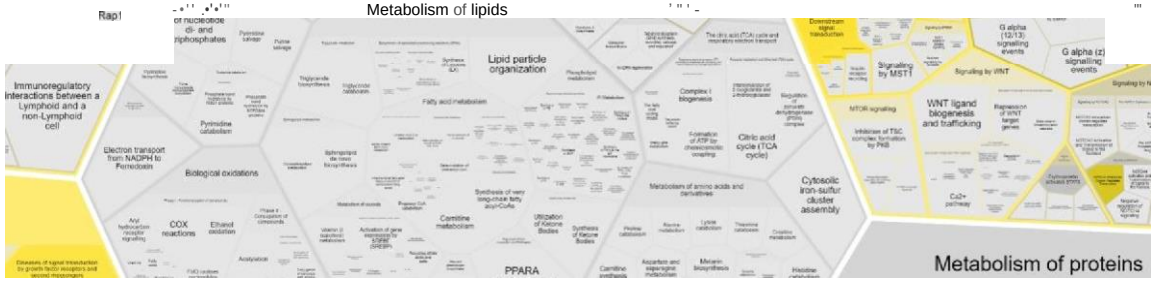
Pyrophosphale

ONA Repaii

Rap1

processing: ubiquitination & Proteasome degradation

Metabolism of lipids



expression

*ransool oT smal molecules

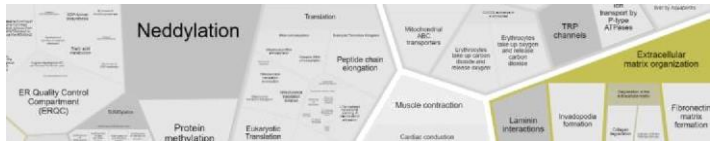
transport and events

Transferrin endocytosis and recycing

Processing of SMDT1

transport

ol NNA



Expression and translocation of olfactory receptors

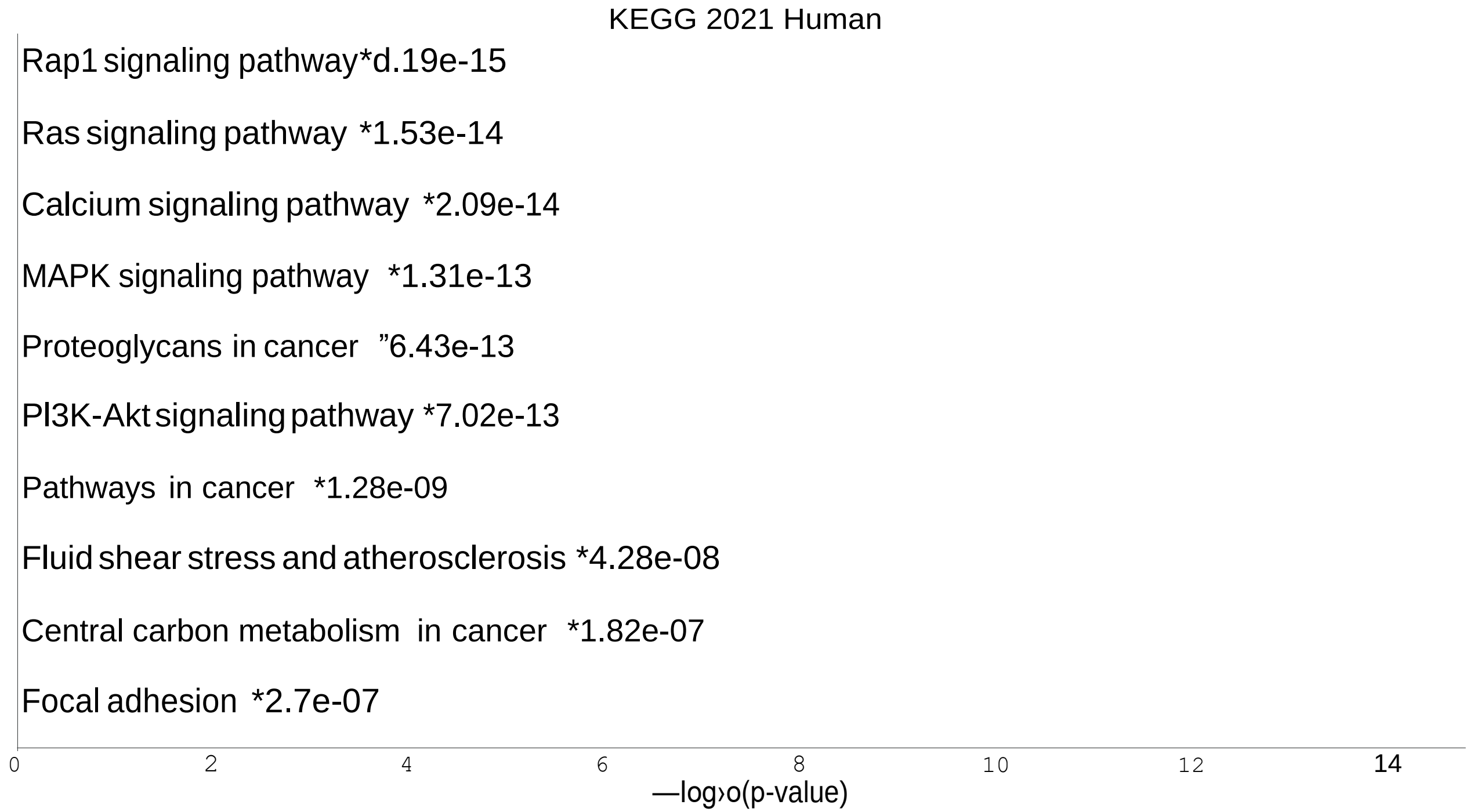
Amyloid fiber formation

Strived Muscle Contraction

Smoov, Muscle Contraction

Syndecan interactions

ESF Figure S2. Voronoi visualization of the hierachical pathways overrepresented in the protein-protein interaction network of the genes that are upregulated in subjects with Adverse Childhood Experiences. This figure was constructed employing the Reactome pathway analysis.


KEGG 2021 Human
Rap1 signaling pathway*d.19e-15
Ras signaling pathway *1.53e-14
Calcium signaling pathway *2.09e-14
MAPK signaling pathway *1.31e-13
Proteoglycans in cancer ”6.43e-13
PI3K-Akt signaling pathway *7.02e-13
Pathways in cancer *1.28e-09
Fluid shear stress and atherosclerosis *4.28e-08
Central carbon metabolism in cancer *1.82e-07
Focal adhesion *2.7e-07
0
2
4
6
8
10
12
14
—log›o(p-value)

ESF Figure S3. Bar graph made using Enrichr and visualized with Appyter showing the top 10 Kegg pathways which are overrepresented in the protein-protein network of the growth factor genes of Adverse Childhood Experiences.

WikiPathway 2021 Human

Focal Adhesion-Pl3K-Akt-mTOR-signaling pathway WP3932 *1.54e-15

Angiogenesis WP1539 *7.53e-15

Pl3K-Akt signaling pathway WP4I72 *4.88e-13

EGFR Tyrosine Kinase Inhibitor Resistance WP4806 *2.22e-11

Pathways Regulating Hippo Signaling WP4540 *5.72e-11

Hippo-Merlin Signaling Dysregulation WP4541 *1.97e-10

Ras signaling WP4223 ”2.51e-09

SARS-CoV-2 altering angiogenesis via NRP1 WP50s5 *3.41e-09

ESC Pluripotency Pathways WP3931 *1.72e-08

Regulation of Actin Cytoskeleton WP51 ”s.27e-08

0 2 4 6 8 10 1z 14

—log›o(p-value)

ESF Figure S4. Bar graph made using Enrichr and Appyter showing the top 10 Wikipathways which are overrepresented in the growth factor network of Adverse Childhood Experiences.