PhD Program in

Food and Human Health

38° Cycle

Evidence for Endocannabinoid-Like Compound Supplementation for Human Health: Palmitoylethanolamide as a Potential Treatment in Clinical High-Risk for Psychosis

PhD Candidate
Dr. Riccardo Bortoletto

Supervisor
Prof. Marco Colizzi

Year 2026

Abstract

This doctoral project investigated the biobehavioral potential of palmitoylethanolamide (PEA), an endocannabinoid-like lipid mediator, as a safe and mechanistically grounded intervention for early states of mental health vulnerability. The role of PEA as a homeostatic regulator of immune–glial activity and glutamatergic signaling is generating growing interest in its therapeutic applications across neuropsychiatric conditions characterized by inflammation-driven dysregulation. The research adopted a translational approach integrating systematic reviews and proof-of-concept studies. The first systematic review synthesized evidence from randomized controlled trials on PEA supplementation across clinical populations, highlighting consistent efficacy in pain management and emerging benefits for neuropsychiatric symptoms, while underscoring methodological heterogeneity and lack of mechanistic insight. The second systematic review focused on the biobehavioral role of PEA in psychosis across different stages of illness and clinical phenotypes. Building on this foundation, an open-label, single-arm proof-of-concept trial was conducted in individuals at clinical high-risk (CHR) for psychosis to assess the feasibility, tolerability, and biobehavioral correlates of ultramicronized PEA supplementation (600 mg/day, 12 weeks). Significant improvements were observed in attenuated psychotic symptoms and functional outcomes, with no treatment-emergent adverse effects. Exploratory analyses identified peripheral neuroimmune biomarkers as potential predictors of treatment responsiveness, suggesting that PEA may act as a homeostatic modulator in this population, most effective in individuals with reactive or pro-inflammatory biological profiles. Overall, this project provides preliminary evidence supporting PEA as a biologically informed and well-tolerated intervention for early and therapy-orphan mental health conditions, including the CHR state for psychosis. By integrating clinical and biological data, it contributes to the advancement of personalized, mechanism-based strategies within the framework of precision psychiatry.

Keywords: Palmitoylethanolamide (PEA); Endocannabinoid system; ALIAmides; Psychosis; Neuroinflammation.

Abstract

Questo progetto di dottorato ha indagato il potenziale biocomportamentale della palmitoiletanolamide (PEA), un mediatore lipidico endocannabinoide-simile, come intervento sicuro e meccanicisticamente fondato per stati di vulnerabilità precoce della salute mentale. Il ruolo della PEA come regolatore omeostatico dell'attività immuno-gliale e della trasmissione glutamatergica sta suscitando crescente interesse per le sue possibili applicazioni terapeutiche in condizioni neuropsichiatriche caratterizzate da disregolazione su base infiammatoria. Il progetto ha seguito un approccio traslazionale, combinando revisioni sistematiche e studi proof-of-concept. La prima revisione sistematica ha sintetizzato le evidenze provenienti da trial clinici randomizzati sull'integrazione di PEA in popolazioni cliniche, evidenziando una efficacia costante nella gestione del dolore e benefici emergenti su sintomi neuropsichiatrici, pur con significativa eterogeneità metodologica e scarsa chiarezza sui meccanismi d'azione. La seconda revisione sistematica ha approfondito il ruolo biocomportamentale della PEA nella psicosi, in diversi stadi e fenotipi di malattia. Su queste basi è stato condotto uno studio proof-of-concept aperto, non randomizzato e a singolo braccio, su individui in stato mentale a rischio (CHR) per psicosi, per valutare la fattibilità, la tollerabilità e i correlati biocomportamentali della supplementazione con PEA in forma ultramicronizzata (600 mg/die, 12 settimane). Sono stati osservati miglioramenti significativi nei sintomi psicotici attenuati e negli esiti funzionali, senza effetti avversi emergenti. Le analisi esplorative hanno identificato biomarcatori neuroimmuni periferici come potenziali predittori di risposta al trattamento, suggerendo che la PEA possa agire come modulatore omeostatico, risultando più efficace negli individui con profili biologici reattivi o pro-infiammatori. Complessivamente, il progetto fornisce evidenze preliminari a supporto della PEA come intervento biologicamente informato e ben tollerato per condizioni mentali precoci o orfane di terapia, inclusi lo stato mentale a rischio di psicosi. Integrando dati clinici e biologici, questo progetto contribuisce allo sviluppo di strategie personalizzate e basate sui meccanismi d'azione nell'ambito della psichiatria di precisione.

Parole chiave: Palmitoiletanolamide (PEA); Sistema endocannabinoide; ALIAmidi; Psicosi; Neuroinfiammazione.

CHAPTER I

General Introduction

Nowadays, understanding the biological foundations of mental health increasingly requires integrating insights from body-brain signaling, particularly those arising from nutrition, neuroinflammation, and neuroendocrine regulation.

This chapter provides the scientific background of the present work, outlining the relevance of dietary supplementation in mental health, the homeostatic role of the endocannabinoid (eCB) system, and the rationale for investigating the dietary supplement palmitoylethanolamide (PEA) as a potential intervention in mental health vulnerability conditions, such as the clinical high-risk (CHR) state for psychosis.

1. Why discuss dietary supplementation in mental health?

The scientific debate surrounding the importance of primary health prevention has increasingly acknowledged the pivotal role of nutrition in both general wellbeing and mental health. While benefits of a balanced diet for maintaining physical health are well established, particularly in reducing the risk of non-communicable diseases and premature mortality [1, 2], recent years have seen a surge of research exploring the effects of dietary patterns on mental functioning.

Diet and nutrition have clearly emerged not only as key determinants of psychological wellbeing, but also as modifiable targets for prevention of major mental health disorders, potentially contributing to reduced psychotropic medication use and fewer hospitalizations in psychiatric settings [3, 4].

Accordingly, the evidence-based use of selected dietary supplements (i.e., nutraceuticals) has markedly increased, aiming to compensate for nutrient deficiencies, suboptimal intake, limited bioavailability, and individual differences in absorption or metabolism. In mental health, despite often inconclusive findings, nutraceuticals are increasingly regarded as either stand-alone or adjunctive interventions to enhance the efficacy of psychotropic medications [3-5]. Overall, while some nutrients have shown efficacy under specific conditions, further research is needed to clarify their mechanisms and assess effects in targeted populations, adopting biomarker-guided approaches to optimize personalized care [5].

Particularly, considering the well-documented association between many psychiatric disorders or *sickness behavior* manifestations (e.g., fatigue, social withdrawal, anhedonia) and elevated levels of central and peripheral biomarkers of oxidative stress and inflammation [6-10], as well as evidence linking the efficacy of both pharmacological and lifestyle interventions to changes in these biomarkers [11, 12], the antioxidant and anti-inflammatory properties of certain dietary supplements (e.g., N-acetylcysteine, omega-3 fish oils) indicated promise for their use in the treatment of neuropsychiatric disturbances [5, 13-16]. Dietary supplementation is therefore no longer regarded merely as a source of essential nutrients, but as a key modulator of processes that influence brain

function, including systemic inflammation and lipid metabolism, shaping vulnerability or resilience to stress and psychiatric disorders.

Within this biological framework, the eCB system, conceptualized as the broader endocannabinoidome (eCBome), emerges as a central regulator of immune, metabolic, and neural homeostasis [17], attracting growing interest for its pharmacological modulation in contemporary medicine.

2. The endocannabinoid system and palmitoylethanolamide: a homeostatic framework

Beyond the major eCBs anandamide (AEA) and 2-arachidonoylglycerol (2-AG), a family of structurally related lipid mediators known as eCB-like compounds has been identified. Among these, the N-acylethanolamines (NAEs) PEA and oleoylethanolamide (OEA), as well as the monoacylglycerol (MAG) 2-oleoylglycerol (2-OG), have been extensively studied for their analgesic, anti-inflammatory, and anorexigenic properties [18-21]. Although they display limited affinity for canonical CB1 and CB2 cannabinoid receptors, these molecules interact with non-cannabinoid targets, thereby contributing to the broader regulatory role of the eCB system in maintaining homeostasis [17, 18].

PEA is a naturally occurring fatty acid amide first isolated from egg yolk and found in common foods. Endogenously, it is synthesized by multiple cell types in response to tissue injury or metabolic stress, as part of the Autacoid Local Injury Antagonists (ALIAmides) family. Through a multifaceted pharmacodynamics, further detailed in the following Chapters, PEA regulates mast-cell degranulation, restrains microglial hyperactivation, and mitigates glutamate-induced excitotoxicity, eventually promoting neuroinflammation balance and synaptic plasticity [19, 22-27].

Consistent with these mechanisms, PEA has gained increasing attention as a nutraceutical compound with therapeutic potential across neuropsychiatric conditions. Preclinical and clinical findings indicate benefits on pain, behavior, seizures, as well as on cognitive and adaptive functioning [28-30]. Given its endogenous nature, favorable safety, and excellent tolerability, PEA represents a biologically grounded and acceptable candidate for preventive or adjunctive interventions in early-stage or subthreshold mental health conditions, in which conventional psychotropic medications may be less suitable or lack consistent efficacy, as in the case of the CHR state for psychosis [31]. This rationale is further supported by evidence indicating that immune–glutamatergic dysregulation is an early and potentially reversible biological feature of the CHR state, and that modulation of the eCB system plays a key role in regulating these processes, thereby positioning PEA as a plausible upstream, homeostatic intervention [32, 33].

3. The clinical high-risk state for psychosis: from prodrome to preventive neuroimmune intervention

Over the past three decades, the construct of CHR state for psychosis has evolved from the earlier notion of "prodrome" as a deterministic precursor of schizophrenia toward a multidimensional and transdiagnostic model of risk [34-36]. This paradigm shift has reframed CHR and the related at-risk mental state (ARMS) as heterogeneous and dynamic syndromes [36, 37], rather than fixed pre-psychotic stages. They are characterized by subthreshold positive psychotic symptoms (e.g., mild perceptual disturbances, delusional ideas) accompanied by distress, impaired role and social functioning, reduced quality of life, and non-psychotic features such as anxiety, depression, and cognitive deficits [38-41]. The epidemiological prevalence of the CHR state is estimated at approximately 1.7% in the general population and slightly below 20% among individuals seeking mental health care [42]. According to recent meta-analysis, outcomes among CHR individuals follow diverse trajectories: while transition occurs in about 20% of cases at two years and 35% at ten years [41], many other cases remit or remain clinically stable over time, reflecting the intrinsic complexity of psychosis vulnerability [35]. This variability underscores the need for personalized early interventions and frames the CHR state as a therapy-orphan yet critical window of opportunity for preventive treatment. Current therapeutic options remain limited in efficacy and safety, calling for low-risk, mechanistic-based strategies that address both symptoms and underlying biology [41, 43, 44]. From a biological standpoint, compelling evidence implicates early neuroinflammatory processes and microglial dysregulation in the pathophysiology of CHR state and first-episode psychosis (FEP), alongside altered cytokine profiles, and elevated peripheral inflammatory markers [32, 33, 45-53]. These abnormalities converge on glutamatergic dysfunction and excitotoxicity, ultimately leading to disrupted dopaminergic firing and the clinical manifestation of overt psychosis. As a result, it is worth exploring whether interventions targeting immune-glutamatergic pathways, including PEA supplementation, can mitigate early symptom expression and reduce transition risk in this population [22, 26].

4. Aims of the doctoral project

This doctoral project explores the potential of PEA as a safe and biologically informed intervention for mental health vulnerability states where current treatment options are limited. The work will combine systematic reviews and empirical investigation to connect clinical observations with their biological underpinnings. Specifically, the project aims to: (i) review and synthesize randomized controlled evidence on the therapeutic use of PEA across all medical and neuropsychiatric conditions, outlining its transdiagnostic potential and current research gaps; (ii) explore PEA's biobehavioral role

across different stages and phenotypes of psychosis, combining insights from preclinical and clinical research; (iii) assess the correlates of PEA supplementation in individuals at CHR for psychosis through an investigator-initiated study, integrating clinical, functional, and biological measures to identify predictors of treatment responsiveness and generating hypotheses regarding potential disease-modifying pathways.

References

1. Swinburn, B.A., et al., *The Global Syndemic of Obesity, Undernutrition, and Climate Change: The Lancet Commission report.* Lancet, 2019. **393**(10173): p. 791-846.
2. *Health effects of dietary risks in 195 countries, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017.* Lancet, 2019. **393**(10184): p. 1958-1972.
3. Sarris, J., et al., *Nutritional medicine as mainstream in psychiatry.* Lancet Psychiatry, 2015. **2**(3): p. 271-4.
4. Sarris, J., et al., *International Society for Nutritional Psychiatry Research consensus position statement: nutritional medicine in modern psychiatry.* World Psychiatry, 2015. **14**(3): p. 370-1.
5. Firth, J., et al., *The efficacy and safety of nutrient supplements in the treatment of mental disorders: a meta-review of meta-analyses of randomized controlled trials.* World Psychiatry, 2019. **18**(3): p. 308-324.
6. Berk, M., et al., *So depression is an inflammatory disease, but where does the inflammation come from?* BMC Med, 2013. **11**: p. 200.
7. Miller, B.J., et al., *Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects.* Biol Psychiatry, 2011. **70**(7): p. 663-71.
8. Berk, M., et al., *Pathways underlying neuroprogression in bipolar disorder: focus on inflammation, oxidative stress and neurotrophic factors.* Neurosci Biobehav Rev, 2011. **35**(3): p. 804-17.
9. Dantzer, R., et al., *From inflammation to sickness and depression: when the immune system subjugates the brain.* Nature Reviews Neuroscience, 2008. **9**(1): p. 46-56.
10. Miller, A.H. and C.L. Raison, *The role of inflammation in depression: from evolutionary imperative to modern treatment target.* Nat Rev Immunol, 2016. **16**(1): p. 22-34.
11. Köhler, C.A., et al., *Peripheral Alterations in Cytokine and Chemokine Levels After Antidepressant Drug Treatment for Major Depressive Disorder: Systematic Review and Meta-Analysis.* Mol Neurobiol, 2018. **55**(5): p. 4195-4206.
12. Schuch, F.B., et al., *Neurobiological effects of exercise on major depressive disorder: A systematic review.* Neurosci Biobehav Rev, 2016. **61**: p. 1-11.
13. Dodd, S., et al., *N-acetylcysteine for antioxidant therapy: pharmacology and clinical utility.* Expert Opin Biol Ther, 2008. **8**(12): p. 1955-62.
14. Simopoulos, A.P., *Omega-3 fatty acids in inflammation and autoimmune diseases.* J Am Coll Nutr, 2002. **21**(6): p. 495-505.

15. Huang, H., et al., *Targeting Neuroinflammation in Schizophrenia: A comprehensive review of mechanisms and pharmacological interventions.* International Immunopharmacology, 2025. **159**: p. 114910.
16. Devoe, D.J., A. Peterson, and J. Addington, *Negative Symptom Interventions in Youth at Risk of Psychosis: A Systematic Review and Network Meta-analysis.* Schizophr Bull, 2018. **44**(4): p. 807-823.
17. Arturo, I.F. and P. Fabiana, *Endocannabinoidome*, in *eLS*. p. 1-10.
18. Kleberg, K., H.A. Hassing, and H.S. Hansen, *Classical endocannabinoid-like compounds and their regulation by nutrients.* Biofactors, 2014. **40**(4): p. 363-72.
19. Clayton, P., et al., *Palmitoylethanolamide: A Natural Compound for Health Management.* Int J Mol Sci, 2021. **22**(10).
20. Caillon, A., et al., *The OEA effect on food intake is independent from the presence of PPARα in the intestine and the nodose ganglion, while the impact of OEA on energy expenditure requires the presence of PPARα in mice.* Metabolism, 2018. **87**: p. 13-17.
21. Vaughn, L.K., et al., *Endocannabinoid signalling: has it got rhythm?* Br J Pharmacol, 2010. **160**(3): p. 530-43.
22. Petrosino, S. and V. Di Marzo, *The pharmacology of palmitoylethanolamide and first data on the therapeutic efficacy of some of its new formulations.* Br J Pharmacol, 2017. **174**(11): p. 1349-1365.
23. Petrosino, S. and A. Schiano Moriello, *Palmitoylethanolamide: A Nutritional Approach to Keep Neuroinflammation within Physiological Boundaries-A Systematic Review.* Int J Mol Sci, 2020. **21**(24).
24. Petrosino, S., et al., *Palmitoylethanolamide counteracts substance P-induced mast cell activation in vitro by stimulating diacylglycerol lipase activity.* J Neuroinflammation, 2019. **16**(1): p. 274.
25. Petrosino, S., et al., *Oral Ultramicronized Palmitoylethanolamide: Plasma and Tissue Levels and Spinal Anti-hyperalgesic Effect.* Front Pharmacol, 2018. **9**: p. 249.
26. Petrosino, S., et al., *The anti-inflammatory mediator palmitoylethanolamide enhances the levels of 2-arachidonoyl-glycerol and potentiates its actions at TRPV1 cation channels.* Br J Pharmacol, 2016. **173**(7): p. 1154-62.
27. Clayton, P., et al., *Palmitoylethanolamide: A Potential Alternative to Cannabidiol.* J Diet Suppl, 2021: p. 1-26.

28. Colizzi, M., et al., *Therapeutic effect of palmitoylethanolamide in cognitive decline: A systematic review and preliminary meta-analysis of preclinical and clinical evidence.* Frontiers in Psychiatry, 2022. **13**.
29. Colizzi, M., et al., *Palmitoylethanolamide and Its Biobehavioral Correlates in Autism Spectrum Disorder: A Systematic Review of Human and Animal Evidence.* Nutrients, 2021. **13**(4).
30. Bortoletto, R., et al., *Is It Time to Test the Antiseizure Potential of Palmitoylethanolamide in Human Studies? A Systematic Review of Preclinical Evidence.* Brain Sciences, 2022. **12**(1).
31. Yung, A.R., et al., *Mapping the onset of psychosis: the Comprehensive Assessment of At-Risk Mental States.* Aust N Z J Psychiatry, 2005. **39**(11-12): p. 964-71.
32. Basavaraju, R., et al., *Hippocampal Glutamate and Positive Symptom Severity in Clinical High Risk for Psychosis.* JAMA Psychiatry, 2022. **79**(2): p. 178-179.
33. Bojesen, K.B., et al., *Cerebral glutamate levels over two years in initially antipsychotic-naïve first-episode patients with psychosis are related to clinical symptoms and cognition.* Molecular Psychiatry, 2025.
34. Colizzi, M., A. Lasalvia, and M. Ruggeri, *Prevention and early intervention in youth mental health: is it time for a multidisciplinary and trans-diagnostic model for care?* Int J Ment Health Syst, 2020. **14**: p. 23.
35. McGorry, P.D. and C. Mei, *Ultra-high-risk paradigm: lessons learnt and new directions.* Evid Based Ment Health, 2018. **21**(4): p. 131-133.
36. Association, A.P., *Diagnostic and Statistical Manual of Mental Disorders*. 5th ed., text rev. ed. 2022: American Psychiatric Publishing.
37. American Psychiatric Association, *Diagnostic and Statistical Manual of Mental Disorders*. 5th ed. 2013, Washington, DC.
38. Devoe, D.J., et al., *Negative Symptoms and Functioning in Youth at Risk of Psychosis: A Systematic Review and Meta-analysis.* Harv Rev Psychiatry, 2020. **28**(6): p. 341-355.
39. Carrión, R.E., et al., *Prediction of Functional Outcome in Individuals at Clinical High Risk for Psychosis.* JAMA Psychiatry, 2013. **70**(11): p. 1133-1142.
40. Fusar-Poli, P., et al., *Prevention of Psychosis: Advances in Detection, Prognosis, and Intervention.* JAMA Psychiatry, 2020. **77**(7): p. 755-765.
41. Minichino, A., et al., *Preventing psychosis in people at clinical high risk: an updated meta-analysis by the World Psychiatric Association Preventive Psychiatry section.* Mol Psychiatry, 2025. **30**(6): p. 2773-2782.

42. Salazar de Pablo, G., et al., *Prevalence of Individuals at Clinical High-Risk of Psychosis in the General Population and Clinical Samples: Systematic Review and Meta-Analysis.* Brain Sci, 2021. **11**(11).
43. Davies, C., et al., *Lack of evidence to favor specific preventive interventions in psychosis: a network meta-analysis.* World Psychiatry, 2018. **17**(2): p. 196-209.
44. Davies, C., et al., *Efficacy and Acceptability of Interventions for Attenuated Positive Psychotic Symptoms in Individuals at Clinical High Risk of Psychosis: A Network Meta-Analysis.* Front Psychiatry, 2018. **9**: p. 187.
45. Day, F.L., et al., *Blunted cortisol awakening response in people at ultra high risk of developing psychosis.* Schizophr Res, 2014. **158**(1-3): p. 25-31.
46. Misiak, B., et al., *Immune-inflammatory markers and psychosis risk: A systematic review and meta-analysis.* Psychoneuroendocrinology, 2021. **127**: p. 105200.
47. Mondelli, V., et al., *Serum immune markers and transition to psychosis in individuals at clinical high risk.* Brain Behav Immun, 2023. **110**: p. 290-296.
48. Nisha Aji, K., et al., *Interaction between peripheral and central immune markers in clinical high risk for psychosis.* Brain Behav Immun Health, 2023. **30**: p. 100636.
49. Park, S. and B.J. Miller, *Meta-analysis of cytokine and C-reactive protein levels in high-risk psychosis.* Schizophr Res, 2020. **226**: p. 5-12.
50. Zhang, T., et al., *Changes in inflammatory balance correlates with conversion to psychosis among individuals at clinical high-risk: A prospective cohort study.* Psychiatry Res, 2022. **318**: p. 114938.
51. Berger, M., et al., *Cortisol awakening response in patients with psychosis: Systematic review and meta-analysis.* Neurosci Biobehav Rev, 2016. **68**: p. 157-166.
52. Taylor, J.H., et al., *Immune and oxidative stress biomarkers in pediatric psychosis and psychosis-risk: Meta-analyses and systematic review.* Brain Behav Immun, 2024. **117**: p. 1-11.
53. Upthegrove, R., N. Manzanares-Teson, and N.M. Barnes, *Cytokine function in medication-naive first episode psychosis: a systematic review and meta-analysis.* Schizophr Res, 2014. **155**(1-3): p. 101-8.

CHAPTER II

Palmitoylethanolamide Supplementation for Human Health: A State-Of-The-Art Systematic Review of Randomized Controlled Trials in Patient Populations

Bortoletto R, Comacchio C, Garzitto M, Piscitelli F, Balestrieri M, Colizzi M. Palmitoylethanolamide supplementation for human health: A state-of-the-art systematic review of Randomized Controlled Trials in patient populations. Brain Behav Immun Health. 2024 Dec 23;43:100927. doi: 10.1016/j.bbih.2024.100927. PMID: 39839988; PMCID: PMC11745966.

Note: Supplementary material associated with this publication is provided in the Appendix section of this thesis.

Abstract

Interest in preventative dietary interventions for human health has increasingly focused on the endocannabinoid (eCB)-like compound palmitoylethanolamide (PEA), a bioactive lipid mediator with anti-inflammatory, analgesic, and neuroprotective properties. This Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020-compliant systematic review aimed at collecting and comprehensively discussing all available data from Randomized Controlled Trials (RCTs) evaluating the efficacy and tolerability of PEA supplementation across human illnesses in patient populations. Overall, 48 eligible ouputs from 47 RCTs were extracted, covering neuropsychiatric (*n*=15), neurological (*n*=17), somatic (*n*=13), and visceral (*n*=11) disturbances, as well as PEA effects on blood/plasma or other tissue biomarkers (*n*=10). The strongest evidence emerged from RCTs exploring PEA impact on pain management and measures of general wellbeing, especially in its ultramicronized/micronized or cold-water dispersible formulations, showing good tolerability compared to controls. Also, alongside symptom improvement, PEA demonstrated to modulate biomarkers early altered in the initial phases of an illness or contributing to its progression, suggesting a disease-modifying potential. This systematic review provided a comprehensive overview of the therapeutic potential of PEA across RCTs, highlighting its versatility either as monotherapy or add-on treatment for various clinical conditions.

Keywords

Peroxisome Proliferator-Activated Receptor α; Glutamate; Neuroinflammation; Nutraceutical; Endocannabinoidome; Cannabidiol

1. Introduction

Recent years have seen increasing research and social awareness around healthy nutrition and dietary habits as key factors in primary prevention [1, 2]. This has contributed to promote public demand for nutraceuticals and dietary supplements as all-natural products, as well as their use in routine clinical practice, either as symptomatic treatments or as strategies with potential preventive properties especially targeting inflammatory processes [3-5]. Indeed, while inflammation is a natural response to remove harmful stimuli and promote tissue repair, an overly intense or prolonged inflammatory response can damage host tissues, cause organ dysfunction, and finally contribute to the medical burden in high-income countries [6, 7]. Concurrently, the search for novel pharmaceuticals, aimed at halting the progression of illnesses by intervening over their underlying mechanisms, has drawn growing attention to the endocannabinoid (eCB) system modulation, particularly due to its role in maintaining homeostasis during disease conditions as part of the enlarged family of lipid mediators known as the "endocannabinoidome" (eCBome) [8, 9]. In addition to the two main eCBs, anandamide (AEA) and 2-arachidonoylglycerol (2-AG), which directly interact with cannabinoid receptors [i.e., type-1 (CB1), type-2 (CB2)], "eCB-like compounds" including the N-acylethanolamines (NAEs) palmitoylethanolamide (PEA) and oleoylethanolamide (OEA), and the monoacylglycerol (MAG) 2-oleoylglycerol (2-OG), have been extensively investigated for their analgesic and anorexigenic properties, as well as for their effect on lipid metabolism [10-16]. Undoubtedly, among eCB-like compounds, PEA use as a dietary supplement is the most widely studied across several clinical conditions, thanks to its analgesic, anti-neuroinflammatory, and neuroprotective features [17-20]. As a naturally occurring lipid, PEA is produced by different cell types to restore disrupted homodynamic balance in response to actual or potential damage, exerting its effects in multiple ways: (i) directly activating the Peroxisome Proliferator Activated Receptor-α (PPAR-α) and G Protein-coupled Receptor 55 (GPR55), subsequently activating the Transient Receptor Potential Vanilloid 1 (TRPV1) channels and increasing CB2 expression [21-23]; (ii) allosterically modulating the TRPV1, that interacts with CB1 and CB2 receptors [21, 24]; (iii) inhibiting the Fatty-Acid Amide Hydrolase (FAAH) and stimulating the Diacylglycerol Lipase (DAGL), thus increasing the endogenous availability of AEA and 2-AG and their actions at CB1, CB2 and TRPV1 receptors [25, 26]; (4) down-regulating mast cell and microglia activation via an "Autacoid Local Inflammation Antagonism" (ALIA) effect [27, 28]. According to its pharmacodynamics, PEA is partially seen as the endogenous equivalent of the phytocannabinoid cannabidiol (CBD) [29], which has been thoroughly studied for its therapeutic potential in multiple neuropsychiatric disorders, chronic pain, and gastrointestinal motility issues, although being not devoid of adverse events (AEs) and raising increasing concern about possible interactions with other medications [30-34]. Alongside the safe and tolerable profile

of exogenous PEA supplementation, the above considerations make it an advantageous alternative to CBD in the long-term treatment of both clinical and general populations, particularly in its most bioavailable formulations [19, 29, 35-37].

1.1. Objectives

The anti-inflammatory properties of PEA may represent a promising therapeutic mechanism underlying its clinical utility across several human illness conditions, as both a symptomatic agent and a disease-modifying drug. This systematic review aims at collecting and discussing all available data generated from Randomized Controlled Trials (RCTs) exploring PEA efficacy and tolerability across all human illness conditions in clinical populations.

2. Materials and methods

2.1. Inclusion and exclusion criteria

All findings emerging from RCTs regarding the effects of PEA supplementation for human diseases in clinical populations were gathered and systematically analyzed according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines [38]. Since this systematic review was intended at collecting the highest level of evidence-based clinical data on PEA supplementation versus comparator treatments, only RCT designs were considered for systematic data extraction, due to their intrinsically lower susceptibility to biases [39]. Patient populations suffering from all illness conditions studied through RCTs with PEA, evaluated using appropriate clinical assessments or assessment scales of targeted symptoms, of any age, ethnicity, and gender, were included. Exclusion criteria were as follows: (i) non-RCTs in which PEA was the intervention of interest; (ii) RCTs in which PEA was not the intervention of interest; (iii) RCTs in which PEA was administered to healthy populations; (iv) case reports, case series, letters to the editor, correspondences, and commentaries; (v) cross-sectional studies, case-control studies, and cohort studies; (vi) reviews, systematic reviews, and meta-analyses.

2.2. Search strategy

A literature search was conducted across both electronic publication repositories (PubMed, Scopus, and Web of Science) and clinical trial registries [ClinicalTrials.gov, World Health Organization (WHO) International Clinical Trials Registry Platform (ICTRP)]. The first search encompassed all English-written original results of eligible studies published up to May 3rd, 2023, without any restrictions in terms of publication date, study duration, and follow-up. A second, more updated search was performed on August 26th, 2024, following the same methodology. To be as much inclusive as

possible, the search was divided as follows: (i) clinical trial registries ClinicalTrials.gov and ICTRP were queried using the search terms “palmitoylethanolamide” and “palmidrol”; (ii) PubMed was queried using the search string “(palmitoylethanolamide OR palmitylethanolamide OR N-2-hydroxyethyl-hexadecanamide OR N-2-hydroxyethyl-palmitate OR N-palmitoylethanolamine OR palmidrol OR impulsin OR um-pea)” and applying the ‘article type’ filters ‘Clinical Trial’ and ‘Randomized Controlled Trial’; (iii) publication repositories Pubmed, Web of Science, and Scopus were queried using a combination of broad-meaning terms describing and/or concerning intervention “(palmitoylethanolamide OR palmitylethanolamide OR N-2-hydroxyethyl-hexadecanamide OR N-2-hydroxyethyl-palmitate OR N-palmitoylethanolamine OR palmidrol OR impulsin OR um-pea)”, study type “(prospective OR randomised OR randomized OR trial OR observational OR rct OR (randomized AND controlled AND trial))”, and study population “(human OR female OR male OR proband OR patient* OR volunteer* OR healthy OR adult OR child)”.

2.3. Data extraction, screening, and risk of bias assessment

Data extraction and screening were conducted by using a web-based systematic review management software (Covidence systematic review software, Veritas Health Innovation, Melbourne, Australia).

Quality of studies assessment was conducted according to the Johanna Briggs Institute (JBI) critical appraisal tool for RCTs [40]. The JBI tool for RCTs consists of a 13-item scale exploring the internal validity (e.g., randomized allocation, level of concealment), similarity of participants and treatments between compared groups, the reliability of outcome measures, and the appropriateness of statistical analysis. Each item was parsed using four evaluation answers: “Yes”, “No”, “Unclear”, and “Not Applicable”. Affirmative answers were summarized as a score from 0 to 13. RCTs with a score equal or lower than 5 were classified as having a poor quality, those with a score between 6 and 7 as having a fair quality, and those with a score equal or higher than 8 as having a good quality [40].

All these steps were performed by two researchers (R.B. and C.C.), independently of each other.

In the instances of conflicting opinions regarding papers' inclusion or quality assessment, a consensus was reached through discussion involving a third senior reviewer (M.C.).

The full study protocol (PROSPERO 2023 CRD42023423617) is available at: https://www.crd.york.ac.uk/prospero/display_record.php?ID=CRD42023423617 (accessed on October 3rd, 2024).

3. Results

3.1. Study selection

Overall, 699 studies were imported from initial data search. After removing duplicates, 351 papers were retrieved. Titles, abstracts, full texts, and reference lists of all records were assessed against inclusion and exclusion criteria, according to a three-step screening approach (Figure 1). A final list of 48 studies was used for systematic reappraisal, including forty-seven RCTs exploring PEA efficacy and tolerability across illness conditions. The identified RCTs explored the effect of PEA supplementation on a heterogeneous group of illnesses enclosing (i) neuropsychiatric disturbances [41-55], (ii) sensory and/or motor neurological disturbances [42-44, 49, 50, 55-67], (iii) somatic disturbances [49, 53, 68-78], (iv) visceral disturbances [45, 79-87], and (v) blood/plasma or other tissue biomarkers alterations [49-51, 53, 70, 74, 75, 79, 80, 88]. Moreover, in some cases the effect of PEA was evaluated in terms of quality of life due to symptom management [49-51, 53, 62, 69, 72, 76-78, 81, 83]. A brief synthesis of the results is presented below and summarized in Table 1.
Additional data on methodological quality of studies are reported in Table 2.
All included studies underwent critical appraisal. Most studies were classified as having good quality, while two studies were categorized as having fair quality [86, 88], as assessed using the JBI critical appraisal tool for RCTs [40]. The detailed risk of bias and quality assessment with the corresponding questions for each item and list of RCTs is shown in Table 3.

3.2. Effect of Palmitoylethanolamide supplementation on neuropsychiatric disturbances across human conditions

Fifteen RCTs addressed this area (Table 1) using similar but not overlapping methodologies in terms of clinical condition of interest [bipolar affective disorder (BPAD) [41], spinal cord injury (SCI) [42], traumatic brain injury (TBI) [43], carpal tunnel syndrome (CTS) [44], primary open angle glaucoma/ocular hypertension (POAG/OH) [45], major depressive disorder (MDD) [46], autism spectrum disorder (ASD) [47], post-operative delirium (POD) [48], relapsing-remitting multiple sclerosis (RR-MS) [49], diabetes [50], disturbed sleep pattern (DSP) [51], schizophrenia (SCZ) [52], knee osteoarthritis (OA) [53], SARS-Cov-2 (COVID-19) infection [54], acute ischemic stroke (AIS) [55]], PEA formulation [undeclared [41, 45, 47, 52], ultramicronized-PEA (umPEA) [42, 44, 46, 49], Co-ultramicronized PEA plus Luteolin (Co-ultraPEALut) [43, 48, 54, 55], LipiSperse PEA [50, 51, 53]], PEA intake route (oral capsule [41, 46, 47, 50-53], sublingual microgranules [42, 44, 54], oral tablet [43-45, 48, 49], oral suspension [55]), PEA dosage (600 mg/bid [41, 42, 44, 46, 47], 700 mg/bid [43, 48, 52, 54, 55], 600 mg/day [44, 45, 49], 300 mg/bid [50, 53], 150 mg/bid [51, 53]), PEA period of exposure (6 weeks [41, 46], 12 weeks [42, 55], 26 weeks [43], 20 weeks [44], 8 weeks [45, 50-54], 10 weeks [47], 3 days [48], 53 weeks [49]), PEA use as monotherapy [44, 45, 48-51, 53, 54] or add-on therapy [41-43, 46, 47, 52, 55], level of concealment (double-blind [41, 42, 45-47, 49, 51-54],

single-blind [43, 48], open-label [44, 55], quadruple-blind [50]), and comparison to placebo [41, 42, 45-47, 49-54] or control treatments [43, 44, 48, 55]. Notably, PEA was well-tolerated throughout the studies, showing no signs of extrapyramidal symptoms [41, 52] or any other relevant adverse events (AEs) [41, 42, 44-48, 50-53, 55] during the entire period of administration.

3.2.1. Effect of Palmitoylethanolamide supplementation on mood, anxiety, and stress

Overall, studies exploring PEA ability to improve mood disturbances compared to placebo or control treatment suggested its efficacy as add-on therapy in reducing either depressive symptoms in citalopram-treated MDD patients [46] and acute manic symptoms in lithium- and risperidone-treated BPAD patients [41], as well as monotherapy in reducing depressive symptoms in diabetic patients with peripheral neuropathy [50]. Contrastingly, no significant reduction of depressive symptoms emerged in PEA-treated BPAD [41], SCI [42], TBI [43], SCZ [52], and knee OA [53] patients, in comparison to placebo or control treatment. A limited number of studies examined PEA effects on anxiety [42, 50, 53] and stress [50, 53], with benefits observed specifically on anxiety symptoms among knee OA patients [53] and a trend towards reduced stress levels among PEA-treated diabetic patients [50]. Lastly, no increase in the occurrence of mood disorders and anxiety, monitored as adverse events, was observed among PEA-treated patients with POAG/OH [45].

3.2.2. Effect of Palmitoylethanolamide supplementation on attention, awareness, and cognition

Five RCTs explored the selective effects of PEA across conditions affecting higher-level cognitive functions [43, 48, 49, 54, 55]. A single trial demonstrated the superiority of PEA compared to placebo add-on to treatment as usual (TAU; i.e., antiplatelet agents, anticoagulants, antiepileptic medications) in the recovery of TBI-related deficits in working memory and executive functions [43]. PEA add-on to TAU resulted in an increased number of thrombolysis-treated AIS patients able to undergo cognitive evaluations after the stroke event, as well as an improvement in cognitive performances, although the superiority over the comparator was not proven [55]. Also, PEA monotherapy proved to be effective in reducing both the incidence and maximum severity of POD, although not affecting POD duration, in comparison to control treatment [48]. On the contrary, no difference between PEA and placebo was observed over measures of calculation ability and auditory information processing speed and flexibility among RR-MS patients, despite reports of subjectively improved cognitive function and health [49]. Finally, PEA monotherapy enhanced measures of Gamma-aminobutyric acid (GABA)ergic activity and synaptic plasticity among patients experiencing long-term post-COVID-19 fatigue and cognitive sequelae (e.g., forgetfulness, cloudiness, difficulty in focusing), although it did not significantly impact clinical measures of cognitive performance [54].

3.2.3. Effect of Palmitoylethanolamide supplementation on sleep

Accumulating studies have been parsing PEA effects on several sleep domains across different clinical conditions, including SCI [42], CTS [44], diabetes [50], and knee OA [53], converging towards the evidence of improved sleep-wake rhythm or sleep adequacy [44, 50], sleep latency [44], sleep-related troubles in daily activities (e.g., shortness of breath or headache) or daytime somnolence [44, 50], and sleep duration [44]. In contrast, mixed or not significant findings emerged regarding PEA efficacy on measures of sleep quality [44, 53], sleep quantity [50], and sleep disturbances, snoring, or insomnia [42, 44, 50]. Additionally, PEA was exclusively effective on self-monitored sleep onset latency among subjects with primary DSP, while it did not have a significant impact on other sleep measures [51]. Notably, there were no observed sleep disturbances reported as adverse events in PEA-treated patients with POAG/OH [45].

3.2.4. Effect of Palmitoylethanolamide supplementation on psychotic symptoms and autistic behaviors

A single study demonstrated the beneficial effects of PEA adjunctive therapy on residual negative psychotic symptoms and general psychopathology measures in risperidone-treated SCZ patients, although not resulting in improved positive psychotic symptoms in comparison to placebo [52]. Results from an RCT among risperidone-treated autistic children yielded the superiority of PEA add-on compared to placebo in the treatment of core symptoms of ASD, particularly in the domains of irritability, hyperactivity, and inappropriate speech, but not stereotypic behavior, either in terms of symptom improvement or response rate [47].

3.3. Effect of Palmitoylethanolamide supplementation on sensory and/or motor neurological disturbances across human conditions

Altogether, seventeen RCTs explored this topic, using similar but not overlapping methodologies in terms of condition of interest [SCI [42], migraine [67], TBI [43], CTS [44, 56, 60, 61], post-COVID-19 olfactory impairment [57-59, 65, 66], compressive-type lumbar sciatica [62], RR-MS [49], burning mouth syndrome (BMS) [63], amyotrophic lateral sclerosis (ALS) [64], diabetes [50], AIS [55]], PEA formulation [umPEA [42, 44, 49, 63, 64], Co-ultraPEALut [43, 55, 57-59, 65, 66], undeclared [56, 60, 61], micronized-PEA (mPEA) [62], LipiSperse PEA [50, 67], Co-ultraPEALut/Alpha-lipoic acid (ALA) [65]], PEA intake route (sublingual microgranules [42, 44, 63], oral tablet [43, 44, 49, 56-61, 64], oral capsule [50, 62, 67], oral sachet [65, 66], oral suspension [55]), PEA dosage (600 mg/bid [42, 44, 63, 64], 700 mg/bid [43, 55, 57-59], 600 mg/day [44, 49, 56, 60, 61], 300 mg/day [62], 300

mg/bid [50, 62, 67], 700 mg/day [65, 66]), PEA period of exposure (12 weeks [42, 55, 58, 59, 65, 66], 8 hours [67], 26 weeks [43], 4 weeks [56, 57], 20 weeks [44], 8 weeks [50, 60, 61, 63], 3 weeks [62], 53 weeks [49], 24 weeks [64]), PEA use as monotherapy [44, 49, 50, 56-63, 65-67] or add-on therapy [42, 43, 55, 64, 66], level of concealment (double-blind [42, 49, 56, 58-63, 67], single-blind [43, 57, 64-66], open-label [44, 55], quadruple-blind [50]), and comparison to placebo [42, 49, 50, 56, 58-63, 65-67] or control treatments [43, 44, 55, 57, 64, 65]. PEA was well-tolerated throughout the studies, showing no relevant AEs during the entire period of administration [42, 44, 50, 55, 58, 60-63, 67].

3.3.1. Effect of Palmitoylethanolamide supplementation on neuropathic pain

The major strand of RCTs reported on PEA effects over neuropathic pain across several conditions, including CTS [44, 56, 60, 61], SCI [42], lumbar sciatica [62], BMS [63], and diabetes [50]. Among the three RCTs investigating PEA effects on neuropathic pain in CTS patients, PEA was found to significantly improve ultrasound [56] and subjective measures of symptom severity and functional status [60, 61] overtime, although yielding conflicting results in terms of hyperalgesia control [44, 60, 61], time without paresthesia during wrist flexion [44, 60, 61], and electrophysiological assessments [56, 60, 61]. Patients with lumbar sciatica treated with PEA experienced significantly greater reduction in pain intensity and improvement in disability measures compared to placebo, with effects increasing in a dose-dependent manner [62]. Neuropathic pain, perceived as burning sensation, was also improved among PEA-treated BMS patients compared to those treated with placebo, and pain reduction persisted even 4 months after PEA discontinuation [63]. An RCT investigated the efficacy of PEA in managing pain associated with diabetic peripheral neuropathy (PNP), revealing a significant decrease in overall pain severity, pain interference with life activities, superficial and deep pain, paroxysmal and evoked pain, and paresthesia [50]. Finally, a study examining the effects of PEA on symptoms of traumatic or non-traumatic SCI found a significantly larger decrease in the use of rescue medications compared to placebo, although there was no impact on sensory examination, global impression, neuropathic pain relief, pain-associated unpleasantness, pain interference with life activities, and health-related quality of life [42].

3.3.2. Effect of Palmitoylethanolamide supplementation on pain associated with migraine

A single RCT addressed PEA effects in migraine patients instructed to take either PEA or placebo at pain onset and 2 hours later in case of persisting episode. The study found PEA superiority to placebo, with more resolved headaches after 2 hours, more resolved headaches after 8 hours, more resolved

migraines of moderate severity at onset, lower pain scores at 1.5 hours, lower pain scores at 4 hours, and reduced need for rescue medications [67].

3.3.3. Effect of Palmitoylethanolamide supplementation on altered olfactory function

Five studies investigated the effects of PEA monotherapy on olfactory impairment recovery in post-COVID-19 patients, both prior to [58, 59], after [57, 65], or alongside olfactory training [66], using overlapping methodologies in terms of PEA formulation (i.e., Co-ultraPEALut, Co-ultraPEALut/ALA) and sensory function assessment. Consistent improvements emerged in global anosmia/hyposmia scores among patients treated with PEA, also showing its superiority against comparators [57-59, 65, 66], as well as in terms of variation in anosmia/hyposmia scores [58], and proportion of patients recovering from anosmia [58, 59]. A study out of two [65] showed significantly higher parosmia resolution rates among Co-ultraPEALut- and Co-ultraPEALut/ALA-treated patients compared to those treated with ALA monotherapy, and among Co-ultraPEALut/ALA-treated patients compared to those treated with Co-ultraPEALut or placebo [59, 65].

3.3.4. Effect of Palmitoylethanolamide supplementation on mobility, spasticity, and sensory-motor deficits

Studies exploring this area produced mixed findings regarding the impact of PEA on motor neurological symptoms. PEA add-on effect did not significantly differ from placebo in terms of stiffness, spasms, and other descriptors of spasticity among SCI patients [42], nor modulated motor disability progression in β1a-IFN-treated patients with RR-MS [49]. On the other hand, self-reported spasticity was higher in PEA-treated SCI patients compared to those on placebo [42]. Also, patients with moderate TBI who were treated with PEA add-on to TAU showed significant ameliorations in their independence and mobility in daily activities, relative to their baseline condition, although superiority over control therapy remains to be clarified [43]. A single study conducted among thrombolysis-treated AIS patients showed that PEA initiation as add-on to TAU (i.e., alteplase) within the first 72 hours after the ischemic event led to improved neurological deficits overtime [55]. Additionally, PEA-treated patients experienced greater improvements in mobility during daily activities and functional independence, showing significantly better outcomes than the comparator group by the end of the treatment [55].

3.3.5. Effect of Palmitoylethanolamide supplementation on altered muscle force and respiratory capacity

Results from a single study interrogating the effect of PEA add-on to riluzole compared to riluzole alone in ALS patients converged towards higher proportion of survivors among PEA-treated patients. Indeed, a slower decline of pulmonary function over time emerged, assessed as percentage of forced vital capacity (FVC%) and bulbar and respiratory functions measurements [64].

3.4. Effect of Palmitoylethanolamide supplementation on somatic disturbances across human conditions

Thirteen studies appraised this area [49, 53, 68-78], using similar but not overlapping methodologies in terms of condition of interest [impacted lower third molar (ILTM) [68], allergic rhinitis (AR) [74], chronic back/joints/limbs pain [69], periodontitis [70], arthritis [75], temporomandibular joint OA [71], upper respiratory tract infections (URTI) [72, 76], RR-MS [49], atopic eczema [77], fibromyalgia (FM) [78], knee OA [53], asteatotic eczema [73]], PEA formulation (mPEA [68], LipiSperse PEA [53, 74, 76, 77], umPEA [49, 69], PEA/baicalin [70], PEA/astaxanthin [75], undeclared [71], Impulsin [72], PEA/Acetyl-L-Carnitine (ALC) [78], PEA/AEA [73]), PEA intake route (oral tablet [49, 68, 69, 71, 72, 75, 78], oral capsule [53, 70, 74, 76], skin-applied moisturizing cream [77], skin-applied emollient cream [73]), PEA dosage (300 mg/bid [53, 68, 71, 76], 300 mg/day [74], 600 mg/bid [69, 78], 800 mg/bid [70, 75], 300 mg + 600 mg/day [71], 600 mg/tid [72], 600 mg/day [49], 150 mg/bid [53], 1.5% concentrated cream/bid [77], 0.3% 2 mg/cm^2/bid [73]), PEA period of exposure (2 weeks [68, 71, 74], 3 weeks [69], 5 days [70], 12 weeks [75, 76, 78], 12 days [72], 53 weeks [49], 8 weeks [53], 4 weeks [73, 77]), PEA use as monotherapy [49, 53, 68, 69, 71-74, 76, 77] or add-on therapy [70, 75, 78], level of concealment (single-blind [68], double-blind [49, 53, 69, 70, 72-74, 76, 77], open-label [75, 78], triple-blind [71]), and comparison to placebo [49, 53, 69, 72, 74, 76] or control treatments [68, 70, 71, 73, 75, 77, 78]. A minority of studies systematically reported on PEA good tolerability during the entire observation period [53, 68-71, 73, 75-78], only mentioning isolated cases of drowsiness [68], palpitations [68], diarrhoea [69, 76], skin rash [76], dizziness [78], nausea [78], blurred vision [78], sleepiness [78], and dry mouth [78].

3.4.1. Effect of Palmitoylethanolamide supplementation on joints, back, limbs, and chronic widespread pain and altered function

Five RCTs explored the efficacy of PEA in this group of disturbances. Three studies addressing PEA effect on temporomandibular [71], knee [53, 75], and hip [75] OA [53, 71] or isolated arthritis [75] pointed towards the evidence of a greater reduction of total and average pain measures [53, 71, 75], decreased worst and least pain levels [71], and improved measures of joint and physical stiffness, joint function, and joint mobility range [53, 71, 75], in comparison to baseline scores [75], placebo

[53], and nonsteroidal anti-inflammatory drugs (NSAIDs) [71]. A pilot RCT conducted in single elderly outpatients (N-of-1 design) evaluated the effectiveness of PEA monotherapy on chronic non-cancer, non-ischemic pain in the back, joints, or limbs, showing a statistically significant improvement in pain intensity and function impairment in three out of seven patients completing the trial compared to placebo [69]. Lastly, a single RCT explored the effects of PEA add-on to duloxetine plus pregabalin compared to duloxetine plus pregabalin alone among FM patients, showing favorable effects on widespread nociplastic pain, with improvements in quality of life measures [78].

3.4.2. Effect of Palmitoylethanolamide supplementation on dental and periodontal inflammation

The efficacy of PEA has been tested in the context of orthodontic treatments, encompassing both surgical [68] and non-surgical [70] approaches. In surgical patients who underwent lower third molar extraction, treatment with PEA led to a significant reduction in postoperative pain and delayed edema onset, but not improved swelling severity, compared to control group [68]. Similarly, PEA administration following scaling and root planing (SRP) in patients with periodontitis significantly improved both short-term postoperative pain relief and long-term outcomes, including clinical attachment level, probing depth, and bleeding on probing, compared to control group [70].

3.4.3. Effect of Palmitoylethanolamide supplementation on respiratory tract infections and allergy symptoms

Two studies investigated PEA effects on patients with URTI, compared to placebo, indicating a significant reduction in episodes of fever [72], headache [72], sore throat [72, 76], nasal congestion [72], nasal discharge [72], cough [72, 76], and a tendency to improved hoarseness [76] and ability to breathe easily [76], as well as a decrease in the number of total patients [72], in-patients [72], out-patients [72], and patients sick at least once [76]. No consistent improvements emerged in terms of illness duration [72, 76]. A single RCT reported on the impact of PEA in AR, indicating beneficial effects on measures of allergy symptoms over time (e.g., nasal congestion, sneezing, itchy nose, runny nose), in both PEA-treated and placebo-treated patients, although with no significant difference between groups [74].

3.4.4. Effect of Palmitoylethanolamide supplementation on cutaneous pain, erythema, and eczema

Three RCTs examined PEA effects on dermatologic issues. PEA significantly improved pain sensation in β1a-IFN cutaneous injection site among RR-MS patients compared to placebo [49]. PEA-treated patients with asteatotic eczema experienced improved skin scaling, dryness, itching, and current perception threshold (CPT), compared to a control emollient product, while no effects were

observed on skin integrity [73]. In both cases, PEA did not impact erythema features [49, 73], especially its width [49]. A third RCT exploring PEA effects in patients with atopic dermatitis showed significant improvements over time on measures of symptom severity, scratches, and itchiness, also yielding its superiority to a standard moisturizer on measures of redness, dryness, and total symptom scores. No significant impact emerged on quality-of-life measures [77].

3.5. Effect of Palmitoylethanolamide supplementation on visceral disturbances across human conditions

Eleven studies addressed this area, using similar but not overlapping methodologies in terms of condition of interest [endometriosis (EMS) [79], irritable bowel syndrome (IBS) [80, 87], POAG [45, 83], chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS) [81], isolated vestibulodynia [82], normal tension glaucoma (NTG) [83, 86], isolated OH [84], primary dysmenorrhea [85]], PEA formulation [mPEA/transpolydatin [79, 82, 85], mPEA/polydatin [80, 87], undeclared [45, 84], PEA/ALA [81], umPEA [83, 86]], PEA intake route (oral tablet [45, 79, 80, 82-87], oral capsule [81]), PEA dosage (400 mg/bid [79], 200 mg/day [80], 600 mg/day [45, 83], 300 mg/day [81], 400 gm/day [82], 300 mg/bid [84, 86], 200 mg/tid [87]), PEA period of exposure (12 weeks [79-81, 84, 87], 8 weeks [45, 82], 16 weeks [83], 10 days [85], 24 weeks [86]), PEA use as monotherapy [45, 79-81, 84-87] or add-on therapy [82, 83], level of concealment (double-blind [45, 79, 80, 82, 84, 87], single-blind [81, 83], open-label [86]), and comparison to placebo [45, 79, 80, 82, 84, 85, 87] or control treatments [81, 83, 86]. Seven studies systematically reported no PEA-related AEs during the observation period [45, 79-81, 83-87].

3.5.1. Effect of Palmitoylethanolamide supplementation on gynecological and genitourinary pain or altered function

Two RCTs exploring gynecological disturbances demonstrated greater improvements of dysmenorrhea [79] and pelvic pain [79, 85] among PEA-treated EMS [79] and primary dysmenorrhea [85] patients compared to those on placebo, although indicating PEA inferiority to Cyclooxygenase-2 (COX-2) inhibitors [79]. Opposedly, dyspareunia improved in EMS patients treated with PEA monotherapy [79], while no improvement was observed in patients with isolated vestibulodynia who received PEA as an add-on to transcutaneous electrical nerve stimulation (TENS) [82]. A study examining PEA effects in CP/CPPS patients showed significant improvements on lower urinary tract symptoms, pain, voiding symptoms, and their impact on quality of life, but not over measures of erectile dysfunction, compared to treatment with Serenoa Repens [81].

3.5.2. Effect of Palmitoylethanolamide supplementation on intraocular pressure and visual field

Four studies addressed this area, converging towards the evidence of a greater reduction of intraocular pressure among PEA-treated patients with POAG [45, 83], NTG [83, 86], and isolated OH [84], compared to those treated with placebo [45, 84], topical therapy alone [83], or untreated [86]. PEA was shown to significantly reduce IOP values among PEA-treated versus comparator-treated patients across all studies [45, 83, 84, 86], alongside improvements on the P50 component of pattern electroretinogram (PERG) [83], peripheral endothelial dysfunction [84], and quality of life scores among POAG/NTG patients [83]. Contrasting results emerged regarding PEA ability to ameliorate measures of visual field [45, 83, 86] and visual acuity [45, 86]. PEA did not significantly impact measures of central corneal thickness [45], cup-to-disk (C/D) ratio [45], and other PERG components [83].

3.5.3. Effect of Palmitoylethanolamide supplementation on gastrointestinal pain or altered function

Two RCTs documented greater improvements of abdominal pain/discomfort among PEA-treated IBS adult [80] and pediatric [87] patients compared to those treated with placebo, alongside significantly higher responder rates across both populations [80, 87]. Only among children, PEA was superior to placebo in reducing the frequency of episodes of abdominal pain and scores of life interference [87]. PEA did not significantly impact measures of epigastric pain frequency [80], bloating frequency [80], episodes of vomiting frequency [80], as well as measures of bowel habit [80, 87] and stool form [80].

3.6. Effect of Palmitoylethanolamide supplementation on blood/plasma or other tissue biomarkers alterations across human conditions

Overall, ten studies addressed this area, focusing on PEA-dependent modulation of blood/plasma [49-51, 53, 74, 75, 79, 80, 88] or other tissue (e.g., gingival crevicular fluid) [70] inflammatory and oxidative stress markers [49, 50, 70, 74, 75, 79, 80, 88], pathology-unspecific and -specific hematology or biochemistry markers [50, 51, 53, 88], and eCBs/NAEs signaling [49, 80]. Across RCTs investigating PEA effects in β1a-IFN-treated RR-MS [49], diabetes [50], SRP-treated periodontitis [70], AR [74], arthritis [75], EMS [79], IBS [80], and COVID-19 [88] compared to placebo, standard treatment, or baseline condition, PEA intake led to significantly adjusted levels of several inflammatory biomarkers, including decreased Interleukin-1β (IL-1β) [70, 75], decreased Interleukin-6 (IL-6) [50, 75, 88], decreased Interferon Gamma (IFN-γ) [49], decreased Interleukin-17 (IL-17) [49], decreased Interleukin-8 (IL-8) [74], decreased Interleukin-4 (IL-4) [74], decreased histamine [74], decreased IL-1β/ IL-10 ratio [70], decreased D-Dimer [88], decreased Cluster of differentiation 3 (CD3+) and Cluster of differentiation 8 (CD8+) lymphocytes absolute count [88],

decreased Anti-SARS-CoV-2 Immunoglobulins G (IgG) [88], increased Lymphocytes count [88], decreased Neutrophil/Lymphocytes ratio [88], decreased free oxygen radicals [88], and at least a tendency to decreased Tumor Necrosis Factor-Alpha (TNF-α) [49, 70, 74, 75, 88]. Contrasting results emerged regarding PEA effects on modulating levels of C-Reactive Protein (CRP), either significantly decreasing [50, 88] or not [79]. Also contrasting results emerged regarding PEA effects on modulating levels of Interleukin-10 (IL-10), either significantly increasing [70] or decreasing [74]. PEA showed no effects on Erythrocyte Sedimentation Rate (ESR) blood levels in EMS patients [79], immune cell and mastocyte activation in IBS patients [80], and fibrinogen in both diabetic [50] and COVID-19 patients [88].

All pathology-unspecific hematology and biochemistry tests remained within normal ranges throughout the studies among PEA-treated DSP [51], knee OA [53], and COVID-19 [88] patients.

Also, PEA treatment was not associated with changes in levels of fasting blood glucose and glycated hemoglobin among diabetic patients [50].

Circulating eCBs/NAEs and related enzymes were modulated by PEA intake compared to placebo among β1a-IFN-treated RR-MS patients, including significant increase in PEA, AEA, and OEA plasma levels, lower expression of FAAH, and an inverse correlation between PEA and cytokines plasma levels [49]. On the contrary, no impact on eCBs/NAEs levels was observed among PEA-treated IBS patients [80].

4. Discussion

Existing reviews interrogating the therapeutic role of PEA encompassed various designs of clinical studies and focused on specific symptoms or PEA formulations [20, 89-94]. However, to the best of our knowledge, this is the first systematic review collecting all RCTs examining the effects of PEA supplementation across human illnesses, aiming to comprehensively address its impact on any targeted symptoms, either measured clinically or using appropriate assessment scales.

This systematic review indicates several prominent treatment implications of PEA supplementation that deserve to be highlighted.

Not surprisingly, the most convincing findings emerged from RCTs examining PEA efficacy on pain severity across several clinical conditions, encompassing neuropathic pain [42, 44, 50, 56, 60-63], migraine-associated pain [67], somatic pain [49, 53, 68-71, 75], visceral pain [79-81, 85, 87], and nociplastic pain [78]. Evidence largely pointed to improved unidimensional [e.g., Numeric Rating Scale (NRS), Visual Analogue Scale (VAS)] [44, 49, 50, 53, 62, 63, 67-71, 78, 79, 85] and multidimensional [e.g., Patient Global Impression of Change (PGIC), Brief Pain Inventory-Diabetic Peripheral Neuropathy (BPI-DPN)] [44, 50, 53, 60, 61, 75, 79-81, 87] pain measures, also

underscoring PEA effect on daily functioning [49, 50, 53, 62, 78], as a result of pain management. When PEA did not significantly impact pain intensity scores, recovered sensory conduction and morphology measures [56] or a reduction in the use of conventional pain medications [42] were at least observed. In the few cases when the nature of the non-placebo comparator was clearly defined, PEA showed its superiority to both NSAIDs [71, 75] and other natural products with anti-inflammatory properties (i.e., Serenoa Repens) [81], but not to COX-2 inhibitors [79]. Recent robust systematic reviews and metanalyses on this topic have already yielded superior pooled estimates of PEA analgesic effect over comparators, particularly in the treatment of chronic pain [20, 91, 94], with favorable outcomes on quality of life [20], sleep quality [20], and physical function [20], as well as no notable side effects [20], although the high methodological heterogeneity (e.g., dosing regimen, PEA formulations) and small primary samples size may have limited the statistical power of the conclusions drawn [20, 91, 94]. Additionally, our systematic reappraisal unveiled a paucity of RCTs showing beneficial effects of PEA in the treatment of neurological motor symptoms occasionally but not exclusively associated to neuropathic pain, including reduced mobility, spasticity, stiffness, and increased fatigability [42, 43, 49]. A more in-depth investigation of this area through RCTs may be relevant, given the evidence of PEA benefits on neuromotor symptoms [55, 64, 95] and muscle recovery [64, 96] both in clinical [55, 64, 95] and healthy [96] populations. Noteworthy, among PEA-treated diabetic [50], RR-MS [49], periodontitis [70], and arthritis [75] patients, both pain improvement [49, 50, 70, 75] and better general wellbeing (e.g., cognitive function, sleep quality) [49, 50] were accompanied by contextual peripheral tissues lowering proinflammatory biomarkers (e.g., TNF-α, IL-1β, CRP, IL-6, IFN-γ, IL-17) [49, 50, 70, 75], increasing anti-inflammatory biomarkers (e.g., IL-10) [70], and enhanced eCBs/NAEs signaling (e.g., lower FAAH expression, increased PEA, AEA, and OEA plasma levels) [49], indicating that PEA exerts its effects over factors either precociously altered in the early phases of a disease [49], or contributing to illness progression [50, 70]. Few other studies explored potential correlations between pain relief and blood/plasma biomarkers modulation, failing to retrieve significant results, and suggesting the possibility that PEA may act through different pathways depending on the clinical condition [53, 79, 80], with the potential for long-lasting effects even after treatment discontinuation (e.g., through TRPV1 desensitization) [80]. Indeed, PEA ability to modulate immune responses and halt the release of inflammatory mediators, driven by its diverse molecular and cellular interactions, appears to be the most accredited mechanism underlying its efficacy in controlling chronic pain, where neuroinflammation contributes to the persistence of pain signals [91, 97]. However, correlation between pain management and inflammatory biomarkers or eCBs/NAEs signaling modulation as a

response to PEA treatment remains largely unexplored across RCTs, warranting the need of further studies to fill this literature gap.

Numerous studies have investigated PEA effects on neuropsychiatric symptoms, either as primary [41, 43, 44, 46-48, 51, 52, 54] or secondary [42, 45, 49, 50, 53, 55] objectives, showing a general trend towards improved wellbeing in PEA-treated patients. First, depressive and acute manic symptoms appeared to be positively affected by PEA add-on to conventional therapy in MDD [46] and BPAD [41] patients respectively, possibly thanks to PEA multi-faceted mechanism of action, including PPAR-α-mediated anti-inflammatory effects, GPR55 agonism (with implications for the modulation of monoaminergic neurons), TRPV1 allosteric modulation, glutamate neuro-toxicity prevention, and neurogenesis promotion [17, 21]. Improved depressive symptoms were also observed among PEA-treated diabetic patients [50], being less clear across other conditions only secondarily affecting mood [42, 43, 53]. Second, most RCTs investigating PEA short- [48] or long-term [43, 49, 54, 55] therapeutic potential in treating conditions associated with impaired cognitive function have yielded solid favorable results, either using psychometric measures [43, 48, 55], or subjective reports [49], or neurophysiological assessments [54], supposedly accounting for PEA ability to counteract neurodegenerative processes through the modulation of oxidative stress and neuroinflammation pathways, including astrocyte and microglia proliferation and neuronal loss [98]. Intriguingly, PEA was also shown to exert its favorable effects on attention, executive function, and memory domains in healthy adults, possibly through increased neurotrophic factors signaling [99]. Third, two out of three RCTs exploring PEA impact on sleep disturbances both as consequence and contributor of neuropathic pain pointed to PEA beneficial effects mostly on measures of sleep adequacy and sleep-related troubles on daily activities [42, 44, 50], whereas no significant improvements emerged from single studies assessing PEA effects on disrupted sleep secondary to somatic pain [53] and on primary DSP [51]. Fourth, PEA add-on to second-generation antipsychotics exerted beneficial effects on both residual negative psychotic symptoms in SCZ [52] and behavioral and speech disturbances in ASD [47], underscoring its potential impact on neurodevelopmental manifestations driven by mesocorticolimbic dopaminergic disruption and glutamate neurotoxicity [100, 101], sustained by inflammatory processes [102-104], and often difficult to treat or unresponsive to conventional psychotropic medications [105-108].

Alongside these two main strains of research, additional evidence appears more scattered but is still worth mentioning. First, narrow but methodologically consistent data emerged from five RCTs exploring Co-ultraPEALut in combination to olfactory training on chronic hyposmia/anosmia recovery and, to a lesser extent, parosmia recovery in COVID-19 patients, possibly reducing lingering neuroinflammation of the olfactory bulb and higher olfactory centers following SARS-CoV-2

infection [57-59, 65, 66, 89]. These findings are also coherent with the previously discussed beneficial effects of PEA on brain fog and cognitive fatigue in the context of long-COVID syndrome [54], which further highlight the anti-neuroinflammatory potential of PEA. Second, aligning with recent meta-analysis [45, 84, 86, 109], PEA was shown to significantly reduce IOP among glaucoma patients across four RCTs [45, 83, 84, 86], due to still unclear mechanisms that may include the modulation of peripheral endothelial dysfunction [84]. Finally, two RCTs converge towards the evidence of PEA efficacy in the context of allergic symptoms, both respiratory [74] and cutaneous [77], possibly manifesting its effects through the reduction of histamine and multiple immune biomarkers levels.

In addition to its primary therapeutic effects, the considerations outlined above highlight how PEA supplementation may serve as a valuable treatment for transdiagnostic symptoms intertwining neuropsychiatric and physical conditions, especially those fueled by neuroinflammatory processes—both in the short and in the long term. This aspect would be particularly noticeable in the case of formulations with smaller particle size [42-44, 46, 48, 49, 54, 55, 57-59, 62-66, 68, 69, 79, 80, 83, 86-88] or those produced using cold-water dispersible technology [50, 51, 53, 67, 74, 76, 77], maximizing PEA bioavailability and efficacy. To this extent, it is noteworthy that most RCTs addressing PEA effects on neuropsychiatric or neurologic disturbances involved micronized/ultramicronized formulations, either alone [42, 44, 46, 49, 62-64] or in combination with flavonoids or other antioxidant compounds (e.g., luteolin, ALA) [43, 48, 54, 55, 57-59, 65, 66], or LipiSperse PEA [50, 51, 53, 67], possibly to favor PEA penetration into the nervous system through the blood-brain barrier [21, 25, 37].

Moreover, PEA safety and tolerability would make it an excellent and acceptable option for individuals already receiving pharmacological treatments for their underlying condition, either during cross-titration if the current medication is being gradually discontinued, or as an add-on therapy when additional support is needed, or in the management of unpleasant side effects [49].

4.1. Limitations and literature gaps

The findings emerging from our systematic review should also be seen considering some limitations, regarding either the characteristics of included studies or the methodological approach adopted. First, despite our work focused on clinical populations, only few studies selected were conducted in large samples (> 100 patients) [51, 53, 58, 59, 62, 66, 72, 74, 76, 78, 85] or at multiple sites [41, 42, 46, 47, 52, 58, 62, 72, 80], with the majority carried out in Italy [43-45, 48, 49, 54-59, 62-66, 68-71, 78-88]. This may result in some differences between the study participants and the population typically treated in routine clinical practice, alongside the frequent inclusion of otherwise healthy patients without any comorbid physical, neurological, or mental health conditions (see Supplementary Table).

To improve the interpretation and generalizability of the findings about PEA supplementation across all clinical conditions, larger multicenter RCTs will be necessary.

Second, despite compelling, converging evidence on the treatment of neuropsychiatric symptoms is still limited, particularly from RCTs investigating PEA effects in populations with primary neuropsychiatric disturbances, suggesting interpreting presented findings with caution. Indeed, adjuvant therapy with PEA showed to enhance the effectiveness of ongoing psychotropic medications in overt mental health conditions, possibly allowing a reduction in the necessary dosage of treatments that are not devoid of severe adverse effects [41, 46, 47, 52]. However, there is still lack of RCTs exploring PEA as monotherapy for conditions that are often treatment-orphan (e.g., first-episode seizures, depressive symptoms in ASD, and clinical high-risk states for psychosis), particularly in the early stages preceding the onset of full-blown neuropsychiatric disorders or during remission phases, with potential disease-modifying properties. Moreover, evidence linking improvements in neuropsychiatric symptoms with inflammatory response or eCBs/AEs signaling is still scarce [49], and it remains unclear whether PEA effects correlate with the modulation of other biological systems. Future studies across all conditions will be needed to systematically investigate the correlation between clinical response to PEA supplementation and the modulation of biological markers, potentially considering its direct impact on microbiome composition and function as well [110-113].

Third, while the use of non-placebo comparators was consistently established across the studies [43, 44, 48, 55, 57, 64, 68, 70, 71, 73, 75, 77-79, 81, 86, 88], in most cases its nature was not explicitly defined, instead described with terms like “standard therapy” or “standard treatment”, thereby limiting comparisons with other renowned anti-inflammatory medications [71, 75, 79, 81]. Also, only a limited number of studies have compared different PEA formulations [65] and dosages [44, 66], unveiling a need for further investigation before definitive conclusions can be drawn regarding PEA efficacy for the treatment of certain disorders. To this end, although most studies were rated as high quality using the JBI critical appraisal for RCTs [40], this tool fails to capture specific details on the trial medication, which were omitted in a few studies [41, 45, 47, 52, 56, 60, 61, 71, 84]. This information, along with different dosages, intake routes, period of exposure, and use as monotherapy or add-on treatment and is crucial for understanding PEA effects and accurately comparing results and side effects across studies.

Finally, although we searched three major medical databases, reviewed two clinical trial registries, and manually checked all reference lists, some RCTs may still have been missed, a limitation inherent to all systematic reviews.

5. Conclusion

This systematic review sought to provide a comprehensive overview of the therapeutic potential of PEA across all RCTs exploring its efficacy and safety in clinical populations. While further research is necessary, accumulating literature has already underscored PEA as a versatile dietary supplement with a favorable safety profile for both long- and short-term treatment of several transdiagnostic symptoms, offering potential disease-modifying effects and a biological profile closely aligned with human physiology.

Figure 1. PRISMA flowchart of search strategy for systematic review.

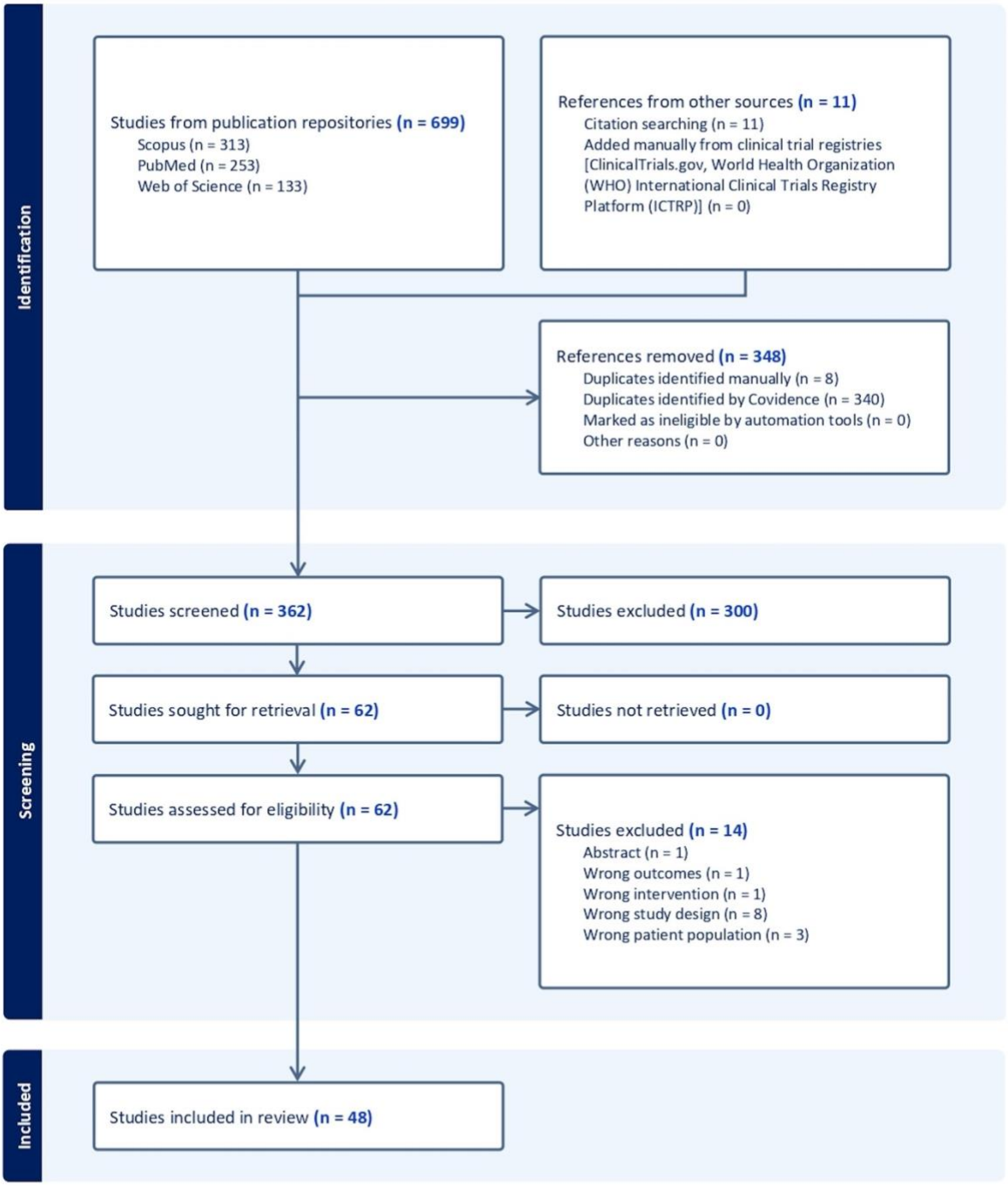


Table 1. Summary results of Randomized Controlled Trials investigating Palmitoylethanolamide supplementation across human diseases.

Study ID	Aim of study	Type of study	Sample size	Outcome measures	Summary outcomes
Abedini et al., 2022 (Iran) [41]	To assess PEA effect on acute mania in BPAD patients	PEA effect on mood, anxiety, and stress	63 (PEA: 32; PLB: 31)	1. Neuropsychiatric measurement (YMRS, HDRS) 2. AEs measurement (ESRS, open-ended questions, comprehensive side effect checklist)	1. Neuropsychiatric measurement: (a) **YMRS: time x treatment interaction**; **YMRS global score**: PEA vs. PLB, NS (Baseline, Week 2, Week 4); **PEA < PLB (Week 6)**; **YMRS score changes**: From Baseline to Week 2: PEA vs. PLB, NS; **From Baseline to Week 4, From Baseline to Week 6: PEA > PLB**; (b) HDRS score changes: From Baseline to Week 6: PEA vs. PLB, NS 2. AEs measurement: (a) ESRS: time x treatment interaction, NS; ESRS global score: PEA vs. PLB, NS (Baseline, Week 1, Week 2, Week 4, Week 6); ESRS score changes: all comparisons, NS; (b) Frequency of adverse events (drowsiness, dizziness, increased appetite, skin rash, diarrhea, dry mouth, sore throat, tachycardia): PEA vs. PLB, NS
Albanese et al., 2022 (Italy) [88]	To assess PEA effect on inflammatory response in COVID-19 patients	PEA effect on blood/plasma or other tissue biomarkers alterations	90 (PEA: 45; CTRL: 45)	Blood/plasma and/or other tissue measurement (Inflammatory markers quantification, oxidative stress markers quantification)	**D-Dimer, CRP, IL-6, CD3 + CD8 + absolute count, Anti-SARS-CoV-2 IgG, Lymphocytes, Neutrophil/Lymphocytes ratio (T1-T0): PEA > CTRL**; Red blood cell, Hemoglobin, Hematocrit, MCV, Neutrophil, PLT, Myoglobin, PT, Fibrinogen, Antithrombin III, Creatinine, GFR, Nitrogenic Acid, D Vitamin, ESR, TNF-α, Liver function, Muscle damage: PEA vs. CTRL, NS; ΔFORD(T1-T0): PEA vs. CTRL, NS; **ΔFORT(T1-T0): PEA < CTRL**
Andresen et al., 2016 (Denmark, Norway, United Kingdom) [42]	To assess PEA effect on neuropathic pain and spasticity in SCI patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on sleep	73 (PEA: 36; PLB: 37)	1. Neurological measurement (NRS, use of rescue medication, neuropathic pain descriptors, evoked	1. Neurological measurement: (a) ITT, secondary ITT, PP populations: PEA vs. PLB, NS; (b) **Use of rescue medication: PEA, ↓; PLB, ↑; treatment effect**; (c) **Self-reported spasticity: PEA, ↑; PLB, ↓; Δ(baseline- LWT): treatment effect**; (d) Stiffness, (e) Spasms, (f) Spasticity angle, (g) NPSI,

		3. PEA effect on neuropathic pain 4. PEA effect on mobility, spasticity, and sensory-motor deficits		pain, MTS, PGIC, NNT) 2. Neuropsychiatric measurement (ISI, MDI, GAD-10) 3. QoL measurement (S-TOPS) 4. AEs measurement (Clinical assessment)	(h) CPSS, (i) Unpleasantness, (j) Pain interference, (k) Sensory examination, (l) Global impression, (m) Pain relief of neuropathic pain, (n) Pain relief at-/below-level of neuropathic pain: PEA vs. PLB, NS (all comparisons) 2. Neuropsychiatric measurement: (a) Anxiety, (b) Depression, (c) Sleep disturbance, (d) Insomnia: PEA vs. PLB, NS (all comparisons) 3. S-TOPS: PEA vs. PLB, NS 4. AEs measurement: PEA vs. PLB, NS
Bacci et al., 2011 (Italy) [68]	To assess PEA effect on swelling and pain in ILTM surgery patients	PEA effect on dental and periodontal inflammation	30 (PEA: 30; CTRL: 30)	1. Somatic measurement (Trismus, swelling, VAS, NSAIDs consumption) 2. AEs measurement (Clinical assessment, post-operative complications)	1. Somatic measurement: (a) **Effect on mean VAS: time; treatment**; T0: PEA vs. CTRL, NS; **T1, T2: PEA < CTRL**; (b) **Effect on mouth opening range: time**; treatment, NS; **gender**; T0, T1, T2: PEA vs. CTRL, NS; (c) Effect on distance from lateral canthus to gonion: all comparisons, NS; (d) **Effect on distance from labial commissure to tragus: time**; treatment, NS; T0, T1, T2: PEA vs. CTRL, NS 2. AEs measurement: PEA vs. PLB, NS
Bonzanino et al., 2024 (Italy) [55]	To assess PEA effect on neurological deficit, independence, disability, and cognitive impairment in AIS patients	1. PEA effect on attention, awareness, and cognition 2. PEA effect on mobility, spasticity, and sensory-motor deficits	60 (PEA: 30; CTRL: 30)	1. Neurological assessment (11-item NIHSS, Barthel index, mRS) 2. Neuropsychiatric measurement (MMSE, MoCA) 3. AEs measurement (Clinical assessment)	1. Neurological measurement: (a) **NIHSS score: T2 < T1 < T0 (PEA, CTRL)**; PEA vs. CTRL, NS; (b) **Barthel index: T2 > T1 > T0 (PEA, CTRL)**; PEA > CTRL, NS (T2); PEA vs. CTRL, NS (T0, T1); (c) **mRS score: T2 < T1 < T0 (PEA, CTRL)**; **PEA < CTRL (T2)**; PEA vs. CTRL, NS (T0, T1) 2. Neuropsychiatric measurement: (a) number of patients able to perform MMSE, (b) number of patients able to perform MoCA: PEA > CTRL, NS (T1, T2); (c) MMSE mean score, (d) MoCA mean score: T2 > T1, NS (PEA) 3. AEs measurement: PEA vs. CTRL, NS

Briskey et al., 2023 (Australia) [74]	To assess PEA effect on allergy symptoms in AR patients	1. PEA effect on respiratory tract infections and allergy symptoms 2. PEA effect on blood/plasma or other tissue biomarkers alterations	108 (PEA: 54; PLB: 54)	1. Somatic measurement (rTNSS, RQLQ) 2. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification, full blood count, enzyme and liver function test)	1. Somatic measurement: (a) rTNSS score: PEA vs. PLB, NS (all timepoints); (b**) rTNSS morning score: ↓ from Day 3 (PEA, PLB)**; (c) **rTNSS evening score: ↓ from Day 2 (PEA, PLB)**; (d) RQLQ score: PEA vs. PLB, NS; (e) **rTNSS score (sub-group with rTNSS > 4): Week 2 < baseline, Week 1**; (f) RQLQ total score: PEA vs. PLB, NS; (g) **RQLQ (all domains): ↓ overtime (PEA, PLB)** 2. Blood/plasma and/or other tissue measurement: (a) **histamine levels change: PEA > PLB**; (b) **IL-4 levels: Week 2 < baseline (PEA, PLB)**; (c) **IL-10 levels**, (d) **IL-8 levels**, (e) **TNF-α levels**, (f) **histamine levels: Week 2 < baseline (PEA)**; Week 2 vs. baseline, NS (PLB)
Briskey et al., 2024 (Australia) [67]	To assess PEA effect on pain and symptom duration in migraine patients	PEA effect on pain associated with migraine	80 (PEA: 40; PLB: 40)	1. Neurological measurement (VAS, migraine duration, migraine severity, use of rescue medication) 2. AEs measurement (Clinical assessment, self-administered questionnaire)	1. Neurological measurement: (a) number of migraines, (b) severity of migraines at onset, (c) use of rescue medication at 4 h: PEA vs. PLB, NS; (d) **number of resolved migraines at 2 h**, (e) **number of resolved migraines at 2 h with no use of rescue medication**, (f) **number of resolved moderate-at-onset migraines**, (g) **number of resolved migraine events at 8 h: PEA > PLB**; (h) **number of unresolved migraine events at 8 h**, (i) **number of reported rescue medication use: PEA < PLB**; (j) **VAS score: 30 min < baseline (PLB)**; **60 min < baseline (PEA)**; PEA vs. PLB, NS (all timepoints); (k) **VAS score change: PEA > PLB (1.5 h, 4 h)**; PEA vs. PLB, NS (other timepoints) 2. AEs measurement: PEA vs. PLB, NS
Campolo et al., 2021 (Italy) [43]	To assess PEA effects on memory and cognitive function in TBI patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on attention,	30 (PEA: 15; CTRL: 15)	1. Neuropsychiatric measurement (GCS, Marshal Score, MMSE, BNCE, BDI)	1. Neuropsychiatric measurement: (a) **MMSE score: PEA > CTRL**; Δ**MMSE: PEA > CTRL**; **MMSE score > 26: PEA > CTRL**; (b) BNCE: PEA vs. CTRL, NS; **BNCE Interference Memory: PEA > CTRL**; (c) BDI: PEA vs. CTRL, NS

		awareness, and cognition 3. PEA effect on mobility, spasticity, and sensory-motor deficits		2. Neurological measurement (Barthel index)	2. Independency (**Barthel index**): ↑ **PEA vs. baseline**; PEA vs. CTRL, NS
Cantone et al., 2024 (Italy) [65]	To assess PEA effect on olfactory function recovery in post-COVID-19 olfactory impairment	PEA effect on altered olfactory function	89 (Co-ultraPEALut: 17; ALA: 21; Co-ultraPEALut/ALA: 28; PLB: 23)	Neurological measurement (Sniffin' sticks)	**TDI score:** PLB post vs. PLB pre, NS; ALA post vs. ALA pre, NS; **Co-ultraPEALut post > Co-ultraPEALut pre**; **Co-ultraPEALut/ALA post > Co-ultraPEALut/ALA pre**; **TDI score at T0: PLB > Co-ultraPEALut, Co-ultraPEALut/ALA, ALA**; other comparisons, NS; **TDI score at T1: Co-ultraPEALut > PLB, ALA; Co-ultraPEALut/ALA > ALA;** other comparisons, NS **Parosmia resolution rates: Co-ultraPEALut > ALA**; **Co-ultraPEALut/ALA > Co-ultraPEALut, ALA, PLB**; other comparisons, NS
Cobellis et al., 2011 (Italy) [79]	To assess PEA effect on chronic pelvic pain in EMS patients	1. PEA effect on gynecological and genitourinary pain or altered function 2. PEA effect on blood/plasma or other tissue biomarkers alterations	61 (PEA: 21; PLB: 20; Celecoxib: 20)	1. Visceral measurement (Pelvic examination, transvaginal US, symptom questionnaire, VAS) 2. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification) 3. AEs measurement (Clinical assessment)	1. Visceral measurement: (a) **Dysmenorrhea,** (b) **Dyspareunia,** (c) **Pelvic Pain: 3 months vs. baseline, ↓ (all groups); Celecoxib < PEA < PLB (at 3 months)** 2. Blood/plasma measurement: (a) ESR, (b) CRP levels: PEA vs. Celecoxib vs. PLB, NS 3. AEs measurement: PEA vs. Celecoxib vs. PLB, NS
Coraci et al., 2018 (Italy) [56]	To assess PEA effect on pain relief in CTS patients	PEA effect on neuropathic pain	56	Neurological measurement (Mean I and III digits SNCV	**Mean SNCV III finger (ΔT)**, **Mean SAP III finger (ΔT): PEA group, ↑**; **PLB group, ↓**; T1 vs. T0, NS (both groups);

				and SNCA, III digit mean SAP, US W/F cross sectional area ratio of the median nerve, BCTQ)	**Mean US W/F (ΔT): PEA group, ↓; PLB, group,** ↑; T1 vs. T0, NS (both groups)
Costagliola et al., 2014 (Italy) [86]	To assess PEA effect on IOP and visual field in NTG patients	PEA effect on intraocular pressure and visual field	32 (PEA: 16; CTRL: 16)	1. Visceral measurement (BCVA, IOP with patient in sitting position at the slit lamp, CCT, visual field, red-free disc photo) 2. AEs measurement (Self-administered questionnaire)	1. Visceral measurement: (a) **IOP**, (b) **visual field (MD, PSD): Month 6 < baseline (PEA)**; Month 6 vs. baseline, NS (CTRL); (c) BCVA changes: PEA, NS; CTRL, NS 2. AEs measurement: PEA vs. CTRL, NS
Cremon et al., 2016 (Italy, Spain, France, Croatia, Bosnia) [80]	To assess PEA effect on mast cell count/activation and symptoms in IBS patients	1. PEA effect on gastrointestinal pain or altered function 2. PEA effect on blood/plasma or other tissue biomarkers alterations	54 (PEA: 29; PLB: 25)	1. Visceral measurement (Symptom questionnaire) 2. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification, Immunohistochemistry, EIA, NGF Emax ImmunoAssay System, ELISA; eCBs/NAEs quantification, LC-APCI-MS; protein expression, Western blot) 3. AEs measurement (Clinical assessment)	1. Visceral measurement: (a) **Effect on abdominal pain/discomfort severity: time, ↓; treatment, ↓; treatment x time interaction,** ↓; responding patients: PEA vs. PLB, NS; (b) **Effect on abdominal pain/discomfort frequency: time,** ↓; **treatment,** ↓; treatment x time interaction, NS; Effect on (c) bloating severity and frequency, (d) bowel habit, (e) dyspeptic and gastro-esophageal reflux symptoms: treatment x time interaction, NS; (f) **Effect on general well-being: time,** ↑; treatment x time interaction, NS 2. Blood/plasma measurement: (a) **Effect on MC count over time: IBS > HC**; treatment, NS; gender, NS; centre, NS; bowel presentation, NS; time, NS; (b) Effect on immune cell/MC activation: IBS vs. HC, NS; treatment, NS; (c) **Effect on eCBs/AEs: OEA levels: IBS < HC**; **CB2 expression: IBS > HC**; AEA, 2-AG, PEA, OEA association with bowel habit: NS; **CB1**

					expression, FAAH levels: IBS-C > IBS-D, IBS-M; treatment, NS 3. AEs measurement: PEA vs. PLB, NS
D'Ascanio et al., 2021 (Italy) [57]	To assess PEA effect on olfactory function recovery in post-COVID-19 olfactory impairment	PEA effect on altered olfactory function	12 (PEA: 7; CTRL: 5)	Neurological measurement (Sniffin' sticks)	**Sniffin' Sticks score improvement (T1-T0): PEA > CTRL**
Di Nardo et al., 2024 (Italy) [87]	To assess PEA effect on pain and other symptoms in IBS patients	PEA effect on gastrointestinal pain or altered function	70 (PEA: 34; PLB: 36)	1. Visceral measurement (IBS-SSS, BSS) 2. AEs measurement (Direct interview)	1. Visceral measurement: (a) **number of complete remissions after 12 weeks (IBS-SSS < 75): PEA > PLB (IBS-D)**; PEA vs. PLB (IBS-C, IBS-M); (b) **abdominal pain frequency (from baseline to Week 12): PEA, ↓**; PLB, NS; (c) **total IBS-SSS**, (d) **pain intensity score**, (e) **life interference score (from baseline to Week 12): PEA, ↓; PLB, ↓;** (f) **total IBS-SS (Week 12)**, (g) **pain intensity score (Week 8, Week 12)**, (h) **pain frequency score (Week 8, Week 12): PEA < PLB**; (i) bowel habit changes: all comparisons, NS 2. AEs measurement: PEA vs. PLB, NS
Di Stadio et al., 2022 (Italy) [58]	To assess PEA effect on olfactory function recovery in post-COVID-19 COD	PEA effect on altered olfactory function	185 (PEA: 130; PLB: 55)	1. Neurological measurement (Sniffin' Sticks) 2. AEs measurement (Clinical assessment)	1. Neurological measurement: (a) **TDI score: PEA post > PEA pre, PLB post**; PLB post vs. PLB pre, NS; PEA pre vs. PLB pre, NS; (b) **TDI Sniffin' Score: PEA post > PLB post**; PEA pre vs. PLB pre, NS; (c) **TDI score recovery: PEA > PLB**; (d) **TDI score variation (PEA group): T3 > T0, T1**; **T0 > T2**; other comparisons, NS 2. AEs measurement: PEA vs. PLB, NS
Di Stadio et al., 2023a (Italy) [59]	To assess PEA effect on olfactory function recovery in post-COVID-19 olfactory impairment	PEA effect on altered olfactory function	130 (PEA: 94; PLB: 36)	Neurological measurement (Sniffin' sticks)	**Sniffin' Sticks score: PEA post > PEA pre, PLB post**; PLB post vs. PLB pre, NS; PEA pre vs. PLB pre, NS **Recovered anosmia patients at T1: PEA > PLB**

					Recovered parosmia patients at T1: PEA vs. PLB, NS
Di Stadio et al., 2023b (Italy) [66]	To assess PEA effect on olfactory function recovery in post-COVID-19 olfactory impairment	PEA effect on altered olfactory function	250 (PEAdaily: 50; PEAbid: 50; PEA+OT: 100; PLB+OT: 50)	Neurological measurement (Sniffin' sticks)	**Sniffin' Sticks score (≥ 3 points improvement, between-groups): PEA+OT > PEAdaily, PEAbid, PLB+OT** **Sniffin' Sticks score (≥ 3 points improvement, within-group): PLB+OT, PEAdaily, PEAbid: Month 3 > baseline**; other comparisons, NS; **PEA+OT: Month 3 > Month 2 > Month 1 > baseline** **Sniffin' Sticks score:** all comparisons, NS (baseline, Month 1); **PEA+OT, PEAdaily, PEAbid > PLB+OT (Month 2, Month 3)**; other comparisons, NS
Evangelista et al., 2018 (Italy) [44]	To assess PEA effect on neuropathic pain intensity and sleep quality in CTS patients	1. PEA effect on sleep 2. PEA effect on neuropathic pain	42 (PEA: 22; CTRL: 20)	1. Neuropsychiatric measurement (NRS, PSQI, PGIC) 2. Neurological measurement (NRS, Tinel's sign, Phalen's test) 3. AEs measurement (Clinical assessment)	1. Neuropsychiatric measurement: (a) **Sleep-wake rhythm (NRS)**: PEA vs. CTRL, NS (baseline, 90 days post-surgery); **PEA < CTRL (60 days pre-surgery)**; (b) **PSQI Total**, (c) **PSQI Sleep latency**, (d) **PSQI Sleep disturbances**, (e) **PSQI Overall sleep quality**, (f) **PSQI Troubles in daily activities**: PEA vs. CTRL, NS (baseline, 90 days post-surgery); **PEA < CTRL (60 days pre-surgery)**; (g) **PSQI Sleep duration**: PEA vs. CTRL, NS (baseline, 90 days post-surgery); **PEA > CTRL (60 days pre-surgery)**; (h) **PGIC: PEA > CTRL** 2. Neurological measurement: (a) **Hyperalgesia score [NRS, Δ(surgery-baseline)]: PEA > CTRL**; (b) **Wrist flexion mean time [Whalen's test, Δ(surgery-baseline)]: PEA > CTRL** 3. AEs measurement: PEA vs. CTRL, NS
Faig-Marti and Martìnez-Catassùs, 2017	To assess PEA effect on clinical and	PEA effect on neuropathic pain	61 (PEA: 30; PLB: 31)	1. Neurological measurement (ENG, Levine's.	1. Neurological measurement: (a) Durkan's test, (b) Phalen's test, (c) ENG data: PEA vs. PLB, NS; (d) Sensitive speed, (e) Sensitive peak amplitude, (f)

(Spain) [60] Faig-Martì and Martìnez-Catassùs, 2020 (Spain) [61]	electrophysiological changes in CTS patients			questionnaire, SSS, FSS, Durkan's test, Phalen's test, VAS) 2. AEs measurement (Clinical assessment)	Motor latency: PLB post-treatment vs. PLB pre-treatment, NS; (g) VAS: PEA vs. PLB, NS; PLB pre-treatment vs. PLB post-treatment, NS; (h) **FSS: PEA pre-treatment > PEA post-treatment**; PLB pre-treatment vs. PLB post-treatment, NS; (i) **SSS: PEA pre-treatment > PEA post-treatment**; **PLB pre-treatment > PLB post-treatment**; (j) **Levine's questionnaire: PLB post-treatment < PLB pre-treatment** 2. AEs measurement: PEA vs. PLB, NS
Gagliano et al., 2011 (Italy) [45]	To assess PEA effect on IOP in POAG/OH patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on sleep 3. PEA effect on intraocular pressure and visual field	42 (PEA: 21; PLB: 21)	1. Visceral measurement (BCVA, anterior and posterior segment findings, vertical and horizontal C/D ratio, Goldmann applanation tonometry, visual field) 2. Neuropsychiatric measurement (MDQ) 3. AEs measurement (Questionnaire)	1. Visceral measurement: (a) **ΔIOP: PEA, ↓**; PLB, NS; (b) **IOP (1 month, 2 months): PEA < PLB**; (c) ΔBCVA, (d) ΔCCT, (e) vertical C/D ratio, (f) ΔMD, (g) ΔPSD: PEA, NS; PLB, NS 2. Neuropsychiatric measurement (ΔMDQ): PEA, NS; PLB, NS 3. AEs measurement: PEA vs. PLB, NS
Germini et al., 2017 (Italy) [69]	To assess PEA effect on chronic pain in geriatric patients	PEA effect on joints, back, limbs, and chronic widespread pain and altered function	10 (PEA: 5; PLB: 5)	1. Somatic measurement (11-point VNS) 2. QoL measurement (BPFS) 3. AEs measurement (Clinical assessment)	1. Somatic measurement (**Effect on pain intensity**): **PEA > PLB (patient 2, patient 9)**; **PEA < PLB (patient 3, patient 5)**; PEA vs. PLB, NS (other patients 2. QoL measurement (**Effect on function impairment**): **PEA > PLB (patient 7)**; PEA vs. PLB, NS (other patients) 3. AEs measurement: PEA vs. PLB, NS
Ghazizadeh-Hashemi et al., 2018 (Iran) [46]	To assess PEA effect on depressive symptoms in MDD patients	PEA effect on mood, anxiety, and stress	54 (PEA: 27; PLB: 27)	1. Neuropsychiatric measurement (HDRS) 2. AEs measurement (Side-effect checklist,	1. Neuropsychiatric measurement: (a) **Effect on HDRS scores: time, ↓**; **treatment, ↓**; **time x treatment interaction, ↓**; (b) **Reduction in HDRS scores: PEA > PLB (week 2, week 4, week 6)**; (c)

				self-reports)	**HDRS improvement: PEA > PLB (week 6)**; (d) **HDRS score male subgroup (week 6): PEA < PLB**; (e) HDRS score female subgroup (week 6): PEA vs. PLB, NS; (e) suicide ideation: PEA vs. PLB, NS (baseline, week 6); (f) **Number of responders: PEA > PLB (week 6)**; PEA vs. PBL, NS (other timepoints); (g) **Number of remissions: PEA > PLB (week 6)**; PEA vs. PLB, NS (other timepoints) 2. AEs measurement (Frequency of adverse events): PEA vs. PLB, NS (daytime drowsiness, morning drowsiness, dizziness, slowed movement, tremor, increased appetite, nervousness, restlessness, skin rash, blurred vision, loss of appetite, fatigue, diarrhea, dry mouth, sore throat, tachycardia)
Giammusso et al., 2017 (Italy) [81]	To assess PEA effect on chronic pain in CP/CPPS patients	PEA effect on gynecological and genitourinary pain or altered function	44	1. Visceral measurement (NIH/CPSI, IIEF5, IPSS) 2. QoL measurement (NIH/CPSI) 3. AEs measurement (Clinical assessment)	1. Visceral measurement, 2. QoL measurement: (a) IPSS score pre-treatment: PEA vs. CTRL, NS; (b) **IPSS score post-treatment: PEA < CTRL**; (c) IIEF5 score pre-treatment, (d) IIEF5 score post-treatment: PEA vs. CTRL, NS; (e) **NIH/CPSI score: post-PEA < pre-PEA**; pre-CTRL vs. post-CTRL, NS; other comparisons, NS 3. AEs measurement: PEA vs. CTRL, NS
Guida et al., 2010 (Italy) [62]	To assess PEA effect on chronic neuropathic pain in compressive-type lumbar sciatica patients	PEA effect on neuropathic pain	636 (PEA: 427; PLB: 209)	1. Neurological measurement (VAS) 2. QoL measurement (RDQ) 3. AEs measurement (Clinical assessment)	1. Neurological measurement: (a) VAS baseline values: PEA(600 mg) vs. PEA(300 mg) vs. PLB, NS; (b) **ΔVAS(T21-baseline) values: PLB < PEA(300 mg) < PEA(600 mg)**; (c) **↑ Perceived efficacy, ↑ PEA dosage** 2. QoL measurement: (a) RDQ baseline values: PEA600 vs. PEA300 vs. PLB, NS; (b) **ΔRDQ(T21-baseline) values: PLB < PEA(300 mg) < PEA(600 mg)** 3. AEs measurement: PEA vs. PLB, NS

Isola et al., 2021 (Italy) [70]	To assess PEA effect on pain and inflammatory response in periodontitis patients	1. PEA effect on dental and periodontal inflammation 2. PEA effect on blood/plasma or other tissue biomarkers alterations	66 (PEA: 33; CTRL: 33)	1. Somatic measurement (PD, BOP, VAS, PI, CAL, GR) 2. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification) 3. AEs measurement (Clinical assessment)	1. Somatic measurement: (a) **PD: 180 days < baseline (PEA)**; 180 days vs. baseline, NS (CTRL); **PEA < CTRL (30, 60, 180 days)**; (b) **BOP: 180 days < baseline (both groups)**; **PEA < CTRL (15, 60, 180 days)**; (c) **PI: 180 days < baseline (CTRL)**; 180 days vs. baseline, NS (PEA); (d) **CAL: PEA > CTRL (30, 60 days)**; (e) **GR: PEA > CTRL (30, 60 days)**; (f) peak post-operative VAS score: 2h (PEA); 12h (CTRL); (g) **median VAS score: PEA < CTRL (6, 12, 24 hours)** 2. Other tissue measurement: (a) **GCF volume: 180 days < baseline (both groups)**; (b) **IL-1β levels: PEA < CTRL (15, 30, 60 days)**; (c) **IL-10 levels: PEA > CTRL (30, 60 days)**; (d) **TNF-α levels: PEA < CTRL (30 days)**; (e) **IL-1β/IL-10 ratio: PEA < CTRL (60, 180 days)** 3. AEs measurement: PEA vs. CTRL, NS
Kadanangode Narayanaswam et al., 2023 (India) [75]	To assess PEA effect on joint pain and inflammatory response in arthritis patients	1. PEA effect on joints, back, limbs, and chronic widespread pain and altered function 2. PEA effect on blood/plasma or other tissue biomarkers alterations	72 (PEA: 36; CTRL: 36)	1. Somatic measurement (WOMAC) 2. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification) 3. AEs measurement: Clinical assessment	1. Somatic measurement: (a) **pain score (WOMAC)**, (b) **joint stiffness (WOMAC)**, (c) **physical stiffness (WOMAC)**: baseline vs. Week 12, NS (CTRL); **Week 12 < baseline (PEA)** 2. Blood/plasma and/or other tissue measurement: (a) **TNF-α levels**, (b) **IL-1β levels**, (c) **IL-6 levels**, (d) **CRP levels:** baseline vs. Week 12, NS (CTRL); **Week 12 < baseline (PEA)** 3. AEs measurement: PEA vs. CTRL, NS
Khalaj et al., 2018 (Iran) [47]	To assess PEA effect as on language and behavior in ASD patients	PEA effect on psychotic symptoms and autistic behaviors	62 (PEA: 31; PLB: 31)	1. Neuropsychiatric measurement (ABC-C) 2. AEs measurement (Clinical assessment)	1. Neuropsychiatric measurement: (a) **ABC-C over time: PEA < PLB (irritability and hyperactivity)**; PEA vs. PLB, NS (other domains); (b) **ABC-C at 10 weeks: PEA < PLB (irritability, hyperactivity)**; PEA < PLB, trend effect (inappropriate speech); PEA vs. PLB, NS; (c) **ABC-C at 5 weeks: PEA < PLB**

					(hyperactivity); PEA < PLB, trend effect (stereotypic behavior, inappropriate speech); PEA vs. PLB, NS (other domains); (d) **ABC-C response rate at 10 weeks: PEA > PLB (hyperactivity, irritability, inappropriate speech)**; PEA vs. PLB, NS (other domains); (e) ABC-C response rate at 5 weeks: PEA vs. PLB, NS (all domains) 2. AEs measurement: PEA vs. PLB, NS
Lunardelli et al., 2019 (Italy) [48]	To assess PEA effect on POD in hip fractured patients	PEA effect on attention, awareness, and cognition	80 (PEA: 40; CTRL: 40)	1. Neuropsychiatric measurement (DOM scale) 2. AEs measurement (Clinical assessment)	1. Neuropsychiatric measurement: (a) **POD incidence: PEA < CTRL**; (b) **Reduction in POD severity: PEA > CTRL**; (c) POD duration: PEA vs. CTRL, NS 2. AEs measurement: PEA vs. CTRL, NS
Marini et al., 2013 (Italy) [71]	To assess PEA effect on pain in TMJ OA patients	PEA effect on joints, back, limbs, and chronic widespread pain and altered function	24 (PEA: 12; CTRL: 12)	1. Somatic measurement (Maximum active mouth opening, VAS) 2. AEs measurement (Self-administered questionnaire)	1. Somatic measurement: (a) **ΔVAS (day 14 - baseline): PEA > CTRL**; (b) **ΔMaximum Mouth Opening (day 14 -baseline): PEA > CTRL** 2. AEs measurement: PEA vs. CTRL, NS
Masek et al., 1974 (Czech Republic) [72]	To assess PEA effect on the incidence and severity of URTI	PEA effect on respiratory tract infections and allergy symptoms	1. First trial: 468 (PEA: 223; PLB: 221) 2. Second trial: 918	1. Somatic measurement (Clinical assessment) 2. QoL measurement (Disability assessment)	First trial: **Episodes of fever/headache/sore throat, Nasal stuffiness/discharge/cough: PEA < PLB**; **Total number of patients: PEA <. PLB (6 weeks)**; PEA vs. PLB, NS (8 weeks); **Total number of days of illness: PEA < PLB (6 weeks, 8 weeks)** Second trial: **Out-patients, In-patients, Total number of patients: PEA < PLB (6 weeks, 8 weeks)**; Average duration of illness/fever: PEA vs. PLB, NS
Murina et al., 2013 (Italy) [82]	To assess PEA effect on vestibulodynia in TENS-treated patients	PEA effect on gynecological and genitourinary pain or altered function	20 (PEA: 10; PLB: 10)	1. Visceral measurement (VAS, Marinoff dyspareunia scale, CPT) 2. AEs measurement	1. Visceral measurement: (a) Post-treatment VAS, (b) Post-treatment Marinoff dyspareunia scale: PEA vs. PLB, NS 2. AEs measurement: PEA vs. PLB, NS

				(Clinical assessment)	
Orefice et al., 2016 (Italy) [49]	To assess PEA effect on IFN-β1a-induced AEs in RR-MS patients	1. PEA effect on attention, awareness, and cognition 2. PEA effect on mobility, spasticity, and sensory-motor deficits 3. PEA effect on cutaneous pain, erythema, and eczema 4. PEA effect on blood/plasma or other tissue biomarkers alterations	29 (PEA: 15; PLB: 14)	1. Somatic measurement (VAS, erythema width) 2. QoL measurement (MSQoL-54) 3. Blood/plasma and/or other tissue measurement (Inflammatory markers quantification, eCBs/NAEs quantification) 4. Neurological measurement (EDSS) 5. Neuropsychiatric measurement (PASAT)	1. Somatic measurement: (a) **VAS**: PEA vs. PLB, NS (baseline); **PEA < PLB (month 6, month 12)**; (b) Erythema width: PEA vs. PLB, NS (baseline, month 6, month 12) 2. QoL measurement: (a) **Cognitive function: PEA > PLB (6 months, 12 months)**; (b) **Change in health: PEA > PLB (6 months)**; (c) other MSQoL domains: PEA vs. PLB, NS 3. Blood/plasma measurement: (a) **IFN-γ**, (b) **IL-17 serum levels**: PEA vs. PLB, NS (month 1); **PEA < PLB (month 3, month 6, month 12)**; (d) **PEA**, (e) **AEA plasma levels**: PEA vs. PLB, NS (month 1); **PEA > PLB (month 3, month 6, month 9, month 12)**; (g) ΔCT FAAH, (h) ΔCT NAAA: PEA vs. PLB, NS; (h) **FAAH mRNA expression (relative to month 1)**: PEA vs. PLB, NS (month 3, month 6, month 9); **PEA < PLB (month 12)**; (j) Correlations between PEA serum levels and other eCBs/NAEs serum levels: **↑ PEA, ↑ OEA, ↑ AEA (month 3, month 6, month 9, month 12)**; (k) Correlations between PEA serum levels and cytokines serum levels: **↑ PEA, ↓ IFN-γ (month 3, month 6, month 12)**; **↑ PEA, ↓ IL-17, ↓ TNF-α (month 3, month 6, month 12)** 4. Neurological measurement: PEA vs. PLB, NS (month 1, month 3, month 6, month 9, month 12) 5. Neuropsychiatric measurement: PEA vs. PLB, NS (month 1, month 6, month 12)
Ottaviani et al., 2018 (Italy) [63]	To assess PEA effect on neuropathic pain in BMS patients	PEA effect on neuropathic pain	35 (PEA: 18; PLB: 17)	1. Neurological measurement (NRS Scoring system)	1. Neurological measurement (Burning intensity over time): PEA vs. PLB, NS (baseline, 30 days); **PEA < PLB (60 days)**; PEA < PLB, NS (4 months after discontinuation)

				2. AEs measurement (Clinical assessment)	2. AEs measurement: PEA vs. PLB, NS
Palma et al., 2016 (Italy) [64]	To assess PEA effect on pulmonary function decline in ALS patients	PEA effect on altered muscle force and respiratory capacity	64 (PEA: 28; CTRL: 36)	Neurological measurement (FVC%, MRC scale, ulnar and phrenic nerve, CMAP amplitudes, ALSFRS-R scale)	**FVC%: baseline > 12 weeks > 24 weeks (CTRL)**; baseline vs. 12 weeks vs. 24 weeks, NS (PEA); MRC, CMAP amplitude, ALSFRS-R total: baseline vs. 12 weeks vs. 24 weeks, NS (both groups); **ALSFRS-R bulbar, ALSFRS-R respiratory: time x treatment effect (PEA)**; time x treatment effect (CTRL), NS; **Proportion of survivors (24 weeks): PEA > CTRL**
Pickering et al., 2022 (Australia) [50]	To assess PEA effect on PNP, sleep quality, and mood changes in diabetic patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on sleep 3. PEA effect on neuropathic pain 4. PEA effect on blood/plasma or other tissue biomarkers alterations	70 (PEA: 35; PLB: 35)	1. Neurological measurement (BPI-DPN, NPSI) 2. Neuropsychiatric measurement (Sleep quality assessment, MOS-Sleep; psychological assessment, DASS-21) 3. Blood/plasma and/or other tissue measurement (HbA1c, FBG, inflammatory markers quantification) 4. AEs measurement (Clinical assessment) 5. QoL measurement (BPI-DPN, NPSI, MOS)	1. Neurological measurement: (a) **effect on pain severity (BPI-PN): time, ↓**; **treatment, ↓**; (b) **pain severity (BPI-PN)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 2, week 4, week 8)**; (c) **pain interference (BPI-PN)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (d) **total pain score (NPSI)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (e) **superficial pain (NPSI)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (f) **deep pain (NPSI)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (g) **paroxysmal pain (NPSI)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (h) **evoked pain (NPSI)**: PEA vs. PLB, NS (baseline, week 4); PEA < PLB (week 8, trend effect); (i) **paraesthesia (NPSI)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (j) Influence of prescribed pain medications on treatment effect: NS 2. Neuropsychiatric measurement: (a**) sleep disturbance**: PEA vs. PLB, NS (baseline); PEA < PLB (week 4, trend effect); **PEA < PLB (week 8)**;

					(b) **sleep adequacy**: PEA vs. PLB, NS (baseline); **PEA > PLB (week 4, week 8)**; (c) sleep quantity: PEA vs. PLB, NS (baseline, week 4, week 8); (d) **daytime somnolence**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (e) snoring: PEA vs. PLB, NS (baseline, week 4, week 8); (f) **shortness of breath or headache**: PEA vs. PLB, NS (baseline, week 4); **PEA < PLB (week 8)**; (g) **sleep problem index**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; (h) **depressive symptoms change**: **PEA > PLB (week 8)**; (i) stress levels change: PEA > PLB (week 8, trend effect); (j) anxiety symptoms change: PEA vs. PLB, NS (week 8) 3. Blood/plasma measurement: (a) FBG, (b) HbA1c mmol/L, (c) HbA1c %, (d) CRP, (e) fibrinogen: PEA vs. PLB, NS (baseline, week 8); (f) **CRP > 5.0 mg/L**, (g) **IL6**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 8)** 4. AEs measurement: PEA vs. PLB, NS
Rao et al., 2021 (Australia) [51]	To assess PEA effect on sleep quality and quantity in DSP patients	1. PEA effect on sleep 2. PEA effect on blood/plasma or other tissue biomarkers alterations	103 (PEA: 55; PLB: 48)	1. Neuropsychiatric measurement (PSQI, wrist actigraphy, consensus sleep diary, PROMIS sleep disturbance questionnaire, SIQ, ESS) 2. QoL measurement (SF-36) 3. Blood/plasma and/or other tissue measurement (Pathology markers	1. Neuropsychiatric measurement: (a) PSQI scores improvement: PEA vs. PLB, NS (all time-points); (b) Sleep actigraphy data: Sleep quality (1-5), Total sleep time (h), Sleep time (h), Interruptions (min), Sleep Continuity (1-5), Sleep percentage (%): PEA vs. PLB, NS (all time-points); (c) **Sleep diary data: Sleep onset latency (min)**: PEA vs. PLB, NS (baseline); **PEA < PLB (week 4, week 8)**; Sleep time (h), Interruptions (n), Interruptions (min): PEA vs. PLB, NS (all time-points); (d): Data from sleep questionnaires: PEA vs. PLB (all time-points) 2. QoL measurement (SF-36): PEA vs. PLB, NS 3. Blood/plasma and/or other tissue measurement: PEA vs. PLB, NS

				quantification, safety markers quantification) 4. AEs measurement (Clinical assessment)	4. AEs measurement (reported gastrointestinal issues): PEA vs. PLB, NS
Rao et al., 2023 (Australia) [76]	To assess PEA effect on the incidence, duration, and severity of URTI	PEA effect on respiratory tract infections and allergy symptoms	426 (PEA: 213; PLB: 213)	1. Somatic measurement (URTI incidence, URTI duration, WURSS-24) 2. QoL measurement (SF-8) 3. AEs measurement (Clinical assessment)	1. Somatic measurement: (a) **incidence of URTI episodes**, (b) **number of participants sick at least once: PEA < PLB**; (c) duration of URTI episodes: PEA vs. PLB, NS; (d) **scratchy throat (WURSS-24)**, (e) **cough (WURSS-24): PEA < PLB**; (f) hoarseness (WURSS-24): PEA < PLB (trend effect); (g) breathe easily (WURSS-24): PEA > PLB (trend effect); (h) other symptoms (WURSS-24): PEA vs. PLB, NS; (i) number of people reporting symptoms (WURSS-24): PEA vs. PLB, NS 2. QoL measurement: all comparisons, NS 3. AEs measurement: PEA vs. PLB, NS
Rao et al., 2024 (Australia) [77]	To assess PEA effect on redness, dry skin, scaling, and itchiness in atopic eczema patients	PEA effect on cutaneous pain, erythema, and eczema	72 (PEA: 36; CTRL: 36)	1. Somatic measurement (SA-EASI, NRS, POEM, use of topical anti-inflammatory creams) 2. QoL measurement (DLQI) 3. AEs measurement (Clinical assessment)	1. Somatic measurement: (a) **redness (SA-EASI)**, (b) **dryness (SA-EASI)**, (c) **Total POEM outcome score: Week 4 < Week 2 < baseline (PEA, CTRL)**; **PEA < CTRL (Week 4)**; other comparisons, NS; (d) **POEM severity scoring: Week 4 < Week 2 < baseline (PEA, CTRL)**; (e) **thickness (SA-EASI)**, (f) **scratches (SA-EASI)**, (g) **itchiness (SA-EASI): Week 4, Week 2 < baseline (CTRL)**; **Week 4 < Week 2 < baseline (PEA)**; PEA vs. CTRL, NS 2. QoL measurement (DLQI): PEA vs. CTRL, NS (all comparisons) 3. AEs measurement: PEA vs. CTRL, NS
Rossi et al., 2020 (Italy) [83]	To assess PEA effect on RGCs function, IOP, visual field, and QoL in POAG or NTG patients	PEA effect on intraocular pressure and visual field	42 (Starting-with-PEA group: 21; Switching-to-PEA group: 19)	1. Visceral measurement (PERG, visual field examination, IOP	1. Visceral measurement: (a) **IOP values:** starting-with-PEA group vs. switching-to-PEA group, NS (baseline); **starting-with-PEA group < switching-to-PEA group** (**month 4)**; **starting-with-PEA**

				assessment) 2. QoL measurement (NEI-VFQ 25) 3. AEs measurement (Clinical assessment)	**group > switching-to-PEA group (month 8)**; (b) Visual field (MD, PSD): starting-with-PEA group vs. switching-to-PEA group, NS (baseline, month 4, month 8); (c) **PERG (P50-wave amplitude)**: starting-with-PEA group vs. switching-to-PEA group, NS (baseline); **starting-with-PEA group > switching-to-PEA group (month 4)**; **starting-with-PEA group < switching-to-PEA group (month 8)**; PERG (other measures): starting-with-PEA group vs. switching-to-PEA group, NS (baseline, month 4, month 8) 2. QoL measurement (**NEI-VFQ 25 Total mean**): starting-with-PEA group vs. switching-to-PEA group, NS (baseline); **starting-with-PEA group > switching-to-PEA group (month 4)**; **starting-with-PEA group < switching-to-PEA group (month 8)** 3. AEs measurement: starting-with-PEA group vs. switching-to-PEA group, NS
Salaffi et al., 2023 (Italy) [78]	To assess PEA effect on chronic pain and general wellbeing in FM patients	PEA effect on joints, back, limbs, and chronic widespread pain and altered function	130 (PEA: 62; CTRL: 68)	1. Somatic measurement (WPI) 2. QoL measurement (FIQR, FASmod) 3. AEs measurement (Clinical assessment)	1. Somatic measurement (**WPI, AUC**): **PEA < CTRL** 2. QoL measurement: (a) **FIQR (AUC)**, (b) **FASmod (AUC): PEA < CTRL** 3. AEs measurement: PEA vs. CTRL, NS
Salehi et al., 2022 (Iran) [52]	To assess PEA effect on negative symptoms in SCZ patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on psychotic symptoms and autistic behaviors	60 (PEA: 30; PLB: 30)	1. Neuropsychiatric measurement (PANSS, HDRS) 2. AEs measurement (ESRS, open-ended questions, comprehensive side effect checklist)	1. Neuropsychiatric measurement: (a) **Effect on PANSS negative: time, ↓; time x treatment interaction, ↓**; (b) **Effect on PANSS positive: time, ↓**; time x treatment interaction, NS; (c) **Effect on PANSS general: time, ↓; time x treatment interaction, ↓**; (d) **Effect on PANSS total: time, ↓; time x treatment interaction, ↓**; (e) Effect on HDRS: time x treatment interaction, NS

					2. AEs measurement: (a) Effect on ESRS global score: time, NS; time x treatment interaction, NS; (b) Frequency of adverse events (drowsiness, dizziness, tremor, increased appetite, nervousness, restlessness, skin rash, blurred vision, fatigue, diarrhea, dry mouth, sore throat, tachycardia): PEA vs. PLB, NS
Steels et al., 2018 (Australia) [53]	To assess PEA effect on pain in knee OA patients	1. PEA effect on mood, anxiety, and stress 2. PEA effect on sleep 3. PEA effect on joints, back, limbs, and chronic widespread pain and altered function 4. PEA effect on blood/plasma or other tissue biomarkers alterations	111 (PEA: 71; PLB: 40)	1. Somatic measurement (WOMAC; NRS; use of rescue medication) 2. Neuropsychiatric measurement (DASS, PSS, PSQI) 3. QoL measurement (SF-36, WOMAC function subdomain) 4. AEs measurement (Clinical assessment) 5. Blood/plasma and/or other tissue measurement	1. Somatic measurement: (a) **WOMAC total score:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 4); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (b) **WOMAC pain score:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 1, week 4); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (c) **WOMAC stiffness score:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 4); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (d) **WOMAC function score:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 4); PEA(300 mg) vs. PLB, NS (week 8); **PEA(600 mg) < PLB (week 8)**; (e) **NRS worst pain yesterday:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 4); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (f) **NRS least pain yesterday:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 4); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (g) **NRS average pain**: PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline); **PEA(300 mg), PEA(600 mg) < PLB (week 1, week 4, week 8)** 2. Neuropsychiatric measurement: (a) **DASS anxiety score:** PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline); **PEA(300 mg), PEA(600 mg) < PLB (week 8)**; (b) DASS depression score: PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS

					(baseline, week 8); (c) PSI stress score: PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 8); (d) sleep quality (PSQI, SF-36): PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 8) 3. Blood/plasma and/or other tissue measurement (Biochemical, hematological, and inflammatory parameters): PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 8) 4. AEs measurement: PEA(300 mg) vs. PEA(600 mg) vs. PLB, NS (baseline, week 8)
Strobbe et al., 2013 (Italy) [84]	To assess PEA effect on systemic endothelial function in OH patients	PEA effect on intraocular pressure and visual field	80 (PEA: 40; PLB: 40)	1. Visceral measurement (Brachial artery FMD, IOP) 2. AEs measurement (Clinical assessment)	1. Visceral measurement: (a) **FMD values: T1, T2, T3 > T0 (starting-with-PEA group)**; other comparisons, NS (starting-with-PEA group); **T3 > T0, T1, T2**; other comparisons, NS (switching-to-PEA group); (b) **IOP values: T1, T2, T3 < T0 (starting-with-PEA group)**; other comparisons, NS (starting-with-PEA group); **T3 < T0, T1, T2 (switching-to-PEA group)**; other comparisons, NS (switching-to-PEA group) 2. AEs measurement: starting-with-PEA group vs. switching-to-PEA group, NS
Tartaglia et al., 2015 (Italy) [85]	To assess PEA effect on pelvic pain in primary dysmenorrhea patients	PEA effect on gynecological and genitourinary pain or altered function	220 (PEA: 110; PLB: 110)	1. Visceral measurement (Questionnaire, 10-point VAS) 2. AEs measurement (Clinical assessment)	1. Visceral measurement (VAS score): (a) **improvement of pelvic pain: PEA > PLB**; (b) **mean improvement of pelvic pain: PEA > PLB** 2. AEs measurement: PEA vs. PLB, NS
Versace et al., 2023 (Italy) [54]	To assess PEA effect over reduced LICI in patients with post-COVID-19 cognitive dyfunction and fatigue	PEA effect on attention, awareness, and cognition	39 (PEA: 17; PLB: 17)	Neuropsychiatric measurement (LICI protocol, SAI, LTP-like cortical plasticity, MoCA, FAB)	**Effect on % change of test MEP (LICI): time x treatment interaction**; **% of test MEP (LICI): PEA group: POST < PRE**; PLB group: POST vs. PRE, NS; Effect on % change of test MEP (SAI): time x treatment interaction, NS; % of test MEP (SAI): POST vs. PRE, NS (PEA

					group, PLB group); **Effect on MEP amplitude (LTP-like cortical plasticity): time x treatment interaction (1 and 10 min following iTBS)**; time x treatment interaction, NS (20 min following iTBS); **Mean amplitude of MEPs (LTP-like cortical plasticity): PEA group: POST > PRE (1 and 10 min following iTBS)**; POST vs. PRE, NS (20 min following iTBS); PLB group: all comparisons, NS RMT protocol: all comparisons, NS Effect on cognitive performance and executive functions (MoCA, FAB): treatment x time interaction, NS
Yuan et al., 2014 (China) [73]	To assess PEA effect on dry skin and itchy sensation in asteatotic eczema patients	PEA effect on cutaneous pain, erythema, and eczema	66 (PEA: 30; CTRL: 30)	1. Somatic measurement (EASI, skin hydration, TEWL, CPT) 2. AEs measurement (Clinical assessment)	1. Somatic measurement: (a) Symptom improvement: **skin scaling**, **dryness**, **itching**: **PEA > CTRL (28 days)**; other symptoms: PEA vs. CTRL, NS; (b) Skin barrier function: **ΔCapacitance: PEA > CTRL (3, 7, 14, 28 days)**; ΔTEWL: PEA vs. CTRL, NS (all time-points); (c) **CPT**: **PEA > CTRL (14 days)**; other time-points: PEA vs. CTRL, NS 2. AEs measurement: PEA vs. CTRL, NS

↑, Increased; ↓, Decreased; <, less/smaller than; >, more/greater than; ≥, more/greater than or equal to; 2-AG, 2-Arachidonoylglycerol; ABC-C, Aberrant Behavior Checklist-Community; AEA, Anandamide; AEs, Adverse Events; AIS, Acute Ischemic Stroke; ALS, Amyotrophic Lateral Sclerosis; ALA, Alpha-lipoic acid; ALSFRS-R, ALS Functional Rating Scale–Revised; AR, Allergic Rhinitis; ASD, Autism Spectrum Disorder; AUC, Area Under the Curve; BCTQ, Boston Carpal Tunnel Questionnaire; BCVA, Best-corrected visual acuity; BDI, Beck Depression Inventory; BMS, Burning mouth syndrome; BNCE, Brief Neuropsychological Cognitive Examination; BOP, Bleeding on probing; BPAD, Bipolar Affective Disorder; BPFS, Back Pain Functional Scale; BPI-DPN, Brief Pain Inventory-Diabetic Peripheral Neuropathy; BSS, Bristol Stool Scale; C/D, Cup-to-disc; CA-125, Cancer Antigen 125; CAL, Clinical attachment level; CB1, Cannabinoid receptor type 1; CB2, Cannabinoid receptor type 2; CCT, Central corneal thickness; CD3, Cluster of differentiation 3 lymphocytes; CD8, Cluster of differentiation 8 lymphocytes; CMAP, Compound Muscle Action Potential; COD, Chronic olfactory dysfunction; Co-ultraPEALut, Co-ultramicronized PEA plus Luteolin; COVID-19, SARS-CoV-2 infection; CP/CPPS, Chronic Prostatitis/Chronic Pelvic Pain syndrome; CPSS, Cincinnati Prehospital Stroke Scale; CPT, Current perception threshold; CRP, C-Reactive Protein; CTRL, Control; CTS, Carpal Tunnel Syndrome; DASS, Depression Anxiety and Stress Scale; DLQI, Dermatology Quality of Life Index; DOM, Delirium-O-Meter; DSP, Disturbed Sleeping Pattern; EASI, Eczema Area and Severity Index; eCBs/NAEs, Endocannabinoids/N-

Acylethanolamines; EDSS, Expanded Disability Status Scale; EIA, Enzyme immunoassay; ELISA, Enzyme-linked immunosorbent assay; EMS, Endometriosis; ENG, Electroneurography; ESR, Erythrocyte Sedimentation Rate; ESRS, Extrapyramidal Symptom Rating Scale; ESS, Epworth Sleepiness Scale; FAAH, Fatty Acid Amide Hydrolase; FAB, Frontal Assessment Battery; FASmod, FBG, Fasting Blood Glucose; FIQR, Fibromyalgia Impact Questionnaire; FM, Fibromyalgia; FMD, Flow-mediated vasodilation; FORD, Free Oxygen Radical Defense; FORT, Free Oxygen Radical Test; FSS, Functional Status Scale; FVC, Forced Vital Capacity; GAD-10, Generalized Anxiety Disorder Assessment; GCF, Gingival crevicular fluid; GCS, Glasgow Coma Scale; GFR, Glomerular Filtration Rate; GR, Gingival recessions; h, hours; HbA1c, Glycated Haemoglobin; HC, Healthy controls; HDRS, Hamilton Depression Rating Scale; IBS, Irritable Bowel Syndrome; IBS-C, IBS with predominant constipation; IBS-D, IBS with predominant diarrhoea; IBS-M, IBS with mixed symptoms; IBS-SSS, IBS-Severity Scoring System; IFN-β1a, Interferon-Beta1a; IFN-γ, Interferon-Gamma; IgG, Immunoglobulin G; IIEF5, International Index of Erectile Function 5; IL-4, Interleukin-4; IL-8, Interleukin-8; IL-10, Interleukin-10; IL-17, Interleukin-17; IL-1β, Interleukin-1Beta; IL-6, Interleukin-6; ILTM, Impacted lower third molar; IOP, Intra-ocular pressure; IPSS, International Prostatic Symptoms Score; ISI, Insomnia Severity Index; iTBS, Intermittent theta burst stimulation; ITT, Intention-to-treat; LC-APCI-MS, Liquid chromatography-atmospheric pressure chemical-ionization-mass spectrometry; LICI, Long-interval intracortical inhibition; LTP, Long-term potentiation; LWT, Last Week of Treatment; MC, Mast cells; MCV, Mean Corpuscular Volume; MD, Mean defect; MDD, Major Depressive Disorder; MDI, Major Depression Inventory; MDQ, Mood Disorder Questionnaire; MEP, Motor evoked potential; min, minutes; MMSE, Mini Mental State Examination; MoCA, Montreal Cognitive Assessment; MOS, Medical Outcome Study; MRC, Medical Research Council; mRNA, Messenger RNA; mRS, Modified Rankin Scale; MSQoL-54, Multiple Sclerosis Quality of Life-54; MTS, Modified Tardieu Scale; NAAA, N-Acylethanolamine Acid Amidase; NEI-VFQ 25, 25 item National Eye Institute Visual Function Questionnaire; NGF, Nerve Growth Factor; NIH/CPSI, National Institutes of Health Chronic Prostatitis Symptom Index; NIHSS, National Institutes of Health Stroke Scale; NNT, Number Needed to Treat; NPSI, Neuropathic Pain Symptom Inventory; NRS, Numeric Rating Scale; NS, Not Significant; NSAIDs, Nonsteroidal anti-inflammatory drugs; NTG, Normal Tension Glaucoma; OA, Osteoarthritis; OEA, Oleoylethanolamide; OH, Ocular hypertension; OT, Olfactory Training; PANSS, Positive and Negative Syndrome Scale; PASAT, Paced Auditory Serial Addition Test; PD, Probing depth; PEA, Palmitoylethanolamide; PERG, Pattern electroretinogram; PGIC, Patient Global Impression of Change; PI, Plaque score; PLB, Placebo; PLT, Platelet; PNP, Peripheral neuropathy; POAG, Primary open glaucoma; POD, Post-operative delirium; POEM, Patient-Oriented Eczema Measure; PP, Per protocol; PROMIS, Patient Reported Outcomes Measurement Information System; PSD, Pattern standard deviation; PSQI, Pittsburgh Sleep Quality Index; PSS, Perceived Stress Scale; PT, Prothrombin Time; QoL, Quality of Life; RDQ, Rolant-Morris disability questionnaire; RGCs, Retinal ganglion cells; RR-MS, Relapsing-Remitting Multiple Sclerosis; rTNSS, Reflective Total Nasal Symptom Score; S-TOPS, Pain Survey; SA-EASI, Self-Assessed Eczema Area and Severity Index; SAI, Short-latency sensory afferent inhibition; SAP, Sensory action potentials; SCI, Spinal Cord Injury; SCZ, Schizophrenia; SF-8, 8-item Health Survey; SF-36, 36-item Health Survey; SIQ, The Sleep Inertia Questionnaire; SNCV, Sensory nerve conduction velocity; SSS, Symptom Severity Scale; TBI, Traumatic Brain Injury; TDI, Threshold, Discrimination and Identification subtests; TENS, Transcutaneous electrical nerve stimulation; TEWL, Transepidermal water loss; TMJ, Temporo-mandibular joint; Tn, Timepoint; TNF-α, Tumor Necrosis Factor-Alpha; URTI, Upper Respiratory Tract Infections; US, Ultrasound; VAS, Visual Analogue Scale; VNS, Visual Numeric Scale; vs., compared to; WOMAC, Western Ontario and McMaster Universities Arthritis Index; WPI, Widespread Pain Index; WURSS-24, Wisconsin Upper Respiratory Symptom Survey; YMRS, Young Mania Rating Scale; Δ, Difference between values; ΔCT, The Comparative Ct Method. Bold font emphasizes statistically significant results.

Table 2. Methodological characteristics of Randomized Controlled Trials investigating Palmitoylethanolamide supplementation across human diseases

Study ID	Level of concealment	Defined population	Age (years, mean ± SD)	Gender (count, %)	PEA measure (dosage, formulation, intake route)	PEA adequate evaluation (period of exposure, use as monotherapy or add-on)
Abedini et al., 2022 (Iran) [41]	Double-blind	Bipolar affective disorder with acute mania (SCID-5, MINI)	PEA: 30.8 ± 9.8 PLB: 32.7 ± 9.0	PEA: Female 9 (28.1%)/Male 23 (71.9%) PLB: Female 11 (35.5%)/Male 20 (64.5%)	Undeclared PEA formulation (600 mg/bid, capsule, oral)	6 weeks, add-on to lth and rsp
Albanese et al., 2022 (Italy) [88]	X	COVID-19 (positive nasopharyngeal swab for SARS-CoV-2)	PEA: 45.6 ± 13.7 CTRL: 55.8 ± 22.5	PEA: Female 28 (62.2%)/Male 17 (37.8%) CTRL: Female 23 (51.1%)/Male 22 (48.9%)	mPEA + umPEA (900 mg/bid, microgranules, sublingual)	4 weeks, add-on to TAU
Andresen et al., 2016 (Denmark, Norway, United Kingdom) [42]	Double-blind	Neuropathic pain following spinal cord injury (SCI-compatible pain distribution, MRI)	PEA: 58.6 ± 11.3 PLB: 54.1 ± 11.7	PEA: Female 14 (39.0%)/Male 22 (61.0%) PLB: Female 5 (14.0%)/Male 32 (86.0%)	umPEA (600 mg/bid, microgranules, sublingual)	12 weeks, add-on to TAU
Bacci et al., 2011 (Italy) [68]	Single-blind	Impacted lower third molars (X-Rays, Pell and Gregory classification)	X	X	mPEA (300 mg/bid, tablet, oral)	2 weeks, monotherapy (6 days before surgery, 9 days afterwards)
Bonzanino et al., 2024 (Italy) [55]	Open-label	Acute ischemic stroke (NIHSS)	Mean age of the sample: 77.9 ± 10.3	Female 27 (45.0%)/Male 33 (55.0%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/bid, suspension, oral)	12 weeks, add-on to TAU (starting within

						72 hours after ischemic event)
Briskey et al., 2023 (Australia) [74]	Double-blind	Allergic rhinitis (rTNSS)	PEA: 45.6 ± 15.6 PLB: 45.3 ± 12.8	PEA: Female 34 (65.4%)/Male 18 (34.6%) PLB: Female 38 (77.6%)/Male 11 (22.4%)	LipiSperse PEA (300 mg/day, capsule, oral)	2 weeks, monotherapy
Briskey et al., 2024 (Australia) [67]	Double-blind	Migraine (ICHD3)	PEA: 41.2 ± 11.5 PLB: 43.8 ± 10.8	PEA: Female 35 (87.5%)/Male 5 (12.5%) PLB: Female 35 (87.5%)/Male 5 (12.5%)	LipiSperse PEA (300 mg/bid, capsule, oral)	8 hours (one dose upon commencement of migraine symptoms, a second dose if migraine had not resolved 2 hours post-dose), monotherapy
Campolo et al., 2021 (Italy) [43]	Single-blind	Traumatic Brain Injury (GCS)	PEA: 49.7 ± 17.9 CTRL: 53.8 ± 17.5	PEA: Female 2 (13.3%)/Male 13 (86.7%) CTRL: Female 4 (26.7%)/Male 11 (73.3%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/bid, tablet, oral)	26 weeks, add-on to TAU
Cantone et al., 2024 (Italy) [65]	Double-blind	Post-COVID-19 anosmia/hyposmia and parosmia (Positive nasopharyngeal swab for SARS-CoV-2)	Co-ultraPEALut: 44.8 ± 12.2 Co-ultraPEALut/ALA: 42 ± 10.3 ALA: 35.9 ± 12.3 PLB: 52.1 ± 11.8	Co-ultraPEALut: Female 11 (64.7%)/Male 6 (35.3%) Co-ultraPEALut/ALA: Female 18 (64.3%)/Male 10 (35.7%) ALA: Female 9 (42.9%)/Male 12 (57.1%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/day, sachet, oral) Co-ultraPEALut/ALA (umPEA 700 mg + luteolin 70 mg + ALA 600 mg/day, sachet, oral)	12 weeks, monotherapy (after OT)

				PLB: Female 13 (56.5%)/Male 10 (43.5)		
Cobellis et al., 2011 (Italy) [79]	Double-blind	Endometriosis (ESHRE, laparoscopic conservative surgery, histological examination)	PEA: 26–37 PLB: 25–41 Celecoxib: 24–35	N/A	mPEA/transpolydatin (mPEA 400 mg + transpolydatin 40 mg/bid, tablet, oral)	12 weeks, monotherapy
Coraci et al., 2018 (Italy) [56]	Double-blind	Minimum/mild idiopathic Carpal tunnel syndrome (Padua's classification)	X	X	Undeclared PEA formulation (600 mg/day, tablet, oral)	4 weeks, monotherapy
Costagliola et al., 2014 (Italy) [86]	Open-label	Normal tension glaucoma (Clinical assessment, IOP, BCVA, visual field test, GHT)	Mean age of the sample: 53.7 (31–67)	PEA: Female 10 (62.5%)/Male 6 (37.5%) CTRL: Female 10 (62.5%)/Male 6 (37.5%)	umPEA (300 mg/bid, tablet, oral)	24 weeks, monotherapy
Cremon et al., 2016 (Italy, Spain, France, Croatia, Bosnia) [80]	Double-blind	Irritable bowel syndrome (Clinical assessment)	PEA: 37.0 ± 10.8 PLB: 40.4 ± 9.8	PEA: Female 18 (62.1%)/Male 11 (37.9%) PLB: Female 11 (44.0%)/Male 14 (56.0%)	mPEA/polydatin (mPEA 200 mg + polydatin 20 mg/day, tablet, oral)	12 weeks, monotherapy
D'Ascanio et al., 2021 (Italy) [57]	Single-blind	Post-COVID-19 anosmia/hyposmia (Positive nasopharyngeal swab for SARS-CoV-2, Sniffin' Sticks)	42.2 ± 14.1	PEA: Female 5 (71.4%)/Male 2 (28.6%) PLB: Female 3 (60.0%)/Male 2 (40.0%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/day, tablet, oral)	4 weeks, monotherapy (after OT)
Di Nardo et al., 2024 (Italy) [87]	Double-blind	Irritable bowel syndrome (Rome IV criteria)	PEA: 13.7 ± 2.3 PLB: 13.8 ± 2.1	PEA: Female 18 (52.9%)/Male 16 (47.1%)	mPEA/polydatin (mPEA 200 mg + polydatin 20 mg/tid, tablet, oral)	12 weeks, monotherapy

				PLB: Female 20 (55.6%)/Male 16 (44.4%)		
Di Stadio et al., 2022 (Italy) [58]	Double-blind	Post-COVID-19 COD (Positive nasopharyngeal swab for SARS-CoV-2, Sniffin' Sticks)	PEA: 42.1 ± 14.5 PLB: 47.0 ± 14.6	PEA: Female 83 (63.8%)/Male 47 (36.2%) PLB: Female 38 (69.0%)/Male 17 (31.0%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/day, tablet, oral)	12 weeks, monotherapy (prior to OT)
Di Stadio et al., 2023a (Italy) [59]	Double-blind	Post-COVID-19 anosmia/parosmia (Positive nasopharyngeal swab for SARS-CoV-2, 16-pen version of the Sniffin' Sticks)	PEA: 36.7 ± 11.8 PLB: 50.5 ± 12.7	PEA: Female 49 (52%)/Male 45 (48%) PLB: Female 21 (58%)/Male 15 (42%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/day, tablet, oral)	12 weeks, monotherapy (prior to OT)
Di Stadio et al., 2023b (Italy) [66]	Double-blind	Post-COVID-19 anosmia/hyposmia (Positive nasopharyngeal swab for SARS-CoV-2, Sniffin' Sticks)	PEAdaily: 39.8 ± 11.5 PEAbid: 37.1 ± 13.9 PEA+OT: 42.5 ± 13.5 PLB+OT: 40.9 ± 11.7	PEAdaily: Female 18 (37.5%)/Male 30 (62.5%) PEAbid: Female 24 (60.0%)/Male 16 (40.0%) PEA+OT: Female 40 (71.4%)/Male 16 (28.6%) PLB+OT: Female 26 (68.4%)/Male 12 (31.6%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/day or /bid, sachet, oral)	12 weeks, monotherapy or add-on to OT
Evangelista et al., 2018 (Italy) [44]	Open-label	Carpal Tunnel Syndrome (Clinical Practice Guidelines, AAOS)	PEA: 57.4 ± 11.5 CTRL: 56.7 ± 17.2	PEA: Female 17 (77.3%)/Male 5 (22.7%)	1. umPEA (600 mg/bid, microgranules, sublingual) 2. umPEA (600 mg/day, tablet, oral)	1. 60 days prior to surgery: (a) 10 days (sublingual), monotherapy; (b) 50

				CTRL: Female 11 (55.0%)/Male 9 (45.0%)		days (oral), monotherapy 2. After surgery: (a) 60 days (oral), monotherapy; (b) 30 days (oral), monotherapy
Faig-Marti and Martìnez-Catassùs, 2017 (Spain) [60] Faig-Martì and Martìnez-Catassùs, 2020 (Spain) [61]	Double-blind	Mild/moderate Carpal Tunnel Syndrome (ENG)	PEA: 51.8 ± 11.1 PLB: 53.3 ± 13.4	PEA: Female 18 (60.0%)/Male 12 (40.0%) PLB: Female 19 (61.3%)/Male 12 (38.7%)	Undeclared PEA formulation (600 mg/day, tablet, oral)	8 weeks, monotherapy
Gagliano et al., 2011 (Italy) [45]	Double-blind	Primary open angle glaucoma/Ocular hypertension (IOP, Clinical assessment)	Group A: 65.0 ± 12.0 Group B: 63.0 ± 11.0	Group A: Female 17 (81.0%)/Male 4 (19.0%) Group B: Female 14 (66.7%)/Male 7 (33.3%)	Undeclared PEA formulation (600 mg/day, tablet, oral)	Group A: 8 weeks, monotherapy, before 4-week washout and 8-week PLB Group B: 8 weeks, monotherapy, after 4-week washout and 8-week PLB
Germini et al., 2017 (Italy) [69]	Double-blind (N-of-1 trial)	Chronic non-cancer non-ischemic pain in the back, joints, or limbs (Clinical assessment)	PEA: 83.2 ± 4.6 PLB: 83.2 ± 4.6	PEA: Female 5 (100%) PLB: Female 5 (100%)	umPEA (600 mg/bid, tablet, oral)	3 weeks, monotherapy (prior to/after a 2-week wash-out period)
Ghazizadeh-Hashemi et al., 2018 (Iran) [46]	Double-blind	Major Depressive Disorder (SCID-5)	PEA: 35.4 ± 7.1 PLB: 33.9 ± 6.6	PEA: Female 8 (30.0%)/Male 19 (70.0%) PLB: Female 11 (40.0%)/Male 16 (60.0%)	umPEA (600 mg/bid, capsule, oral)	6 weeks, add-on to ctp

Giammusso et al., 2017 (Italy) [81]	Single-blind	Chronic prostatitis/chronic pelvic pain syndrome (IPSS, NIH-CPSI, PSA)	Mean age of the sample: 41.32 ±1.686	X	PEA/ALA (PEA 300 mg + ALA 300 mg/day, capsule, oral)	12 weeks, monotherapy
Guida et al., 2010 (Italy) [62]	Double-blind	Radicular and/or core compression of the sciatic nerve and discopathy (Clinical assessment)	PEA(300 mg): 42.0 ± 10.7 PEA(600 mg): 43.0 ± 11.4 PLB: 43.6 ± 11.5	PEA(300 mg): Female 98 (46.2%)/Male 114 (53.8%) PEA(600 mg): Female 99 (46.0%)/Male 116 (54.0%) PLB: Female 103 (49.3%)/Male 106 (50.7%)	mPEA (300 mg/day or 300 mg/bid, capsule, oral)	3 weeks, monotherapy
Isola et al, 2021 (Italy) [70]	Double-blind	Periodontitis (Clinical assessment)	Mean age of the sample: 47.8	Total sample: Female 32 (48.5%)/Male 34 (51.5%)	PEA/baicalin (PEA 800 mg + baicalin 400 mg/bid, capsule, oral)	5 days, add-on to SRP
Kadanangode Narayanaswam et al., 2023 (India) [75]	Open-label	Arthritis (Clinical assessment, DAS 28)	PEA: 41.73 ± 9.85 CTRL: 45.83 ± 6.68	X	PEA/astaxanthin (PEA 800 mg + astaxanthin 2 mg/day, tablet, oral)	12 weeks, add-on to TAU
Khalaj et al., 2018 (Iran) [47]	Double-blind	Autism Spectrum Disorder (DSM-5)	PEA: 6.84 ± 2.1 PLB: 7.42 ± 2.35	PEA: Female 9 (29.0%)/Male 22 (71.0%) PLB: Female 6 (19.4%)/Male 25 (80.6%)	Undeclared PEA formulation (600 mg/bid, capsule, oral)	10 weeks, add-on to rsp
Lunardelli et al., 2019 (Italy) [48]	Single-blind	Post-operative Delirium (DSM-5, CAM, 4AT test, DOM scale)	PEA: 87.2 ± 5.2 CTRL: 87.3 ± 5.6	PEA: Female 33 (82.5%)/Male 7 (17.5%) CTRL: Female 37 (92.5%)/Male 3 (7.5%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/bid, tablet, oral)	Up to 3 days after surgery, monotherapy (at 12-h intervals, not later than 12 h from admission)

Marini et al., 2013 (Italy) [71]	Triple-blind	Temporo-mandibular disorder (RDC/TMD)	X	Total sample: Female 16 (66.7%)/Male 8 (33.3%)	Undeclared PEA formulation (300 mg + 600 mg/day, 300 mg/bid, tablet, oral)	2 weeks, monotherapy (900 mg/day for 7 days, 600 mg/day for 7 days)
Masek et al., 1974 (Czech Republic) [72]	Double-blind	Upper respiratory tract infections (temperature ≥ 37.5°C, headache, sore throat, myalgia, nasal stuffiness, or discharge, productive or dry cough, malaise, fatigue)	First trial: PEA: 37.4 ± 1.5 PLB: 35.4 ± 1.26; Second trial: X	First trial: PEA: Female 125 (66.0%)/Male 98 (34.0%) PLB: Female 136 (61.5%)/Male 85 (38.5%); Second trial: X	Impulsin (300 mg x2/tid, tablet, oral)	12 days, monotherapy
Murina et al., 2013 (Italy) [82]	Double-blind	Vestibulodynia (≥ 6 months history of vulvar pain upon tampon insertion or attempted intercourse, positive cotton swab test result)	PEA: 34.4 (18-48) PLB: 31.8 (23-47)	N/A	PEA/transpolydatin (PEA 400 mg + transpolydatin 40 mg/bid, tablet, oral)	8 weeks, add-on to TENS
Orefice et al., 2016 (Italy) [49]	Double-blind	RR-MS (McDonald's 2010 criteria)	PEA: 30.60 ± 7.60 PLB: 28.93 ± 4.86	PEA: Female 6 (40.0%)/Male 9 (60.0%) PLB: Female 9 (64.0%)/Male 5 (36.0%)	umPEA (600 mg/day, tablet, oral)	53 weeks, monotherapy
Ottaviani et al., 2019 (Italy) [63]	Double-blind	Burning mouth syndrome (IHCC)	X	PEA: Female 13 (72.2%)/Male 5 (27.8%) PLB: Female 16 (94.1%)/Male 1 (5.9%)	umPEA (600 mg/bid, microgranules, sublingual)	8 weeks, monotherapy

Palma et al., 2016 (Italy) [64]	Single-blind	Amyotrophic lateral sclerosis (El Escorial diagnostic criteria)	PEA: 65.3 ± 10.2 CTRL: 62.1 ± 9.5	PEA: Female 12 (42.9%)/Male 16 (57.1%) CTRL: Female 17 (47.2%)/Male 19 (52.8%)	umPEA (600 mg/bid, tablet, oral)	24 weeks, add-on to rlz
Pickering et al., 2022 (Australia) [50]	Quadruple-blind	Peripheral neuropathy in diabetic patients (Clinical assessment, S-LANSS, DN4)	PEA: 65.5 (53–79) PLB: 61.5 (32–75)	PEA: Female 13 (39.4%)/Male 20 (60.6%) PLB: Female 18 (54.5%)/Male 15 (45.5%)	LipiSperse PEA (300 mg/bid, capsule, oral)	8 weeks, monotherapy
Rao et al., 2021 (Australia) [51]	Double-blind	Disturbed sleeping pattern (PSQI)	X	PEA: Female 30 (54.5%)/Male 25 (45.5%) PLB: Female 34 (70.8%)/Male 14 (29.2%)	LipiSperse PEA (150 mg/bid, capsule, oral)	8 weeks, monotherapy (1 h prior to sleep)
Rao et al., 2023 (Australia) [76]	Double-blind	Upper respiratory tract infections (Clinical assessment)	PEA: 40.0 ± 12.5 PLB: 38.9 ± 11.7	PEA: Female 28 (52.8%)/Male 25 (47.2%) PLB: Female 29 (53.7%)/Male 25 (46.3%)	LipiSperse PEA (300 mg/bid, capsule, oral)	12 weeks, monotherapy
Rao et al., 2024 (Australia) [77]	Double-blind	Atopic dermatitis (Clinical assessment)	PEA: 37.4 ± 11.7 CTRL: 36.0 ± 10.6	PEA: Female 27 (84.4%)/Male 5 (15.6%) CTRL: Female 21 (63.6%)/Male 12 (36.4%)	LipiSperse PEA (PEA 1.5% concentrated/bid, moisturizing cream, skin application)	4 weeks, monotherapy
Rossi et al., 2020 (Italy) [83]	Single-blind	Primary open angle glaucoma/Normal tension glaucoma	Starting-with-PEA group: 67.6 (62.3–75.5)	Starting-with-PEA group: Female 11	umPEA (600 mg/day, tablet, oral)	16 weeks, add-on to current topical therapy

		(Clinical assessment, IOP)	Switching-to-PEA group: 66.7 (55.7–70.0)	(52.4%)/Male 10 (47.6%) Switching-to-PEA group: Female 8 (42.1%)/Male 11 (57.9%)		
Salaffi et al., 2023 (Italy) [78]	Open-label	Fibromyalgia (ACR)	PEA: 52.9 ± 12.1 CTRL: 53.4 ± 13.12	PEA: Female 62 (100.0%)/Male 0 (0.0%) PLB: Female 68 (100.0%)/Male 0 (0.0%)	PEA/ALC (PEA 600 mg + ALC 500 mg/bid, tablet, oral)	12 weeks, add-on to dlx and pgb
Salehi et al., 2022 (Iran) [52]	Double-blind	Chronic Schizophrenia (SCID-5)	PEA: 33.8 ± 6.9 PLB: 36.8 ± 9.6	PEA: Female 7 (8.0%)/Male 23 (92.0%) PLB: Female 9 (16.0%)/Male 21 (84.0%)	Undeclared PEA formulation (600 mg/bid, capsule, oral)	8 weeks, add-on to rsp
Steels et al., 2018 (Australia) [53]	Double-blind	Knee osteoarthrits (Clinical assessment)	PEA(300 mg): 57.0 ± 11.0 PEA(600 mg): 58.0 ± 11.0 PLB: 55.0 ± 11.0	PEA(300 mg): Female 20 (66.6%)/Male 16 (44.4%) PEA(600 mg): Female 17 (48.6%)/Male 18 (51.4%) PLB: Female 22 (55.0%)/Male 18 (45.0%)	LipiSperse PEA (150 mg/bid or 300 mg/bid, capsule, oral)	8 weeks, monotherapy
Strobbe et al., 2013 (Italy) [84]	Double-blind	Ocular hypertension (Clinical assessment, IOP)	PEA: 56.8 ± 8.1 PLB: 56.2 ± 10.4	PEA: Female 16 (40.0%)/Male 24 (60.0%) PLB: Female 22 (55.0%)/Male 18 (45.0%)	Undeclared PEA formulation (300 mg/bid, tablet, oral)	12 weeks, monotherapy

Tartaglia et al., 2015 (Italy) [85]	X	Primary dysmenorrhea (Clinical assessment)	PEA: 19,88 ± 2.27 PLB: 19,86 ± 2.35	N/A	PEA/transpolydatin (PEA + transpolydatin once/day, tablet, oral; dosage not given)	10 days, monotherapy (from the 24th day of cycle)
Versace et al., 2023 (Italy) [54]	Double-blind	Post-COVID-19 cognitive dyfunction and fatigue (Clinical assessment, PCR nasopharyngeal swab)	PEA: 53.5 ±10.4 PLB: 48.1 ±10.7	PEA: Female 11 (64.7%)/Male 6 (35.3%) PLB: Female 11 (64.7%)/Male 6 (35.3%)	Co-ultraPEALut (umPEA 700 mg + luteolin 70 mg/bid, microgranules, sublingual)	8 weeks, monotherapy
Yuan et al., 2014 (China) [73]	Double-blind	Asteatotic eczema (Clinical assessment)	PEA: 51.90 ± 10.9 CTRL: 53.43 ± 9.8	PEA: Female 30 (100.0%)/Male 0 (0.0%) CTRL: Female 30 (100.0%)/Male 0 (0.0%)	PEA/AEA (PEA 0.3% + AEA 0.21% 2 mg/cm^2/bid, emollient cream, skin application)	4 weeks, monotherapy

AAOS, American Academy of Orthopaedic Surgeons; ACR, American College of Rheumatology; AEA, Anandamide; ALA, Alpha-lipoic acid; ALC, Acetyl-L-Carnitine; BCVA, Best Corrected Visual Acuity; CAM, Confusion Assessment Method; Co-ultraPEALut, Co-ultramicronized PEA plus Luteolin; COD, Chronic olfactory dysfunction; COVID-19, SARS-CoV-2 infection; CPSI, Chronic Prostatitis Symptom Index; ctp, Citalopram; CTRL, Control; DAS, Disease Activity Score; dlx, Duloxetine; DN4, Neuropathic pain diagnostic questionnaire; DOM, Delirium-O-Meter; DSM, Diagnostic and Statistical Manual of Mental Disorders; ENG, Electroneurography; ESHRE, European Society of Human Reproduction and Embryology; GCS, Glasgow Coma Scale; h, hours; GHT, Glaucoma Hemifield Test; ICHD3, International Classification of Headache Disorders, 3rd Edition; IHCC, International Headache Classification Criteria; IOP, Intra-ocular pressure; lth, Lithium; MINI, The Mini International Neuropsychiatric Interview; mPEA, Micronized-PEA; MRI, Magnetic Resonance Imaging; N/A, Not applicable; NIH, National Institutes of Health; NIHSS, National Institutes of Health Stroke Scale; OT, Olfactory training; PCR, Polymerase Chain Reaction; PEA, Palmitoylethanolamide; pgb, Pregabalin; PLB, Placebo; PSA, Prostate-specific antigen; PSQI, Pittsburgh Sleep Quality Index; RDC/TMD, Reasearch Diagnostic Criteria for TMD; rlz, Riluzole; rsp, Risperidone; rTNSS, Reflective Total Nasal Symptom Score; S-LANSS, Self-reported Leeds Assessment of Neuropathic Symptoms and Signs; SCI, Spinal Cord Injury; SCID, Structured Clinical Interview of DSM; SRP, Scaling and root planning; TAU, Treatment as usual; TENS, Transcutaneous electrical nerve stimulation; TMD, Temporo-mandibular Disorder; umPEA, Ultramicronized-PEA.

Table 3. Risk of bias and quality assessment (Johanna Briggs Institute tool for Randomized Controlled Trials)

Author, year/Question	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10	Q11	Q12	Q13	Quality Appraisal
Abedini et al., 2022 [41]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Albanese et al., 2022 [88]	U	U	Y	U	U	Y	U	Y	Y	Y	Y	Y	N	Fair
Andresen et al., 2016 [42]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Bacci et al., 2011 [68]	Y	U	Y	Y	N	Y	U	Y	Y	U	Y	Y	Y	Good
Bonzanino et al., 2024 [55]	Y	N	Y	N	N	Y	N	Y	Y	U	Y	Y	Y	Good
Briskey et al., 2023 [74]	Y	Y	Y	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Good
Briskey et al., 2024 [67]	Y	Y	Y	Y	U	Y	Y	Y	Y	U	Y	Y	Y	Good
Campolo et al., 2021 [43]	Y	U	Y	U	U	Y	Y	Y	Y	Y	Y	Y	Y	Good
Cantone et al., 2024 [65]	Y	Y	Y	U	Y	Y	Y	Y	Y	U	Y	Y	Y	Good
Cobellis et al., 2011 [79]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Coraci et al., 2018 [56]	Y	Y	Y	Y	U	Y	U	Y	Y	U	Y	Y	U	Good
Costagliola et al., 2014 [86]	U	N	U	N	N	Y	N	Y	Y	Y	Y	Y	Y	Fair
Cremon et al., 2016 [80]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
D'Ascanio et al., 2021 [57]	Y	N	N	Y	N	Y	N	Y	Y	Y	Y	Y	Y	Good
Di Nardo et al., 2024 [87]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Di Stadio et al., 2022 [58]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Di Stadio et al., 2023a [59]	U	Y	N	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Di Stadio et al., 2023b [66]	Y	Y	Y	Y	Y	N	Y	Y	Y	Y	Y	Y	Y	Good

Evangelista et al., 2018 [44]	Y	N	Y	N	N	Y	N	Y	Y	Y	Y	Y	Y	Good
Faig-Marti and Martìnez-Catassùs, 2017 [60]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Faig-Martì and Martìnez-Catassùs, 2020 [61]														
Gagliano et al., 2011 [45]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Germini et al., 2017 [69]	Y	N/A	N/A	Y	Y	N/A	Y	Y	Y	N/A	N/A	Y	Y	Good
Ghazizadeh-Hashemi et al., 2018 [46]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Giammusso et al., 2017 [81]	Y	Y	Y	Y	Y	N	U	Y	Y	Y	Y	Y	Y	Good
Guida et al., 2010 [62]	Y	Y	Y	Y	Y	N	U	Y	Y	Y	Y	Y	Y	Good
Isola et al., 2021 [70]	Y	N	Y	N	N	Y	Y	Y	Y	Y	Y	Y	Y	Good
Kadanangode Narayanaswam et al., 2023 [75]	Y	N	Y	N	N	Y	N	Y	Y	N	Y	Y	Y	Good
Khalaj et al., 2018 [47]	Y	U	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Lunardelli et al., 2019 [48]	Y	Y	Y	Y	N	Y	N	Y	Y	Y	Y	Y	Y	Good
Marini et al., 2013 [71]	Y	Y	Y	Y	Y	N	Y	Y	Y	Y	Y	Y	Y	Good
Masek et al., 1974 [72]	Y	Y	U	Y	Y	Y	U	Y	N	Y	Y	Y	Y	Good
Murina et al., 2013 [82]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Orefice et al., 2016 [49]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Ottaviani et al., 2019 [63]	Y	Y	N	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Palma et al., 2016 [64]	Y	Y	Y	Y	U	Y	U	Y	Y	U	Y	Y	Y	Good
Pickering et al., 2022 [50]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Rao et al., 2021 [51]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good

Rao et al., 2023 [76]	Y	Y	Y	Y	Y	Y	Y	Y	Y	N	Y	Y	Y	Good
Rao et al., 2024 [77]	Y	Y	Y	U	U	Y	U	Y	Y	N	Y	Y	Y	Good
Rossi et al., 2020 [83]	Y	Y	Y	N	N	N	Y	Y	Y	Y	Y	Y	Y	Good
Salaffi et al., 2023 [78]	Y	Y	Y	N	N	Y	N	Y	Y	N	Y	Y	Y	Good
Salehi et al., 2022 [52]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Steels et al., 2018 [53]	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Strobbe et al., 2013 [84]	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Good
Tartaglia et al., 2015 [85]	Y	Y	Y	U	U	U	U	Y	Y	Y	Y	Y	Y	Good
Versace et al., 2023 [54]	N	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good
Yuan et al., 2014 [73]	Y	U	Y	Y	Y	Y	U	Y	Y	Y	Y	Y	Y	Good

Qn, Question; Y, Yes; N, No; U, Unclear; N/A, Not applicable.

References

1. Robinson, S.M., *Improving nutrition to support healthy ageing: what are the opportunities for intervention?* Proc Nutr Soc, 2018. **77**(3): p. 257-264.
2. Papadopoulou, S.K., et al., *Nutritional Status Is Associated with Health-Related Quality of Life, Physical Activity, and Sleep Quality: A Cross-Sectional Study in an Elderly Greek Population.* Nutrients, 2023. **15**(2).
3. Chandra, S., et al., *Nutraceuticals: Pharmacologically Active Potent Dietary Supplements.* Biomed Res Int, 2022. **2022**: p. 2051017.
4. Feart, C., *Dietary Supplements: Which Place between Food and Drugs?* Nutrients, 2020. **12**(1).
5. Dwyer, J.T., P.M. Coates, and M.J. Smith, *Dietary Supplements: Regulatory Challenges and Research Resources.* Nutrients, 2018. **10**(1).
6. Skaper, S.D., *Impact of Inflammation on the Blood-Neural Barrier and Blood-Nerve Interface: From Review to Therapeutic Preview.* Int Rev Neurobiol, 2017. **137**: p. 29-45.
7. Skaper, S.D., et al., *An Inflammation-Centric View of Neurological Disease: Beyond the Neuron.* Front Cell Neurosci, 2018. **12**: p. 72.
8. Arturo, I.F. and P. Fabiana, *Endocannabinoidome*, in *eLS*. p. 1-10.
9. Veilleux, A., V. Di Marzo, and C. Silvestri, *The Expanded Endocannabinoid System/Endocannabinoidome as a Potential Target for Treating Diabetes Mellitus.* Curr Diab Rep, 2019. **19**(11): p. 117.
10. Kleberg, K., H.A. Hassing, and H.S. Hansen, *Classical endocannabinoid-like compounds and their regulation by nutrients.* Biofactors, 2014. **40**(4): p. 363-72.
11. Rahman, S.M.K., et al., *Roles of Endocannabinoids and Endocannabinoid-Like Molecules in Energy Homeostasis and Metabolic Regulation: A Nutritional Perspective.* Annu Rev Nutr, 2021. **41**: p. 177-202.
12. Tutunchi, H., et al., *Effects of oleoylethanolamide supplementation on the expression of lipid metabolism-related genes and serum NRG4 levels in patients with non-alcoholic fatty liver disease: A randomized controlled trial.* Clin Nutr ESPEN, 2023. **58**: p. 311-319.
13. Laleh, P., et al., *Oleoylethanolamide increases the expression of PPAR-Alpha and reduces appetite and body weight in obese people: A clinical trial.* Appetite, 2018. **128**: p. 44-49.
14. Shivyari, F.T., et al., *Examining the oleoylethanolamide supplement effects on glycemic status, oxidative stress, inflammation, and anti-mullerian hormone in polycystic ovary syndrome.* J Ovarian Res, 2024. **17**(1): p. 111.

15. Kazemi, M., et al., *Decreased dysmenorrhea pain in girls by reducing oxidative stress and inflammatory biomarkers following supplementation with oleoylethanolamide: A randomized controlled trial.* J Obstet Gynaecol Res, 2022. **48**(5): p. 1212-1221.
16. Mandoe, M.J., et al., *The 2-monoacylglycerol moiety of dietary fat appears to be responsible for the fat-induced release of GLP-1 in humans.* Am J Clin Nutr, 2015. **102**(3): p. 548-55.
17. Petrosino, S. and A. Schiano Moriello, *Palmitoylethanolamide: A Nutritional Approach to Keep Neuroinflammation within Physiological Boundaries-A Systematic Review.* Int J Mol Sci, 2020. **21**(24).
18. Bortoletto, R., et al., *Is It Time to Test the Antiseizure Potential of Palmitoylethanolamide in Human Studies? A Systematic Review of Preclinical Evidence.* Brain Sciences, 2022. **12**(1).
19. Clayton, P., et al., *Palmitoylethanolamide: A Natural Compound for Health Management.* Int J Mol Sci, 2021. **22**(10).
20. Lang-Illievich, K., et al., *Palmitoylethanolamide in the Treatment of Chronic Pain: A Systematic Review and Meta-Analysis of Double-Blind Randomized Controlled Trials.* Nutrients, 2023. **15**(6).
21. Petrosino, S. and V. Di Marzo, *The pharmacology of palmitoylethanolamide and first data on the therapeutic efficacy of some of its new formulations.* Br J Pharmacol, 2017. **174**(11): p. 1349-1365.
22. Lo Verme, J., et al., *The nuclear receptor peroxisome proliferator-activated receptor-alpha mediates the anti-inflammatory actions of palmitoylethanolamide.* Mol Pharmacol, 2005. **67**(1): p. 15-9.
23. Pertwee, R.G., *GPR55: a new member of the cannabinoid receptor clan?* Br J Pharmacol, 2007. **152**(7): p. 984-6.
24. Ambrosino, P., et al., *Activation and desensitization of TRPV1 channels in sensory neurons by the PPARalpha agonist palmitoylethanolamide.* Br J Pharmacol, 2013. **168**(6): p. 1430-44.
25. Petrosino, S., et al., *The anti-inflammatory mediator palmitoylethanolamide enhances the levels of 2-arachidonoyl-glycerol and potentiates its actions at TRPV1 cation channels.* Br J Pharmacol, 2016. **173**(7): p. 1154-62.
26. Di Marzo, V., et al., *Palmitoylethanolamide inhibits the expression of fatty acid amide hydrolase and enhances the anti-proliferative effect of anandamide in human breast cancer cells.* Biochem J, 2001. **358**(Pt 1): p. 249-55.

27. Petrosino, S., et al., *Palmitoylethanolamide counteracts substance P-induced mast cell activation in vitro by stimulating diacylglycerol lipase activity.* J Neuroinflammation, 2019. **16**(1): p. 274.
28. Scuderi, C., et al., *Palmitoylethanolamide counteracts reactive astrogliosis induced by beta-amyloid peptide.* J Cell Mol Med, 2011. **15**(12): p. 2664-74.
29. Clayton, P., et al., *Palmitoylethanolamide: A Potential Alternative to Cannabidiol.* J Diet Suppl, 2021: p. 1-26.
30. Bonaccorso, S., et al., *Cannabidiol (CBD) use in psychiatric disorders: A systematic review.* Neurotoxicology, 2019. **74**: p. 282-298.
31. Lattanzi, S., et al., *Highly Purified Cannabidiol for Epilepsy Treatment: A Systematic Review of Epileptic Conditions Beyond Dravet Syndrome and Lennox-Gastaut Syndrome.* CNS Drugs, 2021. **35**(3): p. 265-281.
32. Villanueva, M.R.B., et al., *Efficacy, Safety, and Regulation of Cannabidiol on Chronic Pain: A Systematic Review.* Cureus, 2022. **14**(7): p. e26913.
33. Story, G., et al., *Cannabidiol and Intestinal Motility: a Systematic Review.* Curr Dev Nutr, 2023. **7**(10): p. 101972.
34. Madeo, G., et al., *Update on Cannabidiol Clinical Toxicity and Adverse Effects: A Systematic Review.* Curr Neuropharmacol, 2023. **21**(11): p. 2323-2342.
35. Gabrielsson, L., S. Mattsson, and C.J. Fowler, *Palmitoylethanolamide for the treatment of pain: pharmacokinetics, safety and efficacy.* Br J Clin Pharmacol, 2016. **82**(4): p. 932-42.
36. Impellizzeri, D., et al., *Micronized/ultramicronized palmitoylethanolamide displays superior oral efficacy compared to nonmicronized palmitoylethanolamide in a rat model of inflammatory pain.* J Neuroinflammation, 2014. **11**: p. 136.
37. Briskey, D., et al., *Increased bioavailability of curcumin using a novel dispersion technology system (LipiSperse(R)).* Eur J Nutr, 2019. **58**(5): p. 2087-2097.
38. Parums, D.V., *Editorial: Review Articles, Systematic Reviews, Meta-Analysis, and the Updated Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 Guidelines.* Med Sci Monit, 2021. **27**: p. e934475.
39. Burns, P.B., R.J. Rohrich, and K.C. Chung, *The levels of evidence and their role in evidence-based medicine.* Plast Reconstr Surg, 2011. **128**(1): p. 305-310.
40. Barker, T.H., et al., *The revised JBI critical appraisal tool for the assessment of risk of bias for randomized controlled trials.* Jbi Evidence Synthesis, 2023. **21**(3): p. 494-506.

41. Abedini, T., et al., *Efficacy and safety of palmitoylethanolamide as an adjunctive treatment for acute mania: A randomized, double-blind, placebo-controlled trial.* Psychiatry Clin Neurosci, 2022. **76**(10): p. 505-511.
42. Andresen, S.R., et al., *Ultramicronized palmitoylethanolamide in spinal cord injury neuropathic pain: a randomized, double-blind, placebo-controlled trial.* Pain, 2016. **157**(9): p. 2097-2103.
43. Campolo, M., et al., *Co-Ultra PEALut Enhances Endogenous Repair Response Following Moderate Traumatic Brain Injury.* Int J Mol Sci, 2021. **22**(16).
44. Evangelista, M., et al., *Ultra-micronized Palmitoylethanolamide Effects on Sleep-wake Rhythm and Neuropathic Pain Phenotypes in Patients with Carpal Tunnel Syndrome: An Open-label, Randomized Controlled Study.* CNS Neurol Disord Drug Targets, 2018. **17**(4): p. 291-298.
45. Gagliano, C., et al., *Ocular hypotensive effect of oral palmitoyl-ethanolamide: a clinical trial.* Invest Ophthalmol Vis Sci, 2011. **52**(9): p. 6096-100.
46. Ghazizadeh-Hashemi, M., et al., *Palmitoylethanolamide as adjunctive therapy in major depressive disorder: A double-blind, randomized and placebo-controlled trial.* J Affect Disord, 2018. **232**: p. 127-133.
47. Khalaj, M., et al., *Palmitoylethanolamide as adjunctive therapy for autism: Efficacy and safety results from a randomized controlled trial.* J Psychiatr Res, 2018. **103**: p. 104-111.
48. Lunardelli, M.L., et al., *Co-ultraPEALut: Role in Preclinical and Clinical Delirium Manifestations.* CNS Neurol Disord Drug Targets, 2019. **18**(7): p. 530-554.
49. Orefice, N.S., et al., *Oral Palmitoylethanolamide Treatment Is Associated with Reduced Cutaneous Adverse Effects of Interferon-beta1a and Circulating Proinflammatory Cytokines in Relapsing-Remitting Multiple Sclerosis.* Neurotherapeutics, 2016. **13**(2): p. 428-38.
50. Pickering, E., et al., *A randomized controlled trial assessing the safety and efficacy of palmitoylethanolamide for treating diabetic-related peripheral neuropathic pain.* Inflammopharmacology, 2022. **30**(6): p. 2063-2077.
51. Rao, A., et al., *Palmitoylethanolamide for sleep disturbance. A double-blind, randomised, placebo-controlled interventional study.* Sleep Sci Pract, 2021. **5**(1): p. 12.
52. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.

53. Steels, E., et al., *A double-blind randomized placebo controlled study assessing safety, tolerability and efficacy of palmitoylethanolamide for symptoms of knee osteoarthritis.* Inflammopharmacology, 2019. **27**(3): p. 475-485.
54. Versace, V., et al., *Co-ultramicronized palmitoylethanolamide/luteolin normalizes GABA(B)-ergic activity and cortical plasticity in long COVID-19 syndrome.* Clin Neurophysiol, 2023. **145**: p. 81-88.
55. Bonzanino, M., et al., *PEALut in the Dietary Management of Patients with Acute Ischemic Stroke: A Prospective Randomized Controlled Clinical Trial.* J Clin Med, 2024. **13**(2).
56. Coraci, D., et al., *Carpal tunnel syndrome treatment with palmitoylethanolamide: neurophysiology and ultrasound show small changes in the median nerve.* Rheumatol Int, 2018. **38**(7): p. 1307-1309.
57. D'Ascanio, L., et al., *Randomized clinical trial "olfactory dysfunction after COVID-19: olfactory rehabilitation therapy vs. intervention treatment with Palmitoylethanolamide and Luteolin": preliminary results.* Eur Rev Med Pharmacol Sci, 2021. **25**(11): p. 4156-4162.
58. Di Stadio, A., et al., *Ultramicronized Palmitoylethanolamide and Luteolin Supplement Combined with Olfactory Training to Treat Post-COVID-19 Olfactory Impairment: A Multi-Center Double-Blinded Randomized Placebo- Controlled Clinical Trial.* Curr Neuropharmacol, 2022. **20**(10): p. 2001-2012.
59. Di Stadio, A., et al., *Parosmia COVID-19 Related Treated by a Combination of Olfactory Training and Ultramicronized PEA-LUT: A Prospective Randomized Controlled Trial.* Biomedicines, 2023. **11**(4).
60. Faig-Marti, J. and A. Martinez-Catassus, *Use of palmitoylethanolamide in carpal tunnel syndrome: a prospective randomized study.* J Orthop Traumatol, 2017. **18**(4): p. 451-455.
61. Faig-Marti, J. and A. Martinez-Catassus, *Measuring the placebo effect in carpal tunnel syndrome.* J Orthop Traumatol, 2020. **21**(1): p. 1.
62. Guida, G.D.M., M.; De Fabiani, A.; Cantieri, L.; Alexandre, A.; Vassallo, G. M.; Rogai, M.; Lanaia, F.; Petrosino, S., *Palmitoylethanolamide (Normast®) in chronic neuropathic pain by compressive type lumbosciatalgia: Multicentric clinical study.* DOLOR, 2010(25): p. 35-42.
63. Ottaviani, G., et al., *Efficacy of ultramicronized palmitoylethanolamide in burning mouth syndrome-affected patients: a preliminary randomized double-blind controlled trial.* Clin Oral Investig, 2019. **23**(6): p. 2743-2750.
64. Palma, E., et al., *Acetylcholine receptors from human muscle as pharmacological targets for ALS therapy.* Proc Natl Acad Sci U S A, 2016. **113**(11): p. 3060-5.

65. Cantone, E., et al., *Persistent COVID-19 parosmia and olfactory loss post olfactory training: randomized clinical trial comparing central and peripheral-acting therapeutics.* Eur Arch Otorhinolaryngol, 2024. **281**(7): p. 3671-3678.
66. Di Stadio, A., et al., *Treatment of COVID-19 olfactory dysfunction with olfactory training, palmitoylethanolamide with luteolin, or combined therapy: a blinded controlled multicenter randomized trial.* Eur Arch Otorhinolaryngol, 2023. **280**(11): p. 4949-4961.
67. Briskey, D., et al., *Effectiveness of Palmitoylethanolamide (Levagen+) Compared to a Placebo for Reducing Pain, Duration, and Medication Use during Migraines in Otherwise Healthy Participants-A Double-Blind Randomised Controlled Study.* Pharmaceuticals (Basel), 2024. **17**(2).
68. Bacci, C., et al., *Randomized split-mouth study on postoperative effects of palmitoylethanolamide for impacted lower third molar surgery.* ISRN Surg, 2011. **2011**: p. 917350.
69. Germini, F., et al., *N-of-1 Randomized Trials of Ultra-Micronized Palmitoylethanolamide in Older Patients with Chronic Pain.* Drugs Aging, 2017. **34**(12): p. 941-952.
70. Isola, G., et al., *Effectiveness of a nutraceutical agent in the non-surgical periodontal therapy: a randomized, controlled clinical trial.* Clin Oral Investig, 2021. **25**(3): p. 1035-1045.
71. Marini, I., et al., *Palmitoylethanolamide versus a nonsteroidal anti-inflammatory drug in the treatment of temporomandibular joint inflammatory pain.* J Orofac Pain, 2012. **26**(2): p. 99-104.
72. Masek, K., et al., *Prophylactic efficacy of N-2-hydroxyethyl palmitamide (impulsin) in acute respiratory tract infections.* Eur J Clin Pharmacol, 1974. **7**(6): p. 415-9.
73. Yuan, C., et al., *N-palmitoylethanolamine and N-acetylethanolamine are effective in asteatotic eczema: results of a randomized, double-blind, controlled study in 60 patients.* Clin Interv Aging, 2014. **9**: p. 1163-9.
74. Briskey, D., P. Ebelt, and A. Rao, *The Effect of Levagen+ (Palmitoylethanolamide) Supplementation on Symptoms of Allergic Rhinitis-A Double-Blind Placebo-Controlled Trial.* Nutrients, 2023. **15**(23).
75. Kadanangode Narayanaswam, N., et al., *A randomized interventional clinical trial assessing the safety and effectiveness of PeaNoc XL tablets in managing joint pain and inflammation in arthritis patients.* F1000Res, 2023. **12**: p. 895.

76. Rao, A., R. Skinner, and D. Briskey, *The Efficacy of Palmitoylethanolamide (Levagen+) on the Incidence and Symptoms of Upper Respiratory Tract Infection-A Double Blind, Randomised, Placebo-Controlled Trial.* Nutrients, 2023. **15**(20).
77. Rao, A., et al., *Efficacy of Topical Palmitoylethanolamide (Levagen+) for the Management of Eczema Symptoms: A Double-Blind, Comparator-Controlled, Randomized Clinical Trial.* Skin Pharmacol Physiol, 2023. **36**(6): p. 288-295.
78. Salaffi, F., et al., *Palmitoylethanolamide and acetyl-L-carnitine act synergistically with duloxetine and pregabalin in fibromyalgia: results of a randomised controlled study.* Clin Exp Rheumatol, 2023. **41**(6): p. 1323-1331.
79. Cobellis, L., et al., *Effectiveness of the association micronized N-Palmitoylethanolamine (PEA)-transpolydatin in the treatment of chronic pelvic pain related to endometriosis after laparoscopic assessment: a pilot study.* Eur J Obstet Gynecol Reprod Biol, 2011. **158**(1): p. 82-6.
80. Cremon, C., et al., *Randomised clinical trial: the analgesic properties of dietary supplementation with palmitoylethanolamide and polydatin in irritable bowel syndrome.* Aliment Pharmacol Ther, 2017. **45**(7): p. 909-922.
81. Giammusso, B., R. Di Mauro, and R. Bernardini, *The efficacy of an association of palmitoylethanolamide and alpha-lipoic acid in patients with chronic prostatitis/chronic pelvic pain syndrome: A randomized clinical trial.* Arch Ital Urol Androl, 2017. **89**(1): p. 17-21.
82. Murina, F., et al., *Vestibulodynia: synergy between palmitoylethanolamide + transpolydatin and transcutaneous electrical nerve stimulation.* J Low Genit Tract Dis, 2013. **17**(2): p. 111-6.
83. Rossi, G.C.M., et al., *Effect of palmitoylethanolamide on inner retinal function in glaucoma: a randomized, single blind, crossover, clinical trial by pattern-electroretinogram.* Sci Rep, 2020. **10**(1): p. 10468.
84. Strobbe, E., M. Cellini, and E.C. Campos, *Effectiveness of palmitoylethanolamide on endothelial dysfunction in ocular hypertensive patients: a randomized, placebo-controlled cross-over study.* Invest Ophthalmol Vis Sci, 2013. **54**(2): p. 968-73.
85. Tartaglia, E., et al., *Effectiveness of the Association N-Palmitoylethanolamine and Transpolydatin in the Treatment of Primary Dysmenorrhea.* J Pediatr Adolesc Gynecol, 2015. **28**(6): p. 447-50.

86. Costagliola, C., et al., *Effect of palmitoylethanolamide on visual field damage progression in normal tension glaucoma patients: results of an open-label six-month follow-up.* J Med Food, 2014. **17**(9): p. 949-54.
87. Di Nardo, G., et al., *Palmitoylethanolamide and polydatin in pediatric irritable bowel syndrome: A multicentric randomized controlled trial.* Nutrition, 2024. **122**: p. 112397.
88. Albanese, M., et al., *Effects of Ultramicronized Palmitoylethanolamide (um-PEA) in COVID-19 Early Stages: A Case-Control Study.* Pharmaceuticals (Basel), 2022. **15**(2).
89. Capra, A.P., et al., *Efficacy of Palmitoylethanolamide and Luteolin Association on Post-Covid Olfactory Dysfunction: A Systematic Review and Meta-Analysis of Clinical Studies.* Biomedicines, 2023. **11**(8).
90. Santonocito, S., et al., *Orofacial Pain Management: An Overview of the Potential Benefits of Palmitoylethanolamide and Other Natural Agents.* Pharmaceutics, 2023. **15**(4).
91. Scuteri, D., et al., *Effects of Palmitoylethanolamide (PEA) on Nociceptive, Musculoskeletal and Neuropathic Pain: Systematic Review and Meta-Analysis of Clinical Evidence.* Pharmaceutics, 2022. **14**(8).
92. Indraccolo, U., et al., *Looking for Responders among Women with Chronic Pelvic Pain Treated with a Comicronized Formulation of Micronized Palmitoylethanolamide and Polydatin.* Biomed Res Int, 2022. **2022**: p. 8620077.
93. Artukoglu, B.B., et al., *Efficacy of Palmitoylethanolamide for Pain: A Meta-Analysis.* Pain Physician, 2017. **20**(5): p. 353-362.
94. Schweiger, V., et al., *Extended Treatment with Micron-Size Oral Palmitoylethanolamide (PEA) in Chronic Pain: A Systematic Review and Meta-Analysis.* Nutrients, 2024. **16**(11).
95. Brotini, S., C. Schievano, and L. Guidi, *Ultra-micronized Palmitoylethanolamide: An Efficacious Adjuvant Therapy for Parkinson's Disease.* CNS Neurol Disord Drug Targets, 2017. **16**(6): p. 705-713.
96. Mallard, A., et al., *The Effect of Orally Dosed Levagen+ (palmitoylethanolamide) on Exercise Recovery in Healthy Males-A Double-Blind, Randomized, Placebo-Controlled Study.* Nutrients, 2020. **12**(3).
97. Varrassi, G., et al., *A Decades-Long Journey of Palmitoylethanolamide (PEA) for Chronic Neuropathic Pain Management: A Comprehensive Narrative Review.* Pain Ther, 2024.
98. Colizzi, M., et al., *Therapeutic effect of palmitoylethanolamide in cognitive decline: A systematic review and preliminary meta-analysis of preclinical and clinical evidence.* Frontiers in Psychiatry, 2022. **13**.

99. Kim, N., et al., *Formulated Palmitoylethanolamide Supplementation Improves Parameters of Cognitive Function and BDNF Levels in Young, Healthy Adults: A Randomised Cross-Over Trial.* Nutrients, 2024. **16**(4).
100. Colizzi, M., et al., *Palmitoylethanolamide and Its Biobehavioral Correlates in Autism Spectrum Disorder: A Systematic Review of Human and Animal Evidence.* Nutrients, 2021. **13**(4).
101. Bortoletto, R., et al., *Questioning the role of palmitoylethanolamide in psychosis: a systematic review of clinical and preclinical evidence.* Front Psychiatry, 2023. **14**: p. 1231710.
102. Dunleavy, C., et al., *Inflammation in first-episode psychosis: The contribution of inflammatory biomarkers to the emergence of negative symptoms, a systematic review and meta-analysis.* Acta Psychiatr Scand, 2022. **146**(1): p. 6-20.
103. Masi, A., et al., *Cytokine aberrations in autism spectrum disorder: a systematic review and meta-analysis.* Mol Psychiatry, 2015. **20**(4): p. 440-6.
104. Arteaga-Henriquez, G., et al., *Activation of the Monocyte/Macrophage System and Abnormal Blood Levels of Lymphocyte Subpopulations in Individuals with Autism Spectrum Disorder: A Systematic Review and Meta-Analysis.* Int J Mol Sci, 2022. **23**(22).
105. Tseng, P.T., et al., *Assessment of Noninvasive Brain Stimulation Interventions for Negative Symptoms of Schizophrenia: A Systematic Review and Network Meta-analysis.* JAMA Psychiatry, 2022. **79**(8): p. 770-779.
106. Krause, M., et al., *Antipsychotic drugs for patients with schizophrenia and predominant or prominent negative symptoms: a systematic review and meta-analysis.* Eur Arch Psychiatry Clin Neurosci, 2018. **268**(7): p. 625-639.
107. Williams, K., et al., *Selective serotonin reuptake inhibitors (SSRIs) for autism spectrum disorders (ASD).* Cochrane Database Syst Rev, 2013(8): p. CD004677.
108. Hurwitz, R., et al., *Tricyclic antidepressants for autism spectrum disorders (ASD) in children and adolescents.* Cochrane Database Syst Rev, 2012(3): p. CD008372.
109. Crupi, L., et al., *Evaluation of the nutraceutical Palmitoylethanolamide in reducing intraocular pressure (IOP) in patients with glaucoma or ocular hypertension: a systematic review and meta-analysis.* Nat Prod Res, 2024: p. 1-20.
110. Batacan, R., Jr., et al., *Effect of Palmitoylethanolamide Compared to a Placebo on the Gut Microbiome and Biochemistry in an Overweight Adult Population: A Randomised, Placebo Controlled, Double-Blind Study.* Biomedicines, 2024. **12**(7).

111. Brankovic, M., et al., *Therapeutic Potential of Palmitoylethanolamide in Gastrointestinal Disorders.* Antioxidants (Basel), 2024. **13**(5).
112. Pirozzi, C., et al., *Palmitoylethanolamide counteracts high-fat diet-induced gut dysfunction by reprogramming microbiota composition and affecting tryptophan metabolism.* Front Nutr, 2023. **10**: p. 1143004.
113. Minichino, A., et al., *Endocannabinoid system mediates the association between gut-microbial diversity and anhedonia/amotivation in a general population cohort.* Mol Psychiatry, 2021. **26**(11): p. 6269-6276.

Acknowledgement: The authors would like to aknowledge infrastructures from the Department of Medicine (DMED) – University of Udine (Italy).

Funding: This work received no external funding.

CRediT authorship contribution statement:

RB: Conceptualization, Data curation, Investigation, Methodology, Software, Writing-original draft, Writing – review and editing

CC: Conceptualization, Data curation, Investigation, Methodology, Writing – review and editing

MG: Conceptualization, Data curation, Methodology, Writing – review and editing

FP: Conceptualization, Data curation, Methodology, Writing – review and editing

MB: Conceptualization, Data curation, Methodology, Writing – review and editing

MC: Conceptualization, Data curation, Methodology, Supervision, Writing-original draft, Writing – review and editing

CHAPTER III

Questioning the Role of Palmitoylethanolamide in Psychosis: A Systematic Review of Clinical and Preclinical Evidence

Bortoletto R, Piscitelli F, Candolo A, Bhattacharyya S, Balestrieri M, Colizzi M. Questioning the role of palmitoylethanolamide in psychosis: a systematic review of clinical and preclinical evidence. Front Psychiatry. 2023 Jul 18;14:1231710. doi: 10.3389/fpsyt.2023.1231710. PMID: 37533892; PMCID: PMC10390736.

Abstract

Introduction: The endocannabinoid (eCB) system disruption has been suggested to underpin the development of psychosis, fueling the search for novel, better-tolerated antipsychotic agents that target the eCB system. Among these, palmitoylethanolamide (PEA), an N-acylethanolamine (AE) with neuroprotective, anti-inflammatory, and analgesic properties, has drawn attention for its antipsychotic potential. Methods: This Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020-compliant systematic review aimed at reappraising all clinical and preclinical studies investigating the biobehavioral role of PEA in psychosis. Results: Overall, 13 studies were eligible for data extraction (11 human, 2 animal). Observational studies investigating PEA tone in psychosis patients converged on the evidence for increased PEA plasma (6 human) and central nervous system (CNS; 1 human) levels, as a potential early compensatory response to illness and its severity, that seems to be lost in the longer-term (CNS; 1 human), opening to the possibility of exogenously supplementing it to sustain control of the disorder. Consistently, PEA oral supplementation reduced negative psychotic and manic symptoms among psychosis patients, with no serious adverse events (3 human). No PEA changes emerged in either preclinical psychosis model (2 animal) studied. Discussion: Evidence supports PEA signaling as a potential psychosis biomarker, also indicating a therapeutic role of its supplementation in the disorder. Systematic review registration: https://doi.org/10.17605/OSF.IO/AFMTK.

Keywords

Schizophrenia; antipsychotics; bipolar disorder; cannabidiol; major depressive disorder; nutraceuticals

1. Introduction

Psychotic disorders – non-affective (e.g., schizophrenia (SCZ), schizophreniform disorder) and affective psychoses (e.g., bipolar disorder (BPAD), major depressive disorder with psychotic symptoms) – are a heterogeneous group of disabling mental health disturbances [1], sharing common phenomenological, neurobiological, and genetic characteristics [2-5]. These conditions generally emerge between late adolescence and early adulthood [6], with a lifetime prevalence exceeding 3% [7-9], and severely affect the patients' and their families' quality of life. The dopaminergic and glutamatergic hypotheses still play a pivotal role in the attempt to describe the neurobiological mechanisms underlying psychosis [10-13], with potential for an integrated model explaining both positive (e.g., delusions, hallucinations) and negative (e.g., restricted emotional expression, avolition) psychotic symptoms [12].

To date, antipsychotic (AP) medications represent the cornerstone treatment for these conditions, although not always devoid of suboptimal clinical response and unpleasant side-effects [14, 15]. Therefore, the exploration of other perturbed systems potentially underpinning psychotic disorders has aimed at identifying novel therapeutic targets. The endocannabinoid (eCB) system has been recognised as a mediator of the dopaminergic and glutamatergic systems via the cannabinoid receptor 1 (CB1) in the central nervous system (CNS), and found to be altered in the early phases of the disorder [16-19]. Consistently, accumulating evidence has highlighted the therapeutic potential of the eCB system modulation. Particularly, cannabidiol (CBD) has shown promising results for both psychosis and clinical high-risk (CHR) for psychosis state [20-23]. Further, reduced diversity of gut microbiota and gut-brain axis metabolic alterations associated have been indicated as having a putative role in the patho-etiological cascade towards psychosis [24]. To this end, reduced microbiota diversity has been observed to contribute to common SCZ negative symptoms such as anhedonia and amotivation via eCB-like compound palmitoylethanolamide (PEA) fecal levels, warranting the possibility to target the gut microbiota-eCB axis [25]. Finally, growing evidence emphasizes the importance of inflammation and oxidative stress in the stages preceding psychosis onset and throughout illness progression [26, 27].

PEA is an N-acylethanolamine (AE), produced "on demand" by different cell types as a response to actual or potential damage [28, 29]. Importantly, PEA has been proven to down-regulate central and peripheral activity of mast cells and non-neuronal cells (e.g., astrocytes, microglia) [30-32] and to exert protective functions against glutamate neuro-toxicity, accounting for its naturally-occurring anti-inflammatory, analgesic, and anticonvulsant properties [33]. It directly activates the Peroxisome Proliferator Activated Receptor-α (PPAR-α) and the GPR55, allosterically modulates the Transient

Receptor Potential Vanilloid 1 (TRPV1), and indirectly interacts with CB1 and cannabinoid receptor 2 (CB2) [32, 34, 35]. Due to the shared pharmacodynamic properties, PEA is considered as the endogenous equivalent of CBD [36, 37]. A growing body of literature has confirmed the role of PEA in most neurobiological mechanisms underpinning several neuropsychiatric conditions both in clinical and preclinical settings [38-40].

1.1 Objectives

The effect of PEA over neuroinflammation and glutamate signaling may represent a promising biobehavioral mechanism underlying its clinical utility in psychosis. This systematic review aimed to collect and discuss all available clinical and preclinical data generated by studies investigating the role of PEA in non-affective and affective psychoses. We reviewed all interventional and observational studies, employing either retrospective or prospective methodological approaches with any PEA neuro-biological correlates investigated in psychosis.

2. Materials and Methods

2.1. Inclusion and exclusion criteria

All clinical and preclinical evidence about the topic was gathered and systematically reviewed according to Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines [41]. Inclusion criteria were defined as follows: 1. analytic, observational, and interventional studies; 2. studies assessing (i) acute or long-term effects of palmitoylethanolamide (PEA) administration over psychosis-related biological underpinnings (e.g., neuroimmune disruption, hypothalamus-pituitary-adrenal axis dysregulation); and behavioral features (e.g., negative psychotic symptoms, manic symptoms); or (ii) PEA and PEA signaling-related molecular marker (e.g., other endocannabinoids/acylethanolamines, PEA-related enzymes) modulation in peripheral tissues (e.g., plasma, serum), or in the central nervous system (e.g. cerebrospinal fluid, brain tissue) in psychosis and related conditions. Exclusion criteria were defined as outlined: 1. studies in which (i) PEA was not the intervention or outcome of interest (e.g., studies evaluating only exogenous cannabinoid administration or assessing endogenous cannabinoid levels); and (ii) PEA bio-behavioral correlates were not investigated with reference to psychosis; and (iii) PEA bio-behavioral correlates were not directly reported on; 2. reviews; 3. systematic reviews; 4. meta-analyses.

2.2. Search strategy and data extraction

A literature search was performed using electronic databases (PubMed, Scopus, and Web of Science) for any published original study written in English, on 16 January 2023. In order to be as much inclusive as possible, a combination of broad-meaning terms describing and/or concerning PEA ('palmitoylethanolamide', 'palmitylethanolamide', 'N-2-hydroxyethyl-hexadecanamide', 'N-2-hydroxyethyl-palmitate', 'N-palmitoylethanolamine', 'PEA', and 'palmitoyl-ethanolamine') and psychosis ('schizophreni*', 'psychosis', 'psychoses', 'psychotic', 'bipolar', 'mania', 'manic') was adopted. Reference lists of all selected studies were scrutinized to identify any adjunctive eligible evidence. Data screening and extraction were conducted according to a two-step selection process (conventional double-screening), performed by two researchers (R.B. and M.C.) independently from each other. In the instances of conflicting opinions regarding papers' inclusion, a consensus was sought through discussion with a third senior reviewer (M.B.).

2.3. Risk of bias assessment

In light of the methodological heterogeneity of collected evidence, quality of studies assessment was conducted in accordance to an adapted and suitably flexible set of criteria suggested by the Agency for Healthcare Research and Quality (AHRQ) guidance [42], in line with previous research in the field [38-40]. Risk of systematic bias across human studies was ruled out by screening all papers for potential confounding factors (e.g., gender, age, smoking status, level of education). Furthermore, factors possibly accounting for similarities and differences between all studies were assessed, extracting information about study characteristics, including study design (e.g., observational, interventional), defined study population (for human studies: e.g., schizophrenia (SCZ) patients, clinical high-risk (CHR) subjects; for animal studies: e.g., mouse or rat model), age or developmental stage, gender, adequate psychosis model (for animal studies only: e.g., maternal deprivation, methylazoxymethanol acetate (MAM) prenatal exposure), PEA measure (e.g., PEA dosage and administration route, PEA assessment in tissues), adequate PEA evaluation (e.g., time of exposure, single or multiple assessments), defined control group, comparability of subjects (for human studies only), exclusion criteria/adjusting factors (for human studies only), statistical analyses, and declaration of fundings/sponsorship. The full study protocol is available at https://doi.org/10.17605/OSF.IO/AFMTK.

3. Results

3.1. Study selection

Overall, 418 studies were retrieved through the initial data search. After removing duplicates as well as excluding articles owing to article type (e.g., non-systematic reviews, systematic reviews, meta-analyses), titles, abstracts, or full texts of all records were examined against the inclusion and exclusion criteria following a three-step screening process (Figure 1). A final list of thirteen studies was used for systematic analysis in this review, including 11 studies conducted only in human populations and two studies performed in animal models, exploring various aspects of palmitoylethanolamide (PEA) signaling pathway (Table 1). These include (i) *in vivo* PEA add-on treatment exposure in humans with different types of psychoses (e.g., non-affective psychosis, affective psychosis) or psychotic symptoms (e.g., hallucinations) (3 studies; Table 1); (ii) PEA quantitative blood assessment in humans with psychosis clinical high-risk (CHR) state (1 study; Table 1); (iii) PEA quantitative blood assessment in humans with different types of psychoses (e.g., non-affective psychosis, affective psychosis) at different stages of illness (5 studies; Table 1); (iv) PEA quantitative central nervous system (CNS; e.g., brain tissue, cerebrospinal fluid) assessment in humans with schizophrenia (SCZ; 2 studies; Table 1); (v) PEA quantitative brain tissue assessment in animal models of SCZ (2 studies; Table 1). Additional data on methodological quality of studies conducted in humans and animals are reported in Table 2 (a, b). A brief synthesis of the main results is presented below and summarized in Table 1.

Figure 1. PRISMA flowchart of search strategy for systematic review.

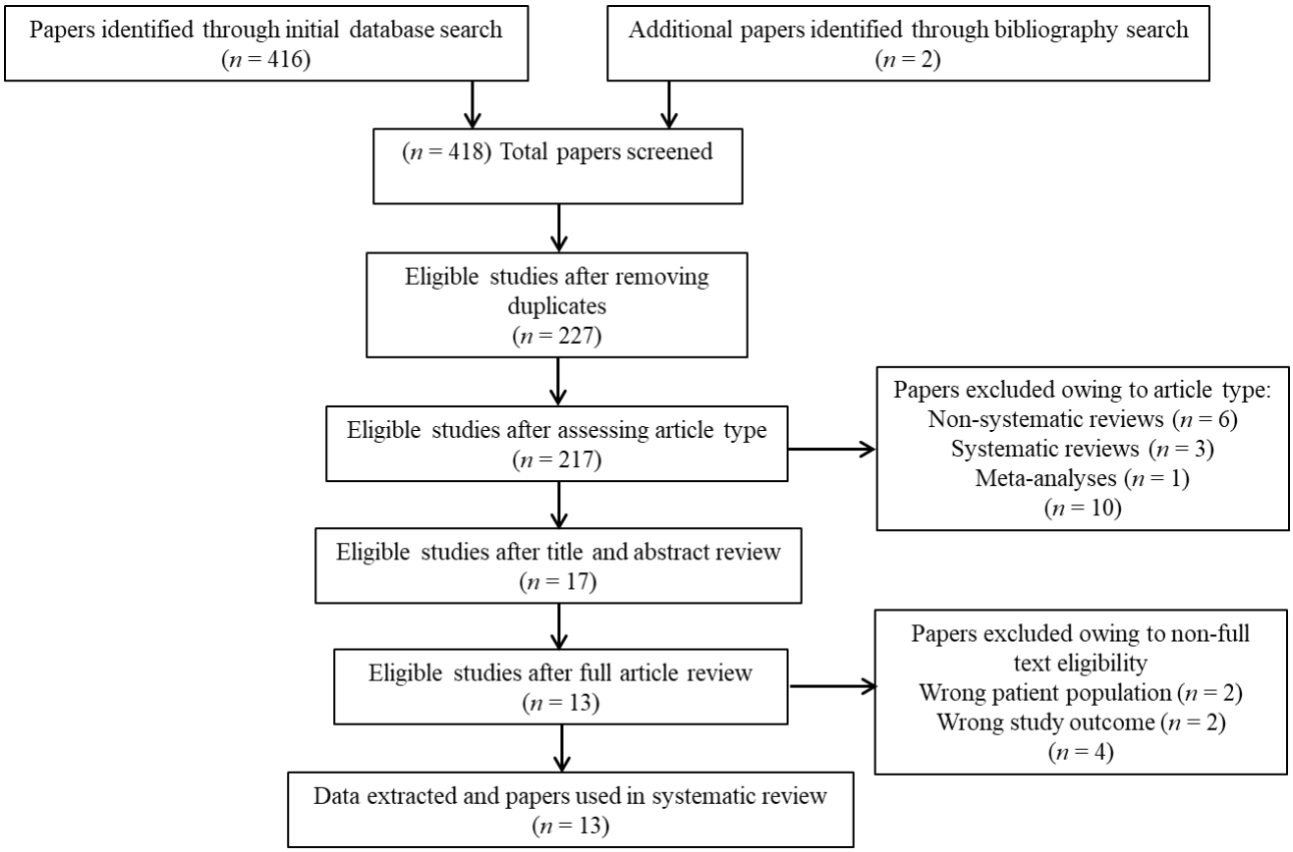


Table 1. Summary of clinical and preclinical studies investigating palmitoylethanolamide and its correlations to psychotic disorders.

Study (year)	Country	Aim of Study	Type of PEA Study	Population	Total Sample Size	Outcome Measure (test name or description)	Summary Results
Leweke et al. (1999)	Germany	To assess PEA and other eCBs/AEs levels in SCZ patients	Quantitative assessment in humans	1. SCZ (*n*=10); 2. HC (*n*=11)	21	eCBs/AEs CSF levels (HPLC, GC/MS)	1. **PEA levels: SCZ > HC;** 2. **AEA levels: SCZ > HC**; 3. OEA levels: SCZ vs. HC, NS
Leweke et al. (2012)	Germany	To assess PEA, other eCBs/AEs, and related enzymes levels in CBD-treated SCZ patients	Quantitative assessment in humans	1. CBD (*n*=20); 2. AMI (*n*=19)	39	eCBs/AEs and related enzymes serum levels (LC/MS, FAAH assay)	CBD group > AMI group: 1. **PEA levels: Day 14, Day 28 > Baseline;** 2. **AEA levels: Day 14, Day 28 > Baseline**; 3. **OEA levels: Day 14 > Baseline;** Day 28 vs. Baseline, NS
Muguruza et al. (2013)	Spain	To assess postmortem PEA and other eCBs/AEs levels in SCZ patients	Quantitative assessment in humans	1. AP-F (*n*=11); 2. AP-T (*n*=8); 3. CTRL (*n*=19)	38	eCBs/AEs brain tissue levels (LC/MS)	1. Effect on eCBs/AEs levels: (a) **2-AG levels: SCZ, ↑**; **brain region**; SCZ x brain region interaction, NS; (b) **AEA levels: SCZ, ↓**; brain region, NS; SCZ x brain region interaction, NS; (c) **DHEA levels: SCZ, ↓**; **brain region**; **SCZ x brain region interaction**; (d) **PEA**, (e) **LEA levels: SCZ, ↓**; **brain region**; SCZ x brain region

	interaction, NS; (f) **OEA levels**: SCZ, NS; **brain region**; SCZ x brain region interaction, NS; 2. **PEA brain tissue levels**: (a) **CB: SCZ < CTRL**; **AP-F < CTRL**; AP-T vs. CTRL, NS; AP-T vs. AP-F, NS; (b) HIP, (c) PFC: all comparisons, NS; 3. **LEA brain tissue levels**: (a) **CB: SCZ < CTRL**; other comparisons, NS; (b) HIP: all comparisons, NS; (c) **PFC: AP-T < CTRL**; other comparisons, NS; 4. **DHEA brain tissue levels:** (a) **CB: SCZ < CTRL**; **AP-F, AP-T < CTRL**; AP-F vs. AP-T, NS; (b) **HIP: SCZ < CTRL**; **AP-F < CTRL**; other comparisons, NS; (c) PFC: all comparisons, NS**;** 5. **AEA brain tissue levels**: (a) **CB: SCZ < CTRL**; **AP-F < CTRL**; other comparisons, NS; (b) **HIP: SCZ < CTRL**; **AP-F, AP-T < CTR**L; AP-

							F vs. AP-T, NS; (c) **PFC: AP-T < CTRL**; other comparisons, NS; 6. **2-AG brain tissue levels**: (a) CB: all comparisons, NS; (b) **HIP: SCZ > CTRL**; **AP-F > CTRL**; other comparisons, NS; (c) **PFC: SCZ > CTRL**; **AP-F > CTRL**; other comparisons, NS; 7. OEA brain tissue levels: all comparisons, NS; 8. **2-AG/PEA ratio**: (a) **CB**, (b) **PFC: SCZ > CTRL**; (c) HIP: SCZ vs. CTRL, NS; 9. **2-AG/other AEs ratio: SCZ > CTRL (all brain regions; all comparisons)**
Koethe et al. (2018)	United States	To assess PEA and other eCBs/AEs levels in SCZ and BPAD discordant twin patients	Quantitative assessment in humans	1. SCZ discordant twin pairs: (a) SCZ (*n*=25); (b) noSCZ (*n*=25); 2. BPAD discordant twin pairs: (a) BPAD (*n*=7); (b)	80	eCBs/AEs plasma levels (LC/MS)	1. **PEA plasma levels: SCZ, noSCZ > HC**; SCZ vs. noSCZ, NS; **BPAD, noBPAD > HC** (NS after Bonferroni's correction); BPAD vs. noBPAD, NS; SCZ-transit vs. SCZ-non transit, NS; **SCZ discordant > BPAD discordant**; 2. **AEA plasma levels:**

				noBPAD (n=7); 3. HC twins (*n*=16)			**SCZ, noSCZ > HC**; SCZ vs. noSCZ, NS; **BPAD, noBPAD > HC**; BPAD vs. noBPAD, NS; **SCZ-transit < SCZ-non transit**; 3. 2-AG plasma levels: SCZ vs. noSCZ vs. HC, NS; BPAD vs. noBPAD vs. HC, NS; **SCZ-transit < SCZ-non transit**; 4. OEA plasma levels: SCZ vs. noSCZ vs. HC, NS; BPAD vs. noBPAD vs. HC, NS; SCZ-transit vs. SCZ-non transit, NS
Appiah-Kusi et al. (2020)	United Kingdom	To assess PEA and other eCBs/AEs levels in CT-exposed CHR patients	Quantitative assessment in humans	1. HC (*n*=58); 2. CHR (*n*=33)	91	eCBs/AEs plasma levels (LC/MS)	1. Group differences on AEs/eCBs **plasma levels**: (a) PEA: CHR > HC (trend effect); (b) **OEA**, (c) **AEA**, (d) **2-AG: CHR > HC**; 2. (a) **CT effect: ↑ PEA, AEA, 2-AG levels**; (b) **CHR effect: ↑ AEA, 2-AG levels**; (c) **CHR x CT interaction: ↑ PEA levels**; ↑ AEA levels (trend effect); 3. **Effects of 2 vs. 1 RF on AEs/eCBs plasma levels**: (a) **PEA**,

							(b) **AEA**, (c) **OEA**, (d) **2-AG: 2RF > 1RF**; 4. **Effects of RF number on AEs/eCBs plasma levels**: (a) **PEA**, (b) **AEA**, (c) **OEA**, (d) **2-AG: noRF < 1RF < 2RF**; 5. ↑ **PEA levels: ↑ total CAARMS score**; ↑ **total CTQ score**; 6. ↑ AEA levels: ↑ total CAARMS score (trend effect)
Ibarra-Lecue et al. (2022)	Spain	To assess PEA and other eCBs/AEs levels in DUAL patients	Quantitative assessment in humans	1. CUD (*n*=26); 2. HC1 (n=24); 3. SCZ (*n*=22); 4. HC2 (n=19); 5. DUAL (*n*=13); 6. HC3 (*n*=10)	114	1. eCBs/AEs plasma levels (HPLC/MS); 2. CB1R protein expression in PLTs (Western blot); 3. Inflammatory response measurements (ELISA)	1. CB1R protein expression: (a) **CUD main effect**; (b) **SCZ main effect**; (c) **CUD x SCZ interaction**; (d) **% from control: CUD, SCZ, DUAL < HC**; 2. eCBs/AEs plasma levels: (a) **SCZ main effect: PEA, OEA**; (b) **CUD main effect: PEA, AEA, DEA, LEA, NADA**; (c) **SCZ x CUD interaction: PEA, AEA, DEA, OEA**; (d) **PEA plasma levels (ng/ml): SCZ > DUAL; SCZ > HC > CUD**; other comparisons,

							NS; (e) **AEA plasma levels (ng/ml): SCZ > DUAL, CUD, HC**; other comparisons, NS; (f) **DEA plasma levels (ng/ml): SCZ > DUAL, CUD**; other comparisons, NS; (g) **OEA plasma levels (ng/ml): SCZ, CUD, HC > DUAL**; other comparisons, NS; (h) **NADA plasma levels (ng/ml): DUAL > HC**; other comparisons, NS; (i) 2-AG, (l) 1-AG, (m) LEA plasma levels (ng/ml): all comparisons, NS; 3. **IL-6 plasma levels (pg/ml): SCZ > DUAL, CUD, HC**; other comparisons, NS
Parksepp et al. (2022)	Estonia	To assess PEA and other eCBs/AEs levels in FEP patients	Quantitative assessment in humans	1. FEP (*n*=54); 2. HC (*n*=58)	112	eCBs/AEs serum levels (HPLC/MS, flow injection analysis tandem MS)	1. eCBs/AEs serum levels: (a) PEA, (b) AEA, (c) LEA, (d) OEA: FEPb > HC (trend effect); FEP(0.6-year) vs. HC, NS; FEP(5.1-year) vs. HC, NS; (e) **2-AG**: FEPb < HC (trend effect); FEP(0.6-year) vs.

							HC, NS; **FEP(5.1-year) > HC**; 2. (eCBs/AEs)/2-AG ratio levels: (a) **PEA/2-AG: FEPb > HC**; FEP(0.6-year) vs. HC, NS; FEP(5.1-year) < HC (trend effect); (b) **AEA/2-AG**, (c) **OEA/2-AG: FEPb > HC**; FEP(0.6-year) vs. HC, NS; FEP(5.1-year) vs. HC, NS; (d) **LEA/2-AG: FEPb > HC**; FEP(0.6-year) vs. HC, NS; **FEP(5.1-year) < HC**; 3. (eCBs/AEs)/AEA ratio levels: (a) PEA/AEA, (b) OEA/AEA: FEPb vs. HC, NS; FEP(0.6-year) vs. HC, NS; FEP(5.1-year) vs. HC, NS; (c) **LEA/AEA**: FEPb vs. HC, NS; FEP(0.6-year) vs. HC, NS; **FEP(5.1-year) < HC**
Topuz et al. (2022)	Turkey	To assess PEA and other eCBs/AEs levels in BPAD patients	Quantitative assessment in humans	1. Past depressive episode: (a) NoPastDEP (*n*=37);	79	eCBs/AEs serum levels (LC/MS)	1. eCBs/AEs serum levels: (a) AEA: NoPastDEP vs. PastDEP, NS; Manic/hypomanic vs. Depressive, NS; (b) **PEA:**

	(b) PastDEP (*n*=42); 2. First episode type: (a) Manic/hypomanic (*n*=52); (b) Depressive (*n*=27)	**NoPastDEP < PastDEP**; **Manic/hypomanic < Depressive**; (c) **OEA: NoPastDEP < PastDEP**; Manic/hypomanic vs. Depressive, NS; (d) AEA: NoPastDEP vs. PastDEP, NS; Manic/hypomanic vs. Depressive, NS; 2. Correlations between illness course and eCBs/AEs serum levels: (a) **PEA: ↑ number of depressive episodes, ↑; ↑ number of hypomanic episodes, ↓; ↑ number of hospitalizations, ↓; ↑ duration of VPA usage, ↓**; other correlations, NS; (b) **AEA: ↑ duration of VPA usage, ↑**; other correlations, NS; (c) **OEA: ↑ age of onset, ↑; ↑ number of depressive episodes, ↑**; other correlations, NS; (d) **2-AG: ↑ number of depressive episodes, ↑**; other correlations, NS; 3. Relation of symptoms and eCBs/AEs serum levels: (a) **PEA:**

	presence of depressive mood, ↑; **presence of increased sexual desire,** ↓; **presence of anxiety,** ↑; **presence of flight of ideas,** ↓; **presence of delusion,** ↓; **presence of grandiosity,** ↓; presence of other symptoms, NS; (b) **AEA: presence of flight of ideas,** ↓; **presence of increased motor activity,** ↓; **presence of Schneiderian symptoms,** ↑; presence of other symptoms, NS; (c) **OEA: presence of depressive mood,** ↑; **presence of anxiety,** ↑; presence of other symptoms, NS; (d) **2-AG: presence of euphoria,** ↑; presence of other symptoms, NS; 4. Relation of medical history and eCBs/AEs serum levels: (a) **PEA: presence of another disease,** ↓; any other medical history, NS; (b) AEA: all medical history, NS; (c) OEA: all medical history, NS; (d) **2-AG:**

							presence of diabetes mellitus, ↓; **presence of another disease, ↓**; any other medical history, NS
Brotini et al. (2017)	Italy	To assess PEA add-on effects on psychotic symptoms in PD patients	In vivo treatment in humans	PD patients	30	nM-EDL assessment (MDS-UPDRS)	Effect on hallucinations and psychosis (nM-EDL scores): post-PEA vs. pre-PEA, NS
Salehi et al. (2022)	Iran	To assess PEA add-on effects on negative symptoms in SCZ patients	In vivo treatment in humans	1. PEA (*n*=25); 2. PLB (*n*=25)	50	1. Symptoms assessment (PANSS, HDRS); 2. Adverse events assessment (ESRS, open-ended questions, comprehensive side effect checklist)	1. **Effect on PANSS negative: time, ↓**; **time x treatment interaction, ↓**; 2. **Effect on PANSS positive: time, ↓**; time x treatment interaction, NS; 3. **Effect on PANSS general: time, ↓**; **time x treatment interaction, ↓**; 4. **Effect on PANSS total: time, ↓**; **time x treatment interaction, ↓**; 5. Effect on HDRS: time x treatment interaction, NS; 6. Effect on ESRS global score: time, NS; time x treatment interaction, NS; 7. Frequency of adverse events (drowsiness, dizziness, tremor,

							increased appetite, nervousness, restlessness, skin rash, blurred vision, fatigue, diarrhea, dry mouth, sore throat, tachycardia): PEA vs. PLB, NS
Abedini et al. (2022)	Iran	To assess PEA add-on effects on acute mania in BPAD patients	In vivo treatment in humans	1. PEA (*n*=32); 2. PLB (*n*=31)	63	1. Symptoms assessment (YMRS, HDRS); 2. Adverse events assessment (ESRS, open-ended questions, comprehensive side effect checklist)	1. Effect on psychometric measures: (a) **YMRS: time x treatment interaction**; (b) ESRS: time x treatment interaction, NS; 2. **YMRS global score**: (a) Baseline, (b) Week 2, (c) Week 4: PEA vs. PLB, NS; (d) **Week 6: PEA < PLB**; 3. **YMRS score changes**: (a) From Baseline to Week 2: PEA vs. PLB, NS; (b) **From Baseline to Week 4**, (c) **From Baseline to Week 6: PEA > PLB**; 4. **HDRS global score**: (a) Baseline: PEA vs. PLB, NS; (b) **Week 6: PEA > PLB**; 5. HDRS score changes: From Baseline to Week 6: PEA vs. PLB, NS; 6. ESRS global score: (a)

							Baseline, (b) Week 1, (c) Week 2, (d) Week 4, (e) Week 6: PEA vs. PLB, NS; 7. ESRS score changes: all comparisons, NS; 8. Frequency of adverse events (drowsiness, dizziness, increased appetite, skin rash, diarrhea, dry mouth, sore throat, tachycardia): PEA vs. PLB, NS
Stark et al. (2019)	Czech Republic/Italy	To assess PEA and other eCBs/AEs brain levels following CBD, CB1R antagonist/inverse agonist, and HAL in MAM rats	Quantitative assessment in animals	1. CTRL+VHI; 2. CTRL+CBD10; 3. CTRL+CBD30; 4. CTRL+AM251; 5. CTRL+HAL; 6. MAM+VHI; 7. MAM+CBD10; 8. MAM+CBD30;	12-15 per group	eCBs/AEs brain levels (LC-APCI/MS)	1. Effects of peripubertal treatment (PND 19-39) on social interactions (SI test) from PND 100: (a) **effect on time: MAM; treatment; MAM x treatment interaction**; (b) **time: MAM+VHI < CTRL+VHI; MAM+CBD30 > MAM+VHI; CTRL+AM251 < CTRL+VHI; MAM+AM251 > MAM+VHI; CTRL+VHI > CTRL+HAL**; other comparisons, NS; (c) effect on number of social interactions: MAM, NS; treatment, NS; MAM x

	9. MAM+AM251; 10. MAM+HAL	treatment interaction, NS; (d) number of interactions: all comparisons, NS; 2. Effects of peripubertal treatment (PND 19-39) on exploratory activity (NORT, OFT) from PND 100: (a) **effect on DI: MAM**; treatment, NS; **MAM x treatment interaction**; (b) **DI: MAM+VHI < CTRL+VHI; MAM+CBD30 > MAM+VHI**; other comparisons, NS; (c) effect on total exploration time: MAM, NS; treatment, NS; MAM x treatment interaction, NS; (d) total exploration time: all comparisons, NS; (e) effect on number of crossings: MAM, NS; treatment, NS; MAM x treatment interaction, NS; (f) number of crossings: all comparisons, NS; (g) effect on number of rearings: MAM, NS; treatment, NS; MAM x treatment interaction, NS; (h) number of

crossings: all comparisons, NS; 3. Effects of peripubertal treatment (PND 19-39) on PFC CB1R expression from PND 100: (a) **effect on mRNA expression**: MAM, NS; treatment, NS; **MAM x treatment interaction**; (b) **% mRNA methylation: MAM+VHI < CTRL+VHI; MAM+CBD30 > MAM+VHI; CTRL+AM251 > CTRL+VHI; CTRL+HAL > CTRL+VHI; MAM+HAL > MAM+VHI**; other comparisons, NS; (c**) mRNA fold change: MAM+VHI > CTRL+VHI; MAM+CBD30 < MAM+VHI; MAM+AM251 < MAM+VHI**; other comparisons, NS; (d) **protein expression level: MAM+VHI > CTRL+VHI**; **CTRL+CBD30 < CTRL+VHI; MAM+CBD30 < MAM+VHI; MAM+AM251 > CTRL+VHI; MAM+HAL >**

	CTRL+VHI; other comparisons, NS; 4. Effects of peripubertal treatment (PND 19-39) on PFC eCBs/AEs expression from PND 100: (a) effect on PEA levels: MAM, NS; treatment, NS; MAM x treatment interaction, NS; (b) PEA levels: all comparisons, NS; (c) **effect on 2-AG levels: MAM; treatment; MAM x treatment interaction**; (d) **2-AG levels: MAM+CBD30 < MAM+VHI; CTRL+AM251 > CTRL+VHI**; **MAM+AM251 < MAM+VHI; CTRL+HAL > CTRL+VHI**; other comparisons, NS; (e) effect on AEA levels: MAM; treatment, NS; MAM x treatment interaction; (f) **AEA levels: CTRL+CBD30 > CTRL+VHI**; other comparisons, NS; (g) effect on OEA levels: MAM, NS; treatment, NS; MAM x

							treatment interaction, NS; (b) OEA levels: all comparisons, NS
Di Bartolomeo et al. (2021)	Czech Republic/Italy	To assess PEA and other eCBs/AEs brain levels following CBD in pTHC rats	Quantitative assessment in animals	1. CTRL+VHI; 2. CTRL+CBD; 3. pTHC+VHI; 4. pTHC+CBD	3-20 per group	eCBs/AEs brain levels (LC/MS)	1. Effects of pTHC on neonatal behavior: (a) **righting (PND 1-2)**, (b) **cliff aversion (PND 2-8)**, (c) **forelimb placing (PND 3-9)**, (d) **forelimb grasping (PND 2-4)**, (e) **bar holding (PND 5)**, (f) **negative geotaxis (PND 3-4)**, (g) **nest time**: **pTHC+VHI < CTRL+VHI**; (h) nest exploration: pTHC+VHI vs. CTRL+VHI, NS 2. Effects of pTHC on PFC eCBs/AEs expression at PND 10: (a) PEA, (b) AEA, (c) OEA levels: pTHC+VHI vs. CTRL+VHI, NS; (d) **2-AG levels: pTHC+VHI < CTRL+VHI**; (e) **Magl**, (f) **Faah mRNA expression: pTHC+VHI > CTRL+VHI**; (g) Cnr1, (h) Trpv1, (i) other eCBs/AEs enzymes

	mRNA expression: pTHC+VHI vs. CTRL+VHI, NS; 3. Effects of pTHC on PFC Drd2 gene expression at PND 10: pTHC+VHI > CTRL+VHI (trend effect); 4. Effects of peripubertal CBD (PND 19-39) on adult behavior: (a) number of crossings, (b) number of rearings (OFT): all comparisons, NS; (c) **SI time: pTHC+VHI < CTRL+VHI**; **pTHC+CBD < pTHC+VHI**; (d) SI number of interactions: all comparisons, NS; (e) **discrimination index (NORT): pTHC+VHI < CTRL+VHI**; **pTHC+CBD > pTHC+VHI**; (f) time (NORT): all comparisons, NS; 5. Effects of peripubertal CBD (PND 19-39) on PFC genes expression from PND 100: (a) %DNA methylation Cnr1 gene: all comparisons, NS; (b) **%DNA**

	methylation Drd2 gene: pTHC+VHI < CTRL+VHI; **CTRL+CBD < CTRL+VHI**; **pTHC+CBD > pTHC+VHI**; other comparisons, NS; (c) **Cnr1 relative gene expression: pTHC+VHI > CTRL+VHI**; other comparisons, NS; (d) **Drd2 relative gene expression: pTHC+VHI > CTRL+VHI; pTHC+CBD > CTRL+CBD**; (e) Cnr1 protein expression level: all comparisons, NS; (f) **Drd2 protein expression level: pTHC+VHI > CTRL+VHI**; other comparisons, NS; 6. Effects of peripubertal CBD (PND 19-39) on PFC eCBs/AEs expression from PND 100: (a) PEA levels: CTRL+VHI vs. pTHC+VHI vs. pTHC+CBD, NS; (b) **2-AG levels: pTHC+VHI < CTRL+VHI**; **pTHC+CBD < CTRL+VHI**; pTHC+CBD vs.

		pTHC+VHI, NS; (c) **AEA levels: pTHC+VHI > CTRL+VHI**; pTHC+VHI vs. pTHC+CBD, NS; pTHC+CBD vs. CTRL+VHI, NS; (d) OEA levels: all comparisons, NS

<, Lower/less than; >, Greater/more than; ↑, Increase; ↓, Decrease; 2-AG, 2-Arachidonoylglycerol; AEA, Anandamide; AEs, Acylethanolamines; AM251, CB1 antagonist/inverse agonist; AMI, Amisulpride; AP-F, Antipsychotic-free patients; AP-T, Antipsychotic-treated patients; BPAD, Bipolar Affective Disorder; CAARMS, Comprehensive Assessment of At-Risk Mental State; CB, Cerebellum; CB1R, Cannabinoid receptor type 1; CBD, Cannabidiol; CBD10, Cannabidiol (10 mg/kg/day); CBD30, Cannabidiol (30 mg/kg/day); CHR, Clinical High-Risk; Cnr1, Cannabinoid CB1 receptor gene; CSF, Cerebrospinal fluid; CT, Childhood trauma; CTQ, Childhood Trauma Questionnaire; CTRL, Control group; CUD, Cannabis Use Disorder; DEA, Docosatetraenylethanolamide; DHEA, N-docosahexaenoylethanolamine; DI, Discrimination index; DNA, Deoxyribonucleic acid; Drd2, Dopamine D2 receptor gene; DUAL, Dual diagnosis; eCBs, Endocannabinoids; ELISA, Enzyme linked immunosorbent assay; ESRS, Extrapyramidal Symptom Rating Scale; FAAH/Faah, Fatty Acid Amide Hydrolase; FEP, First-episode psychosis; FEPb, First-episode psychosis (baseline); GC/MS, Gas Chromatography-Mass Spectrometry; HAL, Haloperidol; HC, Healthy controls; HDRS, Hamilton Depression Rating Scale; HIP, Hippocampus; HPLC, High Pressure Liquid Chromatography; HPLC/MS, High Pressure Liquid Chromatography-Mass Spectrometry; IL-6, Interleukin 6; LC/MS, Liquid Chromatography-Mass Spectrometry; LC-APCI/MS, Liquid Chromatography-Atmospheric Pressure Chemical Ionization-Mass Spectrometry; LEA, Dihomo-linolenoylethanolamine; Magl, Monoacylglycerol lipase; MAM, Methylazoxymethanol acetate; MDS-UPDRS, MDS-Unified Parkinson's Disease Rating Scale; mRNA, Messenger ribonucleic acid; n, Sub-sample size; NADA, N-Arachidonoyldopamine; ng/ml, Nanograms per milliliter; nM-EDL, Non-Motor Aspects of Experiences of Daily Living; NoPastDEP, No prior depressive episodes; NORT, Novel object recognition test; noBPAD, healthy twins of BPAD patients; noSCZ, healthy twins of SCZ patients; NS, Not significant; OEA, Oleoylethanolamide; OFT, Open-field test; PANSS, Positive and Negative Syndrome Scale; PastDEP, At least one prior depressive episode; PD, Parkinson's Disease; PEA, Palmitoylethanolamide; PFC, Prefrontal cortex; PLB, Placebo; PLTs, Platelets; PND, Postnatal day; pTHC, Perinatal THC; RF, Risk factor; SCZ, Schizophrenia; SI, Social interaction; THC, Delta-9-tetrahydrocannabinol; Trpv1, Transient receptor potential vanilloid 1 gene; VHI, Vehicle; VPA, Valproic Acid; vs., Compared to; YMRS, Young Mania Rating Scale. Bold font emphasizes statistically significant results.

Table 2a. Methodological quality of clinical studies investigating palmitoylethanolamide and its correlations to psychotic disorders.

Study (year)	Study design	Defined Study Population	Age (years, Mean ± SD)	Male Gender Count (%)	PEA Measure	Adequate PEA Evaluation	Control	Comparability of Subjects	Excluded/Adjusted for Confounding Factors	Statistical Analyses	Funding or Sponsorship
Leweke et al. (1999)	√ Analytic, observational	√ SCZ or schizophreniform disorder patients: DSM-IV; BPRS	√/X SCZ: 27.7 ± 9.6	√/X SCZ: 7 (70%)	√ CSF levels	√ Single assessment	√ HC	√ Matched for age	X	√ t-test	√
Leweke et al. (2012)	√ Analytic, observational	√ SCZ or schizophreniform disorder patients: DSM-IV; 18-50 years; BPRS ≥ 36; BPRS THOT factor ≥ 12	√ CBD: 29.7 ± 8.3; AMI: 30.6 ± 9.4	√ CBD: 15 (75%); AMI: 17 (89%)	√ Serum levels	√ Multiple assessment (baseline, day 14, day 28)	√ AMI	√/X Matched for age, weight, pulse, blood pressure, gender, PANSS, BPRS, SAS, EPS; not matched for CGI severity, Lorazepam mg/day	√ Excluded if positive UDS, history of SUDs, previous depot antipsychotic treatment (< 3 months before the	√ Mixed effects repeated measures model (unstructured covariance matrix), Fisher's exact test	√

								study), history of treatment resistance, present relevant/unstable condition, pregnancy, or breastfeeding		
Muguruza et al. (2013)	√ Analytic, observational	√ Postmortem SCZ patients' brain samples: DSM-IV	√ AP-F: 45 ± 4; AP-T: 49 ± 5; CTRL: 45 ± 3	√ AP-F: 9 (81.8%); AP-T: 6 (75%); CTRL: 15 (78.9%)	√ Brain tissue levels	√ Single assessment	√ CTRL	√ Matched for age, gender, PMI, pH, RIN, storage (months)	√ Excluded if positive toxicological test for cannabis; HC excluded if history of neuropsychiatric	√ Two-way ANOVA, Fisher LSD test, Pearson's coefficient, t-test, one-way ANOVA,	√

									disorder, history of drug abuse	Dunnett's multiple comparison post hoc test	
Koethe et al. (2018)	√ Analytic, observational	√ SCZ or BPAD discordant twin patients: DSM-III-R; stable clinical condition; SANS; clinical records review; GAF scale	√ SCZ discordant: 30; BPAD discordant: 33; HC twins: 31	√ SCZ discordant: 14 (56%); BPAD discordant: 1 (14.29%); HC twins: 3 (27.27%)	√ Plasma levels	√ Single assessment	√ noSCZ; noBPAD; HC	X	√ Excluded if positive UDS	√ Wilcoxon rank sum test, exact Wilcoxon signed rank test, Bonferroni's correction	√
Appiah-Kusi et al. (2020)	√ Analytic, observational	√ CHR individuals: CAARMS criteria	√ HC: 25.05 ± 4.90; CHR:	√ 1. HC: 53.00%; 2. CHR: 51.00%	√ Plasma levels	√ Single assessment	√ HC	√ Matched for age, gender, current CU	√ Excluded if history of psychotic or manic episode,	√ ANCOVA, t-test, chi-square,	√

			23.82 ± 5.28						past or current CNS disorder, current substance dependence (DSM-IV), IQ < 70, any contraindications to CBD treatment or MRI, drug use during the entire study	correlation analysis	
Ibarra-Lecue et al. (2022)	√ Analytic, observational	√ SCZ, CUD, or DUAL patients: SCID (DSM-	√ CUD: 32.5 ± 1.9; HC1:	√ CUD: 21 (80.77%) ;	√ Plasma levels	√ Single assessment	√ HC1; HC2; HC3	√ Matched for age, gender	√ HC excluded if any neuropsychi	√ Two-way ANOVA,	√

		IV, DSM-IV-TR); ICD	32.8 ± 2.0; SCZ: 48.9 ± 1.8; HC2: 49 ± 2.1; DUAL: 38.0 ± 2.9; HC3: 37.3 ± 3.4;	HC1: 19 (79.17%); SCZ: 13 (59.09%); HC2: 10 (52.63%); DUAL: 12 (92.31%); HC3: 10 (100%)					atric disease, any past 2 years CU	Tukey's test	
Parksepp et al. (2022)	√ Analytic, observational	√ FEP patients: ICD-10; DUP < 3 years; no AP use before	√ FEPb: 26.6 ± 6.1; FEP(0.6-year): 27.3 ± 6.4;	√ FEPb: 31 (57%); FEP(0.6-year): 27 (51%); FEP(5.1-	√ Serum levels	√ Multiple assessment (baseline, 0.59 ± 0.06 years after baseline, 5.15 ± 1.25	√ HC	√/X FEP and HC matched for age, gender, smoking status, BMI; FEP and HC not matched for length of	√ Excluded if current organic or drug-induced psychosis, current	√ Shapiro-Wilk test, t-test, repeated measure ANOVA, Scheffé	√

		the study; 18-45 years	FEP(5.1-year): 31.8 ± 5.9; HC: 24.7 ± 4.5	year): 23 (43%); HC: 24 (44%)		years after baseline)			education; FEP groups matched for AP dose; FEP groups not matched for BMI, BPRS score	psychotic disorders due to other medical conditions; Adjusted for gender, age at first visit, smoking status, Δt between the visits	post hoc test, chi-square test, LME models, maximum likelihood method, likelihood ratio test, FDR procedure	
Topuz et al. (2022)	√ Analytic, observational	√ BPAD patients: DSM-5; euthymic period; 18-65 years	√ 42.40 ± 1.10	√ 35 (44.30%)	√ Serum levels	√ Single assessment	X	NA		X	√ Shapiro-Wilk test, t-test, Mann-Whitney U test, Spearman correlation analysis	√

Brotini et al. (2017)	√ Analytic, observational, interventional	√ levodopa treated PD patients (PDSBB criteria): (a) HY scale > 0; (b) MMSE ≥26/30; (c) age>18 years; (d) levodopa therapy (eventually other PD medication) without modification over four consecutive weeks	√ 73 ± 8	√ 14 (46.67%)	√ um-PEA 600 mg	√ Twice per day administration (12 weeks), then daily administration (36 weeks)	X	NA	√ Excluded if other forms of parkinsonism, other forms of dementia, unreliable patients, non-compliant patients	√ GLMM, Wilcoxon signed-rank test, Bonferroni's correction, Tukey-Kramer adjusted test	√

Salehi et al. (2022)	√ Analytic, observational, interventional	√ SCZ patients: 18-60 years; SCID (DSM-5); illness duration ≥ 2 years; PANSS negative ≥ 15; HDRS < 14; clinical stability on stable risperidone (PANSS total change ≤ 20% on 2 subsequent assessments within 2 weeks)	√ PEA: 33.76 ± 6.93; PLB: 36.80 ± 9.60	√ 1. PEA: 23 (92%); 2. PLB: 21 (84%)	√ PEA 600 mg (oral administration)	√ Twice per day administration (8 weeks)	√ PLB	√ Matched for age, gender, literacy, smoking status, marital status, overall SCZ duration, baseline HDRS, baseline PANSS, baseline ESRS	√ Excluded if IQ < 70, history of head trauma, prior 3 months history of ECT, prior 6 months substance or alcohol dependence, breastfeeding, pregnancy, suicidal ideation, history of neurosurgery, current acute or chronic	√ Shapiro-Wilk test, Q-Q probability graphics, t-test, Levene's test, Fisher's exact test, ANOVA, Greenhouse-Geisser test	√

									medical disease, history of allergy to risperidone or PEA		
Abedini et al. (2022)	√ Analytic, observational, interventional	√ BPAD patients: SCID (DSM-5); MINI; moderate to severe manic episode assessed (YMRS)	√ PEA: 30.78 ± 9.80; PLB: 32.74 ± 9.04	√ 1. PEA: 21 (71.9%); 2. PLB: 20 (64.5%)	√ PEA 600 mg (oral administration)	√ Twice per day administration (6 weeks)	√ PLB	√ Matched for age, gender, education, smoking status, marital status, overall BPAD duration, baseline HDRS, baseline YMRS, baseline ESRS	√ Excluded if IQ < 70, history of allergy to lithium, risperidone, or PEA, substance dependence (except nicotine and caffeine), receiving manic-inducing medications	√ t-test, chi-square, Fisher's exact test, general linear model repeated-measures analysis, Mauchly test, Greenhouse-Geisser test	√

, metabolic disorders such as hypothyroidism or hyperthyroidism, current severe hepatic disease

<, Lower/less than; >, Greater/more than; ≤, Less than or equal to; ≥, More than or equal to; AMI, Amisulpride; ANCOVA, Analysis of Covariance; ANOVA, Analysis of Variance; AP, Antipsychotic; AP-F, Antipsychotic-free patients; AP-T, Antipsychotic-treated patients; BMI, Body Mass Index; BPAD, Bipolar Affective Disorder; BPRS, Brief Psychiatric Rating Scale; CAARMS, Comprehensive Assessment of At-Risk Mental State; CBD, Cannabidiol; CGI, Clinical Global Impression; CHR, Clinical High-Risk; CNS, Central nervous system; CSF, Cerebrospinal fluid; CTRL, Control group; CU, Cannabis use; CUD, Cannabis Use Disorder; DSM-5, Diagnostic and Statistical Manual of mental disorders, Fifth Edition; DSM-III-R, Diagnostic and Statistical Manual of mental disorders, Third Edition Revised; DSM-IV, Diagnostic and Statistical Manual of mental disorders, Fourth Edition; DSM-IV-TR, Diagnostic and Statistical Manual of mental disorders, Fourth Edition-Text Revision; DUAL, Dual diagnosis; DUP, Duration of untreated psychosis; ECT, Electroconvulsive therapy; EPS, Extrapyramidal Symptoms; ESRS, Extrapyramidal Symptom Rating Scale; FDR, False discovery rate; FEP, First-episode psychosis; FEPb, First-episode psychosis (baseline); GAF, Global Assessment of Functioning; GLMM, Generalized Linear Mixed Model; HC, Healthy controls; HDRS, Hamilton Depression Rating Scale; HY, Hoehn and Yahr; ICD, International Classification of Diseases; ICD-10, International Classification of Diseases, Tenth Edition; IQ, Intelligence Quotient; LME, Linear mixed-effects; LSD, Least Significant Difference; mg, Milligrams; mg/day, Milligrams per day; MINI, Mini-International Neuropsychiatric Interview; MMSE, Mini Mental State Examination; MRI, Magnetic Resonance imaging; NA, Not applicable; noBPAD, healthy twins of BPAD patients; noSCZ, healthy twins of SCZ patients; PANSS, Positive and Negative Syndrome Scale; PD, Parkinson's Disease; PDSBB, PD Society Brain Bank; PEA, Palmitoylethanolamide; PLB, Placebo; PMI, Postmortem interval; RIN, RNA intergrity number; SANS, Scale for the Assessment of Negative Symptoms; SAS, Social Anxiety Scale; SCID, Structured Clinical Interview for DSM; SCZ, Schizophrenia; SD, Standard Deviation; SUDs, Substance Use Disorders; THOT, Thought Disorder; UDS, Urine drugs screening; um-PEA, Ultramicronized-PEA; YMRS, Young Mania Rating Scale; Δt, Time period.

Table 2b. Methodological quality of preclinical studies investigating palmitoylethanolamide and its correlations to psychotic disorders.

Study (year)	Study Design	Defined Study Population	Adequate Psychosis Model	Developmental Stage	Gender	PEA Measure	Adequate PEA Evaluation	Control	Statistical Analyses	Funding or Sponsorship
Stark et al. (2019)	√ Analytic, observational	√ Offsprings of MAM-exposed Sprague-Dawley rats	√ MAM 22 mg/Kg ip administration on GD 17	√ from PND 100	√ Male	√ Brain tissue levels	√ Single assessment	√ CTRL+VHI; CTRL+CBD10; CTRL+CBD30; CTRL+AM251; CTRL+HAL	√ Shapiro-Wilk test, two-way ANOVA, Fisher's LSD, t-test	√
Di Bartolomeo et al. (2021)	√ Analytic, observational	√ Offsprings of THC-exposed Sprague-Dawley rats	√ THC 5 mg/Kg oral administration from GD 15 to PND 9	√ PND 10; from PND 100	√ Male	√ Brain tissue levels	√ Double assessment	√ CTRL+VHI; CTRL+CBD	√ two-way ANOVA, Bonferroni's correction, Mann-Whitney U test, t-test, Shapiro-Wilk test	√

AM251, CB1 antagonist/inverse agonist; ANOVA, Analysis of Variance; CBD, Cannabidiol; CBD10, Cannabidiol (10 mg/kg/day); CBD30, Cannabidiol (30 mg/kg/day); CTRL, Control group; GD, Gestational day; HAL, Haloperidol; ip, intra-peritoneal; LSD, Least Significant Difference; MAM, Methylazoxymethanol acetate; mg/Kg, Milligrams per kilogram; PEA, Palmitoylethanolamide; PND, Postnatal day; THC, Delta-9-tetrahydrocannabinol; VHI, Vehicle.

3.2. In vivo palmitoylethanolamide add-on treatment exposure in humans with different types of psychoses or psychotic symptoms

Three human studies have addressed this area (Table 1) using similar but not overlapping methodologies in terms of study population (chronic schizophrenia (SCZ) patients [43], bipolar disorder (BPAD) patients with manic symptoms [44], Parkinson's disease (PD) patients [45]), PEA formulation (oral native PEA [43, 44], oral Ultramicronized (um)-PEA [45]), PEA dosage (600 milligrams (mg) daily [45], 600 mg twice/daily [43-45]), and PEA period of exposure (6 weeks [44], 8 weeks [43], 12 months [45]). Apart from a single study lacking a controlled condition [45], all studies adopted a randomized, double-blind, placebo-controlled design [43, 44]. Overall, results indicated a beneficial effect of PEA adjunctive therapy on residual negative and general psychopathological symptoms, but not positive symptoms, of risperidone-treated SCZ patients [43], as well as on manic symptoms of lithium- plus risperidone-treated BPAD patients [44]. Coherent data emerged regarding depressive symptomatology, which did not appear to be improved in both SCZ and BPAD patients treated with PEA add-on as compared to placebo [43, 44]. A single study addressing the effect of PEA over non-motor symptoms among levodopa-treated PD patients, showed no reduction in the number of subjects presenting with hallucinations and psychosis [45]. Noteworthy, PEA was well tolerated, in the absence of extrapyramidal symptoms or any other relevant side effect across the three studies, and for the entire duration of the compound administration.

3.3. Palmitoylethanolamide quantitative blood assessment in humans with psychosis clinical high-risk state

This systematic reappraisal identified a single human study specifically assessing peripheral blood PEA levels in individuals suffering from CHR state, as compared to healthy controls (HC) [46] (Table 1). PEA levels tended to be elevated in CHR patients, even though just approaching statistical significance. Intriguingly, PEA levels appeared to be significantly higher in those who were both CHR and had been exposed to childhood trauma (CT), compared to individuals having none of the above-mentioned risk factors or one risk factor alone. Furthermore, a significant positive correlation between PEA levels and the severity of CHR state and CT was observed [46].

3.4. Palmitoylethanolamide quantitative blood assessment in humans with different types of psychoses at different stages of illness

Most of the studies retrieved investigated peripheral blood PEA levels in patients with non-affective (e.g., schizophrenia (SCZ) [47-50] and schizophreniform psychosis [47]) or affective (e.g., bipolar disorder (BPAD) [48, 51]) psychoses at different stages of illness (Table 1). Two studies converged on the evidence of higher PEA levels in SCZ patients than in healthy controls (HC) [48, 49], one of which further suggesting significantly higher PEA plasma concentration in SCZ patients compared to patients meeting criteria for cannabis use disorder (CUD) or dual diagnosis of CUD and SCZ [49]. Interestingly, compared to HC, unaffected twin siblings of SCZ patients also showed increased PEA levels, that did not differ from those of patients [48]. Remarkably, antipsychotic (AP)-naïve first-episode psychosis (FEP) patients compared to HC showed a tendency to elevated PEA plasma levels and a significantly higher PEA/2-arachidonoylglycerol (2-AG) ratio, with both that subsided an average of 0.6 and 5.1 years after the initiation of AP treatment [50]. The modulating effect of AP treatment over acylethanolamines (AEs) levels was also investigated through a double-blind, randomized, parallel-group, controlled clinical trial, showing elevated serum PEA concentration in cannabidiol (CBD)-treated SCZ patients, compared with those treated with the antipsychotic amisulpride [47]. Studies measuring PEA levels among BPAD patients showed a less pronounced increase in affected and unaffected siblings of illness-discordant twin couples, when compared to HC [48]. Further, a higher PEA plasma concentration was found in BPAD patients having first episode as depression than in those who had their first episode as mania [51] as well as in those who had at least one depressive episode than in patients who had no prior depressive episodes [51]. Finally, PEA levels were increased according to the number of depressive episodes and the presence of depressive mood and anxiety, while inversely correlating with the number of hypomanic episodes, the number of hospitalizations, the duration of valproate (VPA) treatment, sexual desire, presence of flight of ideas, delusion, and grandiosity [51].

3.5. Palmitoylethanolamide quantitative central nervous system assessment in humans with schizophrenia

Two studies analyzed PEA levels in the CNS of patients with psychosis [52, 53] (Table 1). In particular, PEA levels were reported to be elevated in the cerebrospinal fluid (CSF) of SCZ and schizophreniform psychosis patients compared with healthy controls (HC) [52]. Conversely, a study on postmortem brain samples from subjects diagnosed with SCZ compared to controls indicated lower PEA levels in the cerebellum of antipsychotic (AP)-free patients only [53]. No significant differences in PEA brain quantification were detected among all study groups in the other brain areas investigated [53].

3.6. Palmitoylethanolamide quantitative brain tissue assessment in animal models of schizophrenia

In total, two studies evaluated PEA levels in the prefrontal cortex (PFC) [54, 55], hippocampus (HIP) [54], and nucleus accumbens (NAc) [54] of rats exposed to either prenatal methylazoxymethanol acetate (MAM) [54] or perinatal delta-9-tetrahydrocannabinol (THC) [55], which present with many SCZ-relevant biobehavioral deficits at neonatal age [55] and adulthood [54, 55] (Table 1). While modulating endocannabinoids (eCBs; e.g., anandamide (AEA) and 2-acylglycerol (2-AG) [54, 55]) and other acylethanolamines (AEs; e.g., oleoylethanolamide (OEA) [54]), both MAM and THC exposure did not significantly affect PEA levels in all investigated brain areas, as neither did the peripubertal exposure to cannabidiol (CBD) [54, 55], Cannabinoid receptor type 1 (CB1)-antagonist/agonist AM251 [54], and first-generation antipsychotic haloperidol (HAL) [54], compared to control conditions.

4. Discussion

This is the first systematic review of all evidence exploring the biobehavioral correlates of palmitoylethanolamide (PEA) in psychosis across human and animal studies. Unlike previous research in this field [38-40], the greatest majority of records included consisted of human studies. Existing reviews focusing on the role of the major phytocannabinoid cannabidiol (CBD) as a potential treatment for schizophrenia (SCZ) patients [20, 56, 57] gathered still preliminary evidence supporting its antipsychotic (AP) efficacy, while highlighting its advantageous side effect profile and good tolerability. Targeting similar pathways, PEA may be considered as a viable alternative to CBD with implications for many therapeutic areas, due to its established safety and the development of formulations maximizing its bioavailability [33, 37].

Overall, the present review demonstrated that PEA may be involved in different psychotic phenotypes. Also, it found initial evidence that PEA levels may reflect the severity of the disorder as well as the stage of illness. Evidence was obtained from both interventional studies addressing the AP potential of PEA supplementation, and observational studies of PEA tone in peripheral blood and in the central nervous system (CNS) in the context of clinical high-risk (CHR) for psychosis and psychotic disorders at different stages of illness.

Some important findings from this systematic review deserve to be highlighted. First, despite its promise, evidence regarding the therapeutic potential of PEA supplementation for psychosis is still

limited [43-45] and provides findings about selective efficacy on specific symptoms. In particular, PEA add-on to conventional psychotropic medications did not significantly reduce positive psychotic symptoms [43-45], while ameliorating negative psychotic [43] and acute manic [44] symptoms. Importantly, while not being the focus of this review, results presented here are inconclusive about a potential role of PEA in ameliorating depressive symptoms among psychosis patients [44]. Also, in only one study participants were clearly asked to avoid forms of cognitive behavioral therapies during the trial period [43], thus requiring future interventional studies to clearly rule out a potential masking effect of add-on psychotherapy over symptoms.

Second, a line of research investigating endocannabinoids (eCBs) tone in blood among subjects with genetic vulnerability to psychosis (i.e., unaffected twins of SCZ patients) [48], CHR individuals [46], untreated first-episode psychosis (FEP) patients [50], and longer-course SCZ patients [48, 49], converged on the evidence of increased PEA plasma levels as compared to healthy subjects. Also, among CHR patients, the more severe the clinical picture (i.e., greater symptom severity) and the risk profile (i.e., more severe childhood trauma), the higher were the PEA levels [46]. Based on PEA-related neurobiological mechanisms, the finding of augmented PEA release across different populations may reflect an endogenous attempt to restore homodynamic balance under disease conditions [28, 29]. However, follow-up studies revealed that PEA plasma levels are no longer heightened in AP-treated SCZ patients after 0.6 years of treatment, and further decreased at 5.1 years from baseline [50], potentially suggesting that such biological self-regulation of PEA levels is lost as the diseases progresses. Interestingly, a 2- to 4-week CBD treatment among SCZ patients was associated with higher PEA levels [47] when compared with a 2- to 4-week AP treatment [47], possibly indicating a CBD-specific property in sustaining PEA tone.

Further, differently from SCZ spectrum disorders, PEA plasma level increase was less pronounced in bipolar disorder (BPAD) patients and unaffected twins of BPAD patients, when compared to healthy subjects [48], perhaps accounting for existing phenotypical discrepancies within the affective psychosis group. In fact, PEA plasma levels were greater among BPAD patients having first episode as depression and increased consistently as a function of the lifetime number of depressive episodes [51]. Instead, the occurrence of positive psychotic symptoms, manic symptoms, and hypomanic episodes, was correlated with reduced PEA tone [51]. Based on evidence that BPAD patients with manic onset have an older age at diagnosis and a longer duration of untreated illness than those with depressive onset [58], and that polarity of episodes over time often reflects polarity at onset [59], such different findings among BPAD patients may suggest a changing pattern in PEA levels over time. In other words, PEA plasma levels would be higher in the first phases of the disorder, while declining

because of disease progression, in line with what observed for non-affective psychosis, and thus strengthening the rationale for its supplementation.

Third, CNS PEA levels were found to be augmented in the cerebrospinal fluid (CSF) of young adults with SCZ and schizophreniform disorder [52], whereas reduced in postmortem cerebellar samples of AP-free middle-aged SCZ patients [53], as compared to healthy controls. Similarly, brain PEA levels did not differ between AP-treated middle-aged SCZ patients and healthy controls [53]. Again, it can be assumed that PEA levels are elevated in the early stages of psychosis, potentially reflecting an innate compensatory mechanism, before dropping concomitantly to disease progression. With reference to AP treatment, while on one hand it did not appear to increase PEA levels, on the other it is not clear if it prevented a more pronounced longer-term decrease [53].

Only two studies assessed PEA levels in the prefrontal cortex (PFC) of preclinical models of psychosis. They did not show any significant differences as compared to the control condition [54, 55]. Further animal studies will need to yield a more robust understanding of the neurobiological mechanisms involving PEA in psychosis.

The findings of this review should be seen considering some strengths and limitations. Despite supporting PEA tone alterations in psychosis and effectiveness of PEA supplementation as a therapeutic strategy, research in this field needs to be expanded, especially to fully comprehend the potential of PEA supplementation as add-on therapy for psychosis across all its symptoms dimensions. Indeed, evidence points toward a beneficial effect of PEA over negative psychotic symptoms and acute manic symptoms, reasonably due to the protective role of the compound against altered neuroinflammation and related mechanisms [28, 29], while PEA effect over positive psychotic symptoms remains to be clarified. Further, future follow-up studies will have to investigate whether PEA effect is sustained in the longer-term. Besides, to date, evidence of PEA effect as monotherapy in the clinical stages preceding full-blown psychosis onset to prevent the risk of progression is totally lacking and is worth of exploration. Finally, PEA levels appear to be altered in psychosis. However, further clarification is needed as to whether PEA tone is altered to a different extent depending on the stage of illness and whether this can be considered a biomarker of psychosis. In line with this, whether any AP treatment may be beneficial through PEA tone modulation requires additional investigation. Biobehavioral comparisons between CBD and PEA are also worth of consideration, especially whether CBD antipsychotic effects are mediated by PEA signaling and similar effects can be obtained directly supplementing PEA. The latter would be inevitably more advantageous, due to its safer profile and shorter biological distance from the therapeutic target.

5. Conclusions

This systematic review provided a first overview of all observational and experimental studies of PEA and its biobehavioral correlates in psychosis. Although in its infancy and still limited, research in this field is primarily carried out on humans and provides evidence for both alterations in PEA signaling, implications for psychosis-related behavioral features, and benefits from PEA supplementation. In particular, PEA may be useful to improve negative psychotic symptoms and manic symptoms. Noteworthy, no serious adverse events were reported across all human studies investigating its administration, further supporting PEA potential effectiveness and elevated safety as a therapeutic intervention in psychosis.

References

1. Kraepelin, E., *Die Erscheinungsformen des Irreseins: (The manifestations of insanity).* History of Psychiatry, 1992. **3**(12): p. 509–529.
2. Potash, J.B. and O.J. Bienvenu, *Neuropsychiatric disorders: Shared genetics of bipolar disorder and schizophrenia.* Nat Rev Neurol, 2009. **5**(6): p. 299-300.
3. Craddock, N., M.C. O'Donovan, and M.J. Owen, *Psychosis genetics: modeling the relationship between schizophrenia, bipolar disorder, and mixed (or "schizoaffective") psychoses.* Schizophr Bull, 2009. **35**(3): p. 482-90.
4. Craddock, N. and M.J. Owen, *The Kraepelinian dichotomy - going, going ... but still not gone.* British Journal of Psychiatry, 2010. **196**(2): p. 92-95.
5. Ivleva, E.I., et al., *Genetics and intermediate phenotypes of the schizophrenia--bipolar disorder boundary.* Neurosci Biobehav Rev, 2010. **34**(6): p. 897-921.
6. Fusar-Poli, L., A. Aguglia, and U. Albert, *Editorial: Early identification of affective and non-affective psychoses: From psychopathology to biomarkers.* Front Psychiatry, 2023. **14**: p. 1144943.
7. Perala, J., et al., *Lifetime prevalence of psychotic and bipolar I disorders in a general population.* Arch Gen Psychiatry, 2007. **64**(1): p. 19-28.
8. Whiteford, H.A., et al., *Global burden of disease attributable to mental and substance use disorders: findings from the Global Burden of Disease Study 2010.* Lancet, 2013. **382**(9904): p. 1575-86.
9. Vargas, T.G. and V.A. Mittal, *The Critical Roles of Early Development, Stress, and Environment in the Course of Psychosis.* Annu Rev Dev Psychol, 2022. **4**(1): p. 423-445.
10. Howes, O.D., et al., *The nature of dopamine dysfunction in schizophrenia and what this means for treatment.* Arch Gen Psychiatry, 2012. **69**(8): p. 776-86.
11. Howes, O.D. and R.M. Murray, *Schizophrenia: an integrated sociodevelopmental-cognitive model.* Lancet, 2014. **383**(9929): p. 1677-1687.
12. Howes, O., R. McCutcheon, and J. Stone, *Glutamate and dopamine in schizophrenia: an update for the 21st century.* J Psychopharmacol, 2015. **29**(2): p. 97-115.
13. Howes, O.D., et al., *The Role of Genes, Stress, and Dopamine in the Development of Schizophrenia.* Biol Psychiatry, 2017. **81**(1): p. 9-20.
14. Andrews, G., et al., *Cost-effectiveness of current and optimal treatment for schizophrenia.* Br J Psychiatry, 2003. **183**: p. 427-35; discussion 436.
15. Obradovic, M., A. Mrhar, and M. Kos, *Cost-effectiveness of antipsychotics for outpatients with chronic schizophrenia.* Int J Clin Pract, 2007. **61**(12): p. 1979-88.

16. Giuffrida, A., et al., *Cerebrospinal anandamide levels are elevated in acute schizophrenia and are inversely correlated with psychotic symptoms.* Neuropsychopharmacology, 2004. **29**(11): p. 2108-14.
17. Koethe, D., C. Hoyer, and F.M. Leweke, *The endocannabinoid system as a target for modelling psychosis.* Psychopharmacology (Berl), 2009. **206**(4): p. 551-61.
18. Appiah-Kusi, E., et al., *Abnormalities in neuroendocrine stress response in psychosis: the role of endocannabinoids.* Psychol Med, 2016. **46**(1): p. 27-45.
19. Colizzi, M., et al., *Effect of cannabis on glutamate signalling in the brain: A systematic review of human and animal evidence.* Neurosci Biobehav Rev, 2016. **64**: p. 359-81.
20. Schubart, C.D., et al., *Cannabidiol as a potential treatment for psychosis.* Eur Neuropsychopharmacol, 2014. **24**(1): p. 51-64.
21. McGuire, P., et al., *Cannabidiol (CBD) as an Adjunctive Therapy in Schizophrenia: A Multicenter Randomized Controlled Trial.* Am J Psychiatry, 2018. **175**(3): p. 225-231.
22. Bhattacharyya, S., et al., *Effect of Cannabidiol on Medial Temporal, Midbrain, and Striatal Dysfunction in People at Clinical High Risk of Psychosis: A Randomized Clinical Trial.* JAMA Psychiatry, 2018. **75**(11): p. 1107-1117.
23. Appiah-Kusi, E., et al., *Effects of short-term cannabidiol treatment on response to social stress in subjects at clinical high risk of developing psychosis.* Psychopharmacology (Berl), 2020. **237**(4): p. 1121-1130.
24. Kelly, J.R., et al., *The role of the gut microbiome in the development of schizophrenia.* Schizophr Res, 2021. **234**: p. 4-23.
25. Minichino, A., et al., *Endocannabinoid system mediates the association between gut-microbial diversity and anhedonia/amotivation in a general population cohort.* Mol Psychiatry, 2021. **26**(11): p. 6269-6276.
26. Khandaker, G.M., et al., *Inflammation and immunity in schizophrenia: implications for pathophysiology and treatment.* Lancet Psychiatry, 2015. **2**(3): p. 258-270.
27. Fraguas, D., et al., *Oxidative Stress and Inflammation in First-Episode Psychosis: A Systematic Review and Meta-analysis.* Schizophr Bull, 2019. **45**(4): p. 742-751.
28. Petrosino, S. and V. Di Marzo, *The pharmacology of palmitoylethanolamide and first data on the therapeutic efficacy of some of its new formulations.* Br J Pharmacol, 2017. **174**(11): p. 1349-1365.
29. Petrosino, S. and A. Schiano Moriello, *Palmitoylethanolamide: A Nutritional Approach to Keep Neuroinflammation within Physiological Boundaries-A Systematic Review.* Int J Mol Sci, 2020. **21**(24).

30. Skaper, S.D. and L. Facci, *Mast cell-glia axis in neuroinflammation and therapeutic potential of the anandamide congener palmitoylethanolamide.* Philos Trans R Soc Lond B Biol Sci, 2012. **367**(1607): p. 3312-25.
31. Skaper, S.D., L. Facci, and P. Giusti, *Glia and mast cells as targets for palmitoylethanolamide, an anti-inflammatory and neuroprotective lipid mediator.* Mol Neurobiol, 2013. **48**(2): p. 340-52.
32. Petrosino, S., et al., *Palmitoylethanolamide counteracts substance P-induced mast cell activation in vitro by stimulating diacylglycerol lipase activity.* J Neuroinflammation, 2019. **16**(1): p. 274.
33. Clayton, P., et al., *Palmitoylethanolamide: A Natural Compound for Health Management.* Int J Mol Sci, 2021. **22**(10).
34. De Petrocellis, L., J.B. Davis, and V. Di Marzo, *Palmitoylethanolamide enhances anandamide stimulation of human vanilloid VR1 receptors.* FEBS Lett, 2001. **506**(3): p. 253-6.
35. Petrosino, S., et al., *The anti-inflammatory mediator palmitoylethanolamide enhances the levels of 2-arachidonoyl-glycerol and potentiates its actions at TRPV1 cation channels.* Br J Pharmacol, 2016. **173**(7): p. 1154-62.
36. Couch, D.G., et al., *Palmitoylethanolamide and Cannabidiol Prevent Inflammation-induced Hyperpermeability of the Human Gut In Vitro and In Vivo-A Randomized, Placebo-controlled, Double-blind Controlled Trial.* Inflamm Bowel Dis, 2019. **25**(6): p. 1006-1018.
37. Clayton, P., et al., *Palmitoylethanolamide: A Potential Alternative to Cannabidiol.* J Diet Suppl, 2021: p. 1-26.
38. Colizzi, M., et al., *Palmitoylethanolamide and Its Biobehavioral Correlates in Autism Spectrum Disorder: A Systematic Review of Human and Animal Evidence.* Nutrients, 2021. **13**(4).
39. Bortoletto, R., et al., *Is It Time to Test the Antiseizure Potential of Palmitoylethanolamide in Human Studies? A Systematic Review of Preclinical Evidence.* Brain Sciences, 2022. **12**(1).
40. Colizzi, M., et al., *Therapeutic effect of palmitoylethanolamide in cognitive decline: A systematic review and preliminary meta-analysis of preclinical and clinical evidence.* Frontiers in Psychiatry, 2022. **13**.
41. Parums, D.V., *Editorial: Review Articles, Systematic Reviews, Meta-Analysis, and the Updated Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 Guidelines.* Med Sci Monit, 2021. **27**: p. e934475.

42. West, S., et al., *Systems to rate the strength of scientific evidence.* Evid Rep Technol Assess (Summ), 2002(47): p. 1-11.
43. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.
44. Abedini, T., et al., *Efficacy and safety of palmitoylethanolamide as an adjunctive treatment for acute mania: A randomized, double-blind, placebo-controlled trial.* Psychiatry Clin Neurosci, 2022. **76**(10): p. 505-511.
45. Brotini, S., C. Schievano, and L. Guidi, *Ultra-micronized Palmitoylethanolamide: An Efficacious Adjuvant Therapy for Parkinson's Disease.* CNS Neurol Disord Drug Targets, 2017. **16**(6): p. 705-713.
46. Appiah-Kusi, E., et al., *Childhood trauma and being at-risk for psychosis are associated with higher peripheral endocannabinoids.* Psychol Med, 2020. **50**(11): p. 1862-1871.
47. Leweke, F.M., et al., *Cannabidiol enhances anandamide signaling and alleviates psychotic symptoms of schizophrenia.* Transl Psychiatry, 2012. **2**(3): p. e94.
48. Koethe, D., et al., *Familial abnormalities of endocannabinoid signaling in schizophrenia.* World Journal of Biological Psychiatry, 2019. **20**(2): p. 117-125.
49. Ibarra-Lecue, I., et al., *Cannabis use selectively modulates circulating biomarkers in the blood of schizophrenia patients.* Addiction Biology, 2022. **27**(6).
50. Parksepp, M., et al., *The Expanded Endocannabinoid System Contributes to Metabolic and Body Mass Shifts in First-Episode Schizophrenia: A 5-Year Follow-Up Study.* Biomedicines, 2022. **10**(2).
51. Topuz, R.D., Y. Gorgulu, and M.K. Uluturk, *Could serum endocannabinoid and N-acylethanolamine levels be important in bipolar disorder?* World Journal of Biological Psychiatry: p. 7.
52. Leweke, F.M., et al., *Elevated endogenous cannabinoids in schizophrenia.* NeuroReport, 1999. **10**(8): p. 1665-1669.
53. Muguruza, C., et al., *Quantification of endocannabinoids in postmortem brain of schizophrenic subjects.* Schizophrenia Research, 2013. **148**(1-3): p. 145-150.
54. Stark, T., et al., *Peripubertal cannabidiol treatment rescues behavioral and neurochemical abnormalities in the MAM model of schizophrenia.* Neuropharmacology, 2019. **146**: p. 212-221.

55. Di Bartolomeo, M., et al., *Crosstalk between the transcriptional regulation of dopamine D2 and cannabinoid CB1 receptors in schizophrenia: Analyses in patients and in perinatal Δ9-tetrahydrocannabinol-exposed rats.* Pharmacol Res, 2021. **164**: p. 105357.
56. Bonaccorso, S., et al., *Cannabidiol (CBD) use in psychiatric disorders: A systematic review.* Neurotoxicology, 2019. **74**: p. 282-298.
57. Ghabrash, M.F., et al., *Cannabidiol for the treatment of psychosis among patients with schizophrenia and other primary psychotic disorders: A systematic review with a risk of bias assessment.* Psychiatry Res, 2020. **286**: p. 112890.
58. Wang, Z., et al., *Differences in Demographic and Clinical Characteristics of Patients With Depressive vs. Manic First Episode of Bipolar Disorder.* Front Psychiatry, 2021. **12**: p. 616415.
59. Perugi, G., et al., *Polarity of the first episode, clinical characteristics, and course of manic depressive illness: a systematic retrospective investigation of 320 bipolar I patients.* Compr Psychiatry, 2000. **41**(1): p. 13-8.

Acknowledgement: The authors would like to aknowledge infrastructures from the Department of Medicine (DMED) – University of Udine (Italy).

Funding: This work received no external funding.

CRediT authorship contribution statement:

RB: Conceptualization, Methodology, Investigation, Software, Data Curation, Writing—Original Draft Preparation, Writing—Review and Editing, Validation, Resources, Visualization, Supervision, and Project Administration

FP: Conceptualization, Methodology, Validation, Resources, Writing—Review and Editing, and Visualization

AC: Conceptualization, Methodology, Validation, Resources, Writing—Review and Editing, and Visualization

SB: Conceptualization, Methodology, Validation, Resources, Writing—Review and Editing, and Visualization

MB: Conceptualization, Methodology, Investigation, Validation, Resources, Writing—Review and Editing, and Visualization

MC: Conceptualization, Methodology, Investigation, Software, Data Curation, Writing—Original Draft Preparation, Writing—Review and Editing, Validation, Resources, Visualization, Supervision, and Project Administration

CHAPTER IV

The Endocannabinoid Activity Remodulation for Psychosis Liability in Youth (EARLY) Study: An Open-Label Feasibility Trial of Ultramicronized-Palmitoylethanolamide Oral Supplementation in Clinical High-Risk State for Psychosis

Bortoletto R, Garzitto M, Piscitelli F, Fornasaro S, Scipioni C, Sepulcri O, Fabris M, Curcio F, Balestrieri M, Colizzi M. The Endocannabinoid Activity Remodulation for Psychosis Liability in Youth (EARLY) Study: An Open-Label Feasibility Trial of Ultramicronized-Palmitoylethanolamide Oral Supplementation in Clinical High-Risk State for Psychosis. Brain Sci. 2024 Dec 7;14(12):1230. doi: 10.3390/brainsci14121230. PMID: 39766429; PMCID: PMC11727594.

Abstract

To date, no psychotropic medication has shown to effectively halt progression to psy-chosis among individuals at Clinical High-Risk for psychosis (CHR), fueling the search for novel therapeutic agents. Recent evidence supports Palmitoylethanolamide (PEA) signaling as a poten-tial psychosis biomarker, also indicating a therapeutic role for its supplementation in the treat-ment of psychotic disorders. Nonetheless, the effect of sustained PEA intake in CHR subjects has never been explored so far. We will assess the feasibility of enrolling 20 CHR young adults pre-senting with attenuated psychotic symptoms (APS) in a 12-week, open-label, investiga-tor-initiated, proof-of-concept, single-arm trial of ultramicronized-PEA (um-PEA) 600 mg/day. Once completed the 12-week phase, participants will be proposed to enter a 24-week extension phase of the study. We will examine um-PEA ability to reduce APS and psychic distress, um-PEA safety and tolerability, and the biological basis of um-PEA effect in terms of modulation of in-flammatory response, endocannabinoid (eCB) signaling, and microbiome composition. Our trial aims to address an unmet clinical need in CHR subjects, providing an initial solid basis for the development of future studies evaluating the efficacy and tolerability of PEA supplementation in this group of patients.

Keywords

Schizophrenia; Cannabidiol; Nutraceuticals; Prevention; Transition-age Psychiatry

1. Introduction

The Clinical High-Risk (CHR) for psychosis state has been first described over two decades ago and represents the phase preceding the potential development of full-blown psychosis [1]. Recent meta-analyses of risk of transitioning to psychosis at 2 years among CHR individuals show rates of approximately 16-19%, while risk factors accumulation (risk enrichment) results in increasing rates of up to 35% at 10 years [2, 3]. Compared to the outdated "prodrome" concept, the CHR paradigm provides a less deterministic understanding of schizophrenia (SCZ), thus highlighting the importance of detection, assessment, and early intervention on milder symptoms preceding psychosis onset to halt disease progression [4-6]. However, to date, no psychotherapeutic or psychopharmacological interventions have shown to effectively prevent progression to psychosis among CHR subjects [3, 7], nor to relieve them from attenuated psychotic symptoms (APS) [8], negative symptoms [9], or impaired global functioning [10]. Compelling evidence of the endocannabinoid (eCB) system early disruption in the neurophysiological pathway leading to schizophrenia onset has fostered the search for novel medications (e.g., cannabidiol, CBD) targeting the eCB system for both psychosis patients and CHR individuals [11-16].

To this extent, growing research is focusing on the role of the eCB-like compound Palmitoylethanolamide (PEA) as a potential therapeutic agent in the context of several neuropsychiatric conditions [17-19]. PEA is a naturally occurring N-Acylethanolamine (AE), proven to modulate the central and peripheral neuroimmune systems and to exert protective functions against glutamate neurotoxicity [20-22]. It directly activates the Peroxisome Proliferator Activated Receptor-α (PPAR-α) and G protein-coupled receptor 55 (GPR55), allosterically modulates the Transient Receptor Potential Vanilloid 1 (TRPV1), and indirectly interacts with type 1- (CB1) and type 2- (CB2) cannabinoid receptors [23], sharing similar pharmacodynamic properties with the phytocannabinoid CBD [24]. We recently conducted a systematic review aimed at investigating the biobehavioral role of PEA in psychotic disorders [25]. Overall, evidence from clinical studies included in our reappraisal converges on the involvement of PEA in different psychotic phenotypes. A robust strain of research indicates an endogenous attempt of PEA to restore homodynamic balance under disease conditions in CHR individuals [26], non-affective psychosis patients [27-30], and affective psychosis patients [28, 31] at different stages of illness. Specifically, PEA blood tone would increase in the initial phases of a psychotic disorder, while subsiding concomitantly to disease progression. Consistently, PEA levels in the central nervous system (CNS) were found to be elevated in the early stages of SCZ [32], before declining as the disease progresses [33]. Limited but promising evidence was also obtained from interventional studies assessing the potential of PEA therapeutic supplementation in SCZ patients [34], bipolar disorder patients with manic symptoms [35], and Parkinson's disease patients

with psychotic symptoms [36]. PEA showed a selective efficacy on negative psychotic (e.g., alogia) [34] and acute manic [35] symptoms, while not significantly ameliorating positive psychotic symptoms (e.g., delusions) [34-36]. To the best of our knowledge, to date there is no evidence of the effect of PEA treatment in the clinical stages preceding the onset of psychosis as well as to prevent the risk of disease progression.

We propose a feasibility study of sustained ultramicronized-PEA (um-PEA) supplementation in CHR individuals to address the lack of a first-line treatment for this group of subjects, focusing on study recruitment and patient acceptability.

2. Materials and Methods

2.1. Trial design

This is a 12-week, open-label, investigator-initiated, proof-of-concept, single-arm study (Phase-2 Pilot Study) assessing the effect of um-PEA 600 mg/day in CHR individuals presenting with APS [1]. Within the first 12 months from the first participant recruitment, we will assess the feasibility of enrolling a minimum of 20 CHR participants and whether at least 80% of participants have completed the 12-week follow-up. After completing the initial 12-week phase, participants will be proposed to enter a 24-week extension phase of the study to evaluate the clinical stability of the treatment. Based on clinical judgement of the improvement obtained at that point, um-PEA might be titrated to 1200 mg/day. Each participant will undergo um-PEA treatment for a maximum of 36 weeks (Table 1). Our purpose is to examine: (i) um-PEA ability to alleviate APS and anxiety symptoms in CHR patients; (ii) um-PEA safety and tolerability; (iii) the biological basis of um-PEA effect. The study was approved by the Department of Medicine (DMED) at University of Udine (Institutional Review Board: 93/2023) in June 2023 (https://osf.io/wn2kf).

Table 1. Study flowchart.

	Screening phase	**Feasibility phase**			**Extension phase**	
Procedures	**Screening (Day -7)**	**Baseline (Day 0)**	**T1 (4 weeks ± 7 days)**	**T2 (12 weeks ± 14 days)**	**T3 (24 weeks ± 14 days)**	**T4 (36 weeks ± 14 days)**
Informed consent	X					
Medical history	X					
Physical examination	X	X		X		X
Electrocardiogram	X	X		X		X
Urinalysis	X			X		X

AUDIT [37], FTND [38], SDS [39], CEQ [40]	X					
TFLB [41]		X	X	X	X	
Pregnancy test	X	X	X	X	X	
Hematology, biochemistry	X			X		X
Concomitant medication	X	X	X	X	X	X
Exclusion criteria	X	X				
Intelligence quotient	X					
UKU-SERS [42]		X	X	X	X	X
Um-PEA dispensing		X	X	X	X	
Compliance assessment			X	X	X	X
CAARMS [1]	X	X	X	X	X	X
HADS [43], GF Social/Role scales [44]		X	X	X	X	X
Microbiome, inflammation, eCBome blood and stool assessments		X	X	X	X	X

AUDIT, Alcohol Use Disorders Identification Test; FTND, The Fagerstorm Test for Nicotine Dependence; SDS, Severity of Dependence Scale; CEQ, Cannabis Experience Questionnaire; TFLB, The Timeline Followback; UKU-SERS, UKU Side Effect Rating Scale; Um-PEA, Ultramicronized-Palmitoylethanolamide; CAARMS, The Comprehensive Assessment of At-Risk Mental State; HADS, Hospital Anxiety and Depression Scale; GF, Global Functioning.

2.2. Sample size calculation

The primary analysis (see below) will focus on total severity of APS as evaluated with the Comprehensive Assessment of At-Risk Mental State (CAARMS) [1], measured in three longitudinal assessments (i.e., Feasibility phase: Baseline, T1, and T2). Accepting a type-I error (α) of 5% and a type-II error (β) of 20%, we would need a sample of 8-12 participants to detect an effect of large size (partial-$\eta 2 \geq 0.140$). This value was obtained considering a moderate-to-large test-retest reliability of measurements over time ($+0.500 < r \leq +0.700$) and the absence of correction for sphericity. With an intermediate sphericity correction ($\varepsilon = 0.750$), the required sample size would be 10-14; in the worst case ($\varepsilon = 0.500$), instead, 12-19 participants would be needed. We therefore propose the recruitment of 20 participants, considered coherent with a pilot study and sufficient to evaluate feasibility outcomes of clinically significant size, also considering a low drop-out rate (i.e., ≤20%). When large statistically significant results are obtained on total severity of APS, specific APS severities will be further investigated (i.e., considering four P1 scales). For this secondary endpoint, on average, a sample-size

of 17 participants would be sufficient to detect large effects, also considering a Bonferroni's correction for multiple independent comparisons (i.e., α=0.012). Power analyses were carried out with G*Power-3.1.9.7 [45].

2.3. Objectives

2.3.1. Feasibility endpoints

We will record (i) the number of subjects giving consent for participating to the trial; (ii) the proportion of participants completing the initial 12-week follow-up. Patients will be defined as compliant in the presence of a pill count greater than 50% the expected number taken. Patients who are defined as non-compliant with the medication will be coded as protocol deviators.

2.3.2. Research endpoints

We will primarily address whether um-PEA added to treatment as usual (TAU) in CHR patients improves APS. Our secondary research questions are whether um-PEA added to TAU in CHR patients improves APS to the extent that patients no longer satisfy the diagnostic criteria for the CHR state, relieves APS-associated distress, improves CHR-related anxiety symptoms, and ameliorates social and role functioning, as well as general psychopathology and wellbeing measures. Therefore, our primary endpoint will be the assessment of APS severity as measured using the CAARMS [1]. As per the secondary endpoints: (i) APS-associated distress will be measured using the CAARMS [1]; (ii) severity of CHR-related anxiety symptoms will be measured using the Hospital Anxiety and Depression Scale (HADS) [43]; (iii) the level of impaired global functioning will be measured using the Social and Role functioning Scale [44]; (iv) clinical remission, defined as no longer meeting the APS criteria [1] will be measured using the CAARMS [1]; (v) general psychopathology and wellbeing changes will be measured using the CAARMS [1].

All the study endpoints for the 12-week clinical trial will be assessed by comparing follow-up (FUP) visits and baseline. For those participants continuing in the 24-week extension phase, change (FUP visit minus baseline) in the severity of psychotic symptoms as measured using the CAARMS will be compared with the group of those willing to discontinue um-PEA and continue with TAU. Should the psychopathological condition require TAU adjustments based on clinical judgement, this will be mentioned in a specific section of the trial case report form (CRF; https://osf.io/2vayh, accessed on 5 November 2024).

2.3.3. Safety endpoints

We will evaluate if sustained um-PEA treatment is well tolerated, with minimal side-effects. Incidence of adverse effects during the study period will be measured using the UKU side-effect rating scale [42].

2.4. Biological measures

Blood tests for haematology (i.e., full blood count and haemoglobin), biochemistry (i.e., urea and electrolytes, liver function test, lipid profile), sex hormones quantification [i.e., follicle-stimulating hormone (FSH), luteinizing hormone (LH)], and hypothalamic-pituitary-adrenal axis activity-related indices quantification (i.e., morning cortisol at 8 a.m.) will be carried out at the clinical research recruitment hub as per their standard procedure. Cytokines and chemokines will be analyzed on available serum samples using customized multiplex immunoenzymatic assays (Ella instrument, Bio-Techne, USA) to characterize the inflammatory profile. Serum samples will be collected for performing "metabolic fingerprinting" analysis using Surface-Enhanced Raman Scattering (SERS) spectroscopy, a rapid, label-free, and non-destructive method revealing molecular information about the biochemical composition of biofluids by analysing shifts in SERS spectral profiles [46-48]. Additionally, blood and faecal samples will be collected for the measurement of eCB mediators using liquid chromatography-mass spectrometry (LC/MS) techniques [49, 50], and the determination of the microbial composition in faeces using Next Generation Sequencing (NGS) of 16SRNA and shotgun metagenomics methodologies [51].

2.5. Study setting and participants

2.5.1. Screening and recruitment

The study will take place at the Unit of Psychiatry and Eating Disorders of the Udine University Hospital, a university clinical research facility in Italy. Help-seeking individuals presenting at the Unit of Psychiatry and Eating Disorders upon referral from General Practitioners (GPs) or other mental healthcare professionals from Udine catchment area and identified as CHR-APS patients by the clinical team will be considered for enrolment against inclusion and exclusion criteria. They will be involved into an internal pilot, that will progress to the open-label trial. Patients expressing an interest in participating to the study will be approached by study researchers and given a patient information sheet and consent form (https://osf.io/nrc5b, accessed on 5 November 2024).

Those who agree to take part in the study will be invited for a screening visit.

2.5.2. Inclusion criteria

The following are the participants' inclusion criteria:

- Individuals diagnosed with CHR-APS, as defined using CAARMS criteria [1];
- Aged 18-35 years;
- To be able to understand and communicate in Italian;
- To be able to give informed consent.

2.5.3. Exclusion criteria

- The following are the participants' exclusion criteria:
- Lifetime history of a psychotic or manic episode lasting 7 days or longer;
- Active suicidal ideation indicating significant current risk or history of serious suicide attempt in the opinion of the Principal Investigator (PI), as evaluated at the screening stage;
- Lifetime neurological disorders (e.g., epilepsy, except febrile convulsions) or severe intercurrent physical illness;
- Current treatment with psychotropic medication, with the exception of Selective Serotonin Reuptake Inhibitor (SSRI) stable monotherapy (at least 8 months);
- Lifetime treatment with antipsychotic medication for more than 7 days;
- Intelligence quotient (IQ) < 70;
- Female patients who are pregnant, lactating or not using an acceptable effective form contraception if they are at risk of falling pregnant;
- Taking part in another pharmacological trial.

2.5.4. Demographics

At the time of the screening visit, socio-demographic and clinical characteristics of each participant will be recorded in the trial CRFs (https://osf.io/2vayh, accessed on 5 November, 2024), including (i) age, (ii) sex, (iii) height, (iv) weight, (v) body mass index (BMI), (vi) reason for first referral, (vii) housing situation, (viii) marital status, (ix) nationality, (x) level of education, (xi) working situation, (xii) handedness, (xiii) prior contact with child & adolescent mental health service (CAMHS), (xiv) prior contact with adult mental health service (AMHS), (xv) predating hospitalizations for mental health issues, (xvi) predating contact with social services, (xvii) predating diagnosis, (xviii) age at predating diagnosis, (xix) comorbid neurodevelopmental disorders (NDDs), (xx) comorbid physical illnesses, (xxi) impairment in daily autonomies, (xxii) intellectual disability, (xxiii) brain magnetic resonance imaging (MRI), (xxiv) childhood adversities (CAs; i.e., sexual/physical/psychological/ emotional abuse, physical/emotional neglect, exploitation, peer victimization/bullyism), (xxv) any CAs, (xxvi) recent stressful life events (SLEs), (xxvii) predating treatment with any psychotropic medications, (xxviii) type of predating medication, (xxix) current treatment with any psychiatric

medications, (xxx) type of current medication, (xxxi) lifetime addicting substance use (any), (xxxii) current addicting substance use (any), (xxxiii) lifetime cannabis use, (xxxiv) current cannabis use, (xxxv) lifetime alcohol use, (xxxvi) current alcohol use, (xxxvii) lifetime stimulant use, (xxxviii) current stimulant use, (xxxix) NDDs family history, (xl) major mental health disorders (MMHDs) family history, (xli) substance use disorders (SUDs) family history, (xlii) suicide/suicide attempt family history, (xliii) neurological disorders family history.

Substance use information will be collected as screening measures, using dedicated interviews [37-40].

2.6. Intervention

Participants will initially undertake a 12-week daily treatment with oral um-PEA 600 mg in tablet form (Normast®). During the 24-week extension phase of the study, the trial medication will be taken from once a day up to twice a day (600-1200 mg per day), based on clinical judgment of the improvement obtained at that point, around mealtime. Being a food supplement/nutraceutical, um-PEA can be purchased at pharmacies without a medical prescription [52]. While unknown risks cannot be excluded, serious adverse events including overdose have not been documented [53, 54]. Um-PEA will be obtained by a pharmaceutical company operating under good manufacturing practice conditions with appropriate certification.

The information presented on the labels for um-PEA will comply with applicable national and local regulations. Only qualified physicians clearly given this role by the PI will prescribe the study medication on the study delegation log. Only people designated by the PI will collect medication. Only um-PEA supplied for this study will be dispensed against the study specific prescription. Full accountability records will be completed including recording the batch, expiry date, people dispensing/checking the prescription, quantity and date of drug returns, empty packaging. Nothing will be destroyed without the authorization from the PI.

Um-PEA will be stored at room temperature (< 25 °C) and not kept in a refrigerator, in compliance with local regulations. It will be stored in a secure area away from other treatments and clearly marked for this study.

2.7. Data management and confidentiality

Study-related information will be stored securely at the study site. All participants' information will be stored in locked file cabinets in areas with limited access. All laboratory specimens, reports, data collection, process and administrative forms, will be identified by a coded identification (ID) number only to maintain participant confidentiality and stored separately from records containing names or

other personal identifiers. Local databases will be secured with password-protected access systems. Forms, lists, logbooks, appointment books, and any other listings that link participant ID numbers to other identifying information will be stored in a separate, locked file in an area with limited access. Participants' study information will not be released outside of the study without the written permission of the participant.

2.8. Statistical methods

2.8.1. Data verification, Statistical monitoring, and Analysis

At the study start, a structured data verification plan will be agreed and developed by the Head Statistician (M.G.) of the research team. Quality of data will be routinely checked in terms of consistency across paper- and electronic-based entry systems. Statistical monitoring will include patient severity level, eventual withdrawals, baseline data, FUP visits data, and adverse effects. The research team will approve a Statistical Analysis Plan within the early stages of the study, and before the Head Statistician summarizes any data (i.e., with univariate and repeated measures analysis, including preliminary assessment and management of any outliers and assessment of the metric qualities of measurements, such as normality and sphericity, where necessary). Proportions will be presented for each feasibility outcome. Interim analyses are not planned. The Head Statistician will both carry out and interpret statistical analysis. Our primary analysis will involve a generalized linear model with CAARMS psychotic symptom severity as the dependent measure and time as a repeated measure (possibly supplemented with analyses at the participant level, as random effects in mixed models). We are mainly interested in whether there is a significant benefit over time with um-PEA treatment: therefore, particular interest will be placed on time as a fixed factor on the severity of outcomes. CAARMS distress and total score as well as HADS and GF ratings will be analysed in a similar manner. We will also conduct paired tests (both parametric and non-parametric) based on baseline and endpoint assessments to examine measures of CAARMS severity, CAARMS frequency/duration, and secondary outcomes. Trend and rates of adverse effects will be reported, as well as reasons for dropouts from the trial.

2.8.2. Missing data

We expect withdrawn patients to be missing at random, and very few participants withdrawing consent. All patients having at least one post-baseline outcome measure will be included in each analysis. Nonetheless, patterns of missing data will be explored to investigate any evidence against missing at random, whose impact on the analysis will be considered by introducing imputation methods (i.e., Multiple Imputation or MissForest approach, depending on the missing data type).

2.9. Withdrawal of participants

In accordance with the Declaration of Helsinki [55], all participants have the right to withdraw from the study at any time, without the need to provide a reason, and without affecting their future medical care. Participants will be informed of this right prior to giving consent. If a participant decides to withdraw, it will be clarified that they are only discontinuing the study treatment while continuing follow-up visits. Should a participant wish to fully withdraw from the study, their decision will be respected. Efforts will be made to determine the reason for withdrawal, although participants are not required to provide any explanation. Data already collected will be retained and included in the final analysis. The PI may also withdraw participants for various reasons, including but not limited to protocol violations, intercurrent illness, adverse events, serious adverse events, suspected unexpected serious adverse reactions, administrative reasons, or if participation in the trial interferes with ongoing care or results in symptomatic worsening.

In these cases, participants will continue to be followed up with the same schedule of research assessments as those who remain in the study, until they complete the 12-week follow-up period or until they develop frank psychosis, whichever occurs first. Participants who experience progression to first-episode psychosis will exit the study intervention, be classified as treatment failure, and only be assessed for safety outcomes until completing the 12-week follow-up period.

3. Discussion

Despite substantial advancements in early detection and prognosis, current research still converges towards the lack of effective interventions for transition to psychosis, subtle psychotic symptoms, functional status, and depression, among CHR individuals [3, 5, 56]. Noteworthy, APS, negative symptoms, and impairment in social and role functioning affect the quality of life of CHR individuals similarly to those of psychosis patients [57]. The present proof-of-concept Phase-2 study proposes the use of a nutraceutical, the fatty acid amide PEA in its ultramicronized form, to treat APS and reduce psychic distress in CHR individuals. In parallel, um-PEA modulation of potential CHR state biomarkers across the inflammatory, endocannabinoid, and microbiome systems will be assessed.

This study protocol should be seen considering some strengths and limitations. First, our study provides a reasonably extended treatment for help-seeking young adults that may not benefit from psychotherapy techniques and are often concerned about the possible unpleasant side-effects of psychotropic medications. Also, the use of conventional anti-psychotics before formulating the diagnosis of a clear-cut psychotic disorder raises concerns about the potential risk of accelerating disease progression [58], as well as implications from forensic and ethical standpoints [59, 60].

Second, the trial will be pursued with minimal risks for study participants, being PEA a well-established Food for Special Medical Purposes (FSMP) for prolonged therapy in several clinical conditions [52], nearly devoid of known or potential adverse effects [61]. Oral PEA has shown well-documented safety and tolerability in numerous clinical trials with over 1000 individuals with doses 300-1200 mg per day both in healthy and sick populations [53, 54], with negligible acute and repeat dose toxicity [62]. Also, no invasive techniques will be implemented throughout the study.

Third, the feasibility of this proof-of-concept trial will justify its continuation as a larger Phase-3 study investigating PEA efficacy, tolerability, and acceptance in CHR subjects.

To this extent, randomization and blinding of future studies will be needed to help reduce possible biases. Also, being a single-site study, selected CHR individuals may differ to some extent from the population treated in normal life, making it difficult to interpret and generalize the results. Indeed, in line with the evidence that around 50% mental health disturbances start by late adolescence [4], when patients are referred to transition-age youth psychiatry services [63], future studies would probably benefit from the inclusion of underage participants (e.g., aged 15-18 years). Moreover, although CHR youths with APS are those most referred to mental health outpatient services [64], future studies should also consider including patients from the CHR-vulnerability group and the CHR-brief limited intermittent psychotic symptoms (BLIPS) group [1]. This approach could help capture potential variations in clinical responses across the CHR spectrum. Finally, more stringent systems for monitoring patient compliance with therapy (e.g., medication diaries, text messaging [65]) would enable more rigorous tracking of medication adherence by participants in future trials.

Net of the above considerations, an open-label single-arm design appears suitable for an early-phase trial in this specific clinical population [66] and may represent the initial ground to build a greater understanding of the possible correlations between behavior and biomarkers modulation as a response to PEA supplementation in CHR individuals.

References

1. Yung, A.R., et al., *Mapping the onset of psychosis: the Comprehensive Assessment of At-Risk Mental States.* Aust N Z J Psychiatry, 2005. **39**(11-12): p. 964-71.
2. Salazar de Pablo, G., et al., *Probability of Transition to Psychosis in Individuals at Clinical High Risk: An Updated Meta-analysis.* JAMA Psychiatry, 2021. **78**(9): p. 970-978.
3. Salazar de Pablo, G., et al., *Transition to psychosis in randomized clinical trials of individuals at clinical high risk of psychosis compared to observational cohorts: a systematic review and meta-analysis.* Eur Psychiatry, 2021. **64**(1): p. e51.
4. Colizzi, M., A. Lasalvia, and M. Ruggeri, *Prevention and early intervention in youth mental health: is it time for a multidisciplinary and trans-diagnostic model for care?* Int J Ment Health Syst, 2020. **14**: p. 23.
5. Fusar-Poli, P., et al., *Prevention of Psychosis: Advances in Detection, Prognosis, and Intervention.* JAMA Psychiatry, 2020. **77**(7): p. 755-765.
6. McGorry, P.D. and C. Mei, *Ultra-high-risk paradigm: lessons learnt and new directions.* Evid Based Ment Health, 2018. **21**(4): p. 131-133.
7. Davies, C., et al., *Lack of evidence to favor specific preventive interventions in psychosis: a network meta-analysis.* World Psychiatry, 2018. **17**(2): p. 196-209.
8. Davies, C., et al., *Efficacy and Acceptability of Interventions for Attenuated Positive Psychotic Symptoms in Individuals at Clinical High Risk of Psychosis: A Network Meta-Analysis.* Front Psychiatry, 2018. **9**: p. 187.
9. Devoe, D.J., A. Peterson, and J. Addington, *Negative Symptom Interventions in Youth at Risk of Psychosis: A Systematic Review and Network Meta-analysis.* Schizophr Bull, 2018. **44**(4): p. 807-823.
10. Devoe, D.J., et al., *Interventions and social functioning in youth at risk of psychosis: A systematic review and meta-analysis.* Early Interv Psychiatry, 2019. **13**(2): p. 169-180.
11. Appiah-Kusi, E., et al., *Abnormalities in neuroendocrine stress response in psychosis: the role of endocannabinoids.* Psychol Med, 2016. **46**(1): p. 27-45.
12. Giuffrida, A., et al., *Cerebrospinal anandamide levels are elevated in acute schizophrenia and are inversely correlated with psychotic symptoms.* Neuropsychopharmacology, 2004. **29**(11): p. 2108-14.
13. Koethe, D., C. Hoyer, and F.M. Leweke, *The endocannabinoid system as a target for modelling psychosis.* Psychopharmacology (Berl), 2009. **206**(4): p. 551-61.

14. Pertwee, R.G., *The diverse CB1 and CB2 receptor pharmacology of three plant cannabinoids: delta9-tetrahydrocannabinol, cannabidiol and delta9-tetrahydrocannabivarin.* Br J Pharmacol, 2008. **153**(2): p. 199-215.
15. McGuire, P., et al., *Cannabidiol (CBD) as an Adjunctive Therapy in Schizophrenia: A Multicenter Randomized Controlled Trial.* Am J Psychiatry, 2018. **175**(3): p. 225-231.
16. Filipiuc, L.E., et al., *Major Phytocannabinoids and Their Related Compounds: Should We Only Search for Drugs That Act on Cannabinoid Receptors?* Pharmaceutics, 2021. **13**(11).
17. Colizzi, M., et al., *Palmitoylethanolamide and Its Biobehavioral Correlates in Autism Spectrum Disorder: A Systematic Review of Human and Animal Evidence.* Nutrients, 2021. **13**(4).
18. Bortoletto, R., et al., *Is It Time to Test the Antiseizure Potential of Palmitoylethanolamide in Human Studies? A Systematic Review of Preclinical Evidence.* Brain Sciences, 2022. **12**(1).
19. Colizzi, M., et al., *Therapeutic effect of palmitoylethanolamide in cognitive decline: A systematic review and preliminary meta-analysis of preclinical and clinical evidence.* Frontiers in Psychiatry, 2022. **13**.
20. Petrosino, S., et al., *Palmitoylethanolamide counteracts substance P-induced mast cell activation in vitro by stimulating diacylglycerol lipase activity.* J Neuroinflammation, 2019. **16**(1): p. 274.
21. Skaper, S.D., L. Facci, and P. Giusti, *Glia and mast cells as targets for palmitoylethanolamide, an anti-inflammatory and neuroprotective lipid mediator.* Mol Neurobiol, 2013. **48**(2): p. 340-52.
22. Skaper, S.D. and L. Facci, *Mast cell-glia axis in neuroinflammation and therapeutic potential of the anandamide congener palmitoylethanolamide.* Philos Trans R Soc Lond B Biol Sci, 2012. **367**(1607): p. 3312-25.
23. Petrosino, S., et al., *The anti-inflammatory mediator palmitoylethanolamide enhances the levels of 2-arachidonoyl-glycerol and potentiates its actions at TRPV1 cation channels.* Br J Pharmacol, 2016. **173**(7): p. 1154-62.
24. Couch, D.G., et al., *Palmitoylethanolamide and Cannabidiol Prevent Inflammation-induced Hyperpermeability of the Human Gut In Vitro and In Vivo-A Randomized, Placebo-controlled, Double-blind Controlled Trial.* Inflamm Bowel Dis, 2019. **25**(6): p. 1006-1018.
25. Bortoletto, R., et al., *Questioning the role of palmitoylethanolamide in psychosis: a systematic review of clinical and preclinical evidence.* Front Psychiatry, 2023. **14**: p. 1231710.
26. Appiah-Kusi, E., et al., *Childhood trauma and being at-risk for psychosis are associated with higher peripheral endocannabinoids.* Psychol Med, 2020. **50**(11): p. 1862-1871.

27. Leweke, F.M., et al., *Cannabidiol enhances anandamide signaling and alleviates psychotic symptoms of schizophrenia.* Transl Psychiatry, 2012. **2**(3): p. e94.
28. Koethe, D., et al., *Familial abnormalities of endocannabinoid signaling in schizophrenia.* World Journal of Biological Psychiatry, 2019. **20**(2): p. 117-125.
29. Ibarra-Lecue, I., et al., *Cannabis use selectively modulates circulating biomarkers in the blood of schizophrenia patients.* Addiction Biology, 2022. **27**(6).
30. Parksepp, M., et al., *The Expanded Endocannabinoid System Contributes to Metabolic and Body Mass Shifts in First-Episode Schizophrenia: A 5-Year Follow-Up Study.* Biomedicines, 2022. **10**(2).
31. Topuz, R.D., Y. Gorgulu, and M.K. Uluturk, *Could serum endocannabinoid and N-acylethanolamine levels be important in bipolar disorder?* World Journal of Biological Psychiatry: p. 7.
32. Leweke, F.M., et al., *Elevated endogenous cannabinoids in schizophrenia.* NeuroReport, 1999. **10**(8): p. 1665-1669.
33. Muguruza, C., et al., *Quantification of endocannabinoids in postmortem brain of schizophrenic subjects.* Schizophrenia Research, 2013. **148**(1-3): p. 145-150.
34. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.
35. Abedini, T., et al., *Efficacy and safety of palmitoylethanolamide as an adjunctive treatment for acute mania: A randomized, double-blind, placebo-controlled trial.* Psychiatry Clin Neurosci, 2022. **76**(10): p. 505-511.
36. Brotini, S., C. Schievano, and L. Guidi, *Ultra-micronized Palmitoylethanolamide: An Efficacious Adjuvant Therapy for Parkinson's Disease.* CNS Neurol Disord Drug Targets, 2017. **16**(6): p. 705-713.
37. Saunders, J.B., et al., *Development of the Alcohol Use Disorders Identification Test (AUDIT): WHO Collaborative Project on Early Detection of Persons with Harmful Alcohol Consumption--II.* Addiction, 1993. **88**(6): p. 791-804.
38. Fagerstrom, K., et al., *The Fagerstrom Test for Nicotine Dependence as a predictor of smoking abstinence: a pooled analysis of varenicline clinical trial data.* Nicotine Tob Res, 2012. **14**(12): p. 1467-73.
39. Gossop, M., et al., *The Severity of Dependence Scale (SDS): psychometric properties of the SDS in English and Australian samples of heroin, cocaine and amphetamine users.* Addiction, 1995. **90**(5): p. 607-14.

40. Birnbaum, M.L., et al., *Factor structure of the Cannabis Experiences Questionnaire in a first-episode psychosis sample.* Early Interv Psychiatry, 2019. **13**(3): p. 495-501.
41. Sobell, L.C., J.A. Cunningham, and M.B. Sobell, *Recovery from alcohol problems with and without treatment: prevalence in two population surveys.* Am J Public Health, 1996. **86**(7): p. 966-72.
42. Lingjaerde, O., et al., *The UKU side effect rating scale. A new comprehensive rating scale for psychotropic drugs and a cross-sectional study of side effects in neuroleptic-treated patients.* Acta Psychiatr Scand Suppl, 1987. **334**: p. 1-100.
43. Snaith, R.P. and A.S. Zigmond, *The hospital anxiety and depression scale.* Br Med J (Clin Res Ed), 1986. **292**(6516): p. 344.
44. Cornblatt, B.A., et al., *Preliminary findings for two new measures of social and role functioning in the prodromal phase of schizophrenia.* Schizophr Bull, 2007. **33**(3): p. 688-702.
45. Faul, F., et al., *G*Power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences.* Behavior Research Methods, 2007. **39**(2): p. 175-191.
46. Moisoiu, V., et al., *SERS liquid biopsy: An emerging tool for medical diagnosis.* Colloids Surf B Biointerfaces, 2021. **208**: p. 112064.
47. Premasiri, W.R., J.C. Lee, and L.D. Ziegler, *Surface-enhanced Raman scattering of whole human blood, blood plasma, and red blood cells: cellular processes and bioanalytical sensing.* J Phys Chem B, 2012. **116**(31): p. 9376-86.
48. Fornasaro, S., V. Sergo, and A. Bonifacio, *The key role of ergothioneine in label-free surface-enhanced Raman scattering spectra of biofluids: a retrospective re-assessment of the literature.* FEBS Lett, 2022. **596**(10): p. 1348-1355.
49. Ferrara, A.L., et al., *Altered Metabolism of Phospholipases, Diacylglycerols, Endocannabinoids, and N-Acylethanolamines in Patients with Mastocytosis.* J Immunol Res, 2019. **2019**: p. 5836476.
50. Maia, J., et al., *The endocannabinoidome in human placenta: Possible contribution to the pathogenesis of preeclampsia.* Biofactors, 2023. **49**(4): p. 887-899.
51. Minichino, A., et al., *Endocannabinoid system mediates the association between gut-microbial diversity and anhedonia/amotivation in a general population cohort.* Mol Psychiatry, 2021. **26**(11): p. 6269-6276.
52. Clayton, P., et al., *Palmitoylethanolamide: A Natural Compound for Health Management.* Int J Mol Sci, 2021. **22**(10).

53. Chirchiglia, D., et al., *Effects of Add-On Ultramicronized N-Palmitol Ethanol Amide in Patients Suffering of Migraine With Aura: A Pilot Study.* Front Neurol, 2018. **9**: p. 674.
54. Marini, I., et al., *Palmitoylethanolamide versus a nonsteroidal anti-inflammatory drug in the treatment of temporomandibular joint inflammatory pain.* J Orofac Pain, 2012. **26**(2): p. 99-104.
55. Shrestha, B. and L. Dunn, *The Declaration of Helsinki on Medical Research involving Human Subjects: A Review of Seventh Revision.* J Nepal Health Res Counc, 2020. **17**(4): p. 548-552.
56. Catalan, A., et al., *Annual Research Review: Prevention of psychosis in adolescents - systematic review and meta-analysis of advances in detection, prognosis and intervention.* J Child Psychol Psychiatry, 2021. **62**(5): p. 657-673.
57. Fusar-Poli, P., et al., *Disorder, not just state of risk: meta-analysis of functioning and quality of life in people at high risk of psychosis.* Br J Psychiatry, 2015. **207**(3): p. 198-206.
58. Raballo, A., M. Poletti, and A. Preti, *The temporal dynamics of transition to psychosis in individuals at clinical high-risk (CHR-P) shows negative prognostic effects of baseline antipsychotic exposure: a meta-analysis.* Transl Psychiatry, 2023. **13**(1): p. 112.
59. NICE. *Psychosis and Schizophrenia in Adults: Prevention and Management (CG178)*. 2014; Available from: www.nice.org.uk/guidance/cg178.
60. NICE. *Psychosis and Schizophrenia in Children and Young People: Recognition and Management (CG155)*. 2016; Available from: www.nice.org.uk/guidance/cg155.
61. Gabrielsson, L., S. Mattsson, and C.J. Fowler, *Palmitoylethanolamide for the treatment of pain: pharmacokinetics, safety and efficacy.* Br J Clin Pharmacol, 2016. **82**(4): p. 932-42.
62. Nestmann, E.R., *Safety of micronized palmitoylethanolamide (microPEA): lack of toxicity and genotoxic potential.* Food Sci Nutr, 2017. **5**(2): p. 292-309.
63. Colizzi, M., et al., *Lessons Learnt From Running a Transition-Age Youth Mental Health Outpatient Clinic in Italy: The PRecocity of Intervention in Adolescent Medicine (PRIMA) Experience.* Early Interv Psychiatry, 2024.
64. Fusar-Poli, P., et al., *Diagnostic and Prognostic Significance of DSM-5 Attenuated Psychosis Syndrome in Services for Individuals at Ultra High Risk for Psychosis.* Schizophr Bull, 2018. **44**(2): p. 264-275.
65. Bobrow, K., et al., *Efficacy of a text messaging (SMS) based intervention for adults with hypertension: protocol for the StAR (SMS Text-message Adherence suppoRt trial) randomised controlled trial.* BMC Public Health, 2014. **14**: p. 28.
66. Jones, N.E., *Pathways to Treatment Development*, in *Encyclopedia of Behavioral Neuroscience, 2nd edition (Second Edition)*, S. Della Sala, Editor. 2022, Elsevier. p. 226-239.

Acknowledgement: The authors would like to acknowledge infrastructure from the University of Udine and the Friuli Centrale Health University Authority (ASUFC). The authors would like to acknowledge Epitech Group for supporting their research activity through the generous supply of the study medication.

Funding: This work received no external funding.

CRediT authorship contribution statement:

RB: Conceptualization, Methodology, Writing-original draft, Writing – review and editing

MG: Conceptualization, Methodology, Formal analysis, Writing-original draft, Writing – review and editing

FP: Conceptualization, Methodology, Writing – review and editing

SF: Conceptualization, Methodology, Writing – review and editing

CS: Conceptualization, Methodology, Writing – review and editing

OS: Conceptualization, Methodology, Writing-original draft, Writing – review and editing

MF: Conceptualization, Methodology, Writing – review and editing

FC: Conceptualization, Methodology, Writing – review and editing

MB: Conceptualization, Methodology, Writing – review and editing, Project administration

MC: Conceptualization, Methodology, Writing-original draft, Writing – review and editing, Supervision, Project administration

CHAPTER V

Effects of Palmitoylethanolamide in Clinical High-Risk for Psychosis: A Nonrandomized Open-Label Trial

Bortoletto R, Garzitto M, Basaldella M, Scipioni C, Sepulcri O, Fabris M, Curcio F, Balestrieri M, Colizzi M. Effects of palmitoylethanolamide in clinical high-risk for psychosis: A nonrandomized open-label trial. Brain Behav Immun Health. 2025 Nov 22;50:101141. doi: 10.1016/j.bbih.2025.101141. PMID: 41378189; PMCID: PMC12689190.

Note: Supplementary material associated with this publication is provided in the Appendix section of this thesis.

Abstract

Clinical high-risk (CHR) for psychosis state still lacks effective and safe treatments. Recent evidence supports the anti-neuroinflammatory properties of fatty acid palmitoylethanolamide (PEA) dietary supplementation across the psychosis spectrum. Sixteen subjects at CHR for psychosis with attenuated psychotic symptoms (APS) enrolled in a 12-week, open-label, nonrandomized, single-arm clinical trial of ultramicronized-PEA (um-PEA, 600 mg/day). Biobehavioral assessments were conducted at baseline, 4 weeks, and 12 weeks, particularly using the Comprehensive Assessment of At-Risk Mental States (CAARMS) and quantifying changes in peripheral neuroimmune biomarkers. Linear mixed-effects models showed significant reductions in CAARMS total APS ($\Delta_{12\text{-weeks}}$=-3.8 units, -30.0%) and Unusual Thought Content (UTC; $\Delta_{12\text{-weeks}}$=-2.0, -52.5%). No treatment-emergent side effects were reported. In exploratory analyses including neuroimmune biomarkers as covariates and potential moderators, lower baseline Interleukin (IL)-1β was associated with greater improvement in UTC, while time-varying IL-10/IL-6 ratio, Interferon (IFN)-γ, and IL-6 were linked to changes in CAARMS total APS and UTC.

A reduction in APS was observed in subjects at CHR for psychosis receiving PEA supplementation, possibly through immune-inflammatory modulation, warranting further research into its therapeutic potential for this condition.

Trial Registration: ClinicalTrials.gov Identifier NCT06037993.

Keywords

Schizophrenia; At-risk mental state; Acylethanolamines; Cannabinoids; Glutamate; Neuroinflammation

1. Introduction

Interventions for individuals at clinical high-risk (CHR) state for psychosis [1] remain an unmet clinical need among young people presenting to mental health services. Most first episodes of psychotic disorders are preceded not only by the core attenuated or brief psychotic symptoms characteristic of the CHR state—which themselves are associated with severe functional impairment affecting the lives of young people and their families—but also by co-occurring or subsequent conditions including depression, anxiety, personality disorder, and substance misuse. Early detection and intervention strategies therefore aim to reduce both the risk of transition to first-episode psychosis (FEP) onset and the duration of untreated illness (DUI), while also relieving such a burden of distressing symptoms. However, to date, no specific treatment has proven clear therapeutic superiority [2, 3]. Compelling research underlines the pathophysiological relevance of altered neuroimmune biomarkers (e.g., cytokines, stress-related hormones) across the psychosis spectrum, including the CHR state [4-9] and progression to FEP [6, 10-12], identifying this system as a potential target for novel therapies. Emerging evidence suggests that endogenous levels of the fatty acid amide palmitoylethanolamide (PEA) are elevated in CHR and early psychosis but tend to decline as the illness progresses [13-18]. This trajectory parallels findings supporting the benefits of PEA dietary supplementation in improving negative symptoms in schizophrenia, possibly through its anti-neuroinflammatory and neuroprotective properties [13, 19, 20]. Given the small proportion of CHR subjects eventually transitioning to psychosis [21], the need for novel medications to be the safest is essential. In this regard, recent studies highlight the therapeutic potential of PEA's exogenous counterpart, cannabidiol, for CHR, showing promising results in a population still lacking effective and reasonably tolerable treatments [2, 22].

However, despite its safer profile and shorter biological distance from the therapeutic target, the effect of PEA supplementation in CHR remains unexplored.

This study aims to fill this gap through the following objectives.

1.1.Objectives

To assess trajectories of CHR symptoms and peripheral neuroimmune biomarkers during PEA supplementation.

To assess associations between baseline levels of peripheral neuroimmune biomarkers and subsequent symptom trajectories during PEA supplementation.

To assess associations between changes in peripheral neuroimmune biomarkers and longitudinal trajectories of CHR symptoms during PEA supplementation.

2. Material and Methods

2.1. Participants

People at CHR state for psychosis with attenuated psychotic symptoms (APS) [1], presenting at the Unit of Psychiatry and Eating Disorders of the Udine University Hospital, were recruited as part of a proof-of-concept study on the effects of PEA supplementation over a two-year period (November 2022 – November 2024).

Eligible subjects met the ultra-high-risk (UHR)-APS criteria measured with the Comprehensive Assessment of At-Risk Mental States (CAARMS) [1], were aged 18-35 years, fluent in Italian, and capable of providing written informed consent (https://osf.io/nrc5b, accessed on 15 June 2025). Given the clinical and biological heterogeneity within the CHR population, the sample was restricted to those with APS, who typically present with more severe symptomatology and are most frequently referred to mental health services [23]. Exclusion criteria comprised a prior diagnosis of a psychotic disorder or manic episode according to the Diagnostic and Statistical Manual of Mental Disorders – Fifth Edition, text revision (DSM-5-TR) [24], lifetime treatment with antipsychotic medications, active suicidal ideation, history of neurological disorders or severe intercurrent physical illness, intelligence quotient (IQ) < 70, and pregnancy or breastfeeding.

2.2. Study design and clinical assessments

Eligible participants enrolled in a 12-week, open-label, single-arm, proof-of-concept trial with PEA in its ultramicronized form (um-PEA, 600 mg/day) as an add-on to treatment as usual (TAU). As um-PEA is a dietary supplement, it can be purchased in pharmacies without a medical prescription. For the trial, um-PEA was supplied by a pharmaceutical company operating under Good Manufacturing Practice (GMP) conditions and with appropriate certification. The study medication (Normast®, um-PEA 600 mg tablet) was administered once daily at home, during breakfast. The choice of a relatively low dosage (600 mg/day) was based on the mechanistic objectives of the trial, given the well-documented tolerability and safety of um-PEA in both clinical populations and healthy individuals across the 300–1200 mg range. Adherence was monitored by pill count, conducted by the experimenters (R.B., M.Bas.) at each follow-up visit. TAU included stable interventions [e.g., psychotherapy and/or selective serotonin reuptake inhibitors (SSRIs)] that had been ongoing for at least 8 months prior to enrollment, with persistent APS at baseline indicating suboptimal response.

Clinical assessments were conducted at baseline (BL) and repeated at 4 (FU-1) and 12 (FU-2) weeks, using the CAARMS [1], the Hospital Anxiety and Depression Scale (HADS) [25], the Global Functioning (GF): Social and Role scales [26], and the UKU Side-Effect Rating Scale [27]. The primary endpoint was the reduction in the intensity of total APS in CHR participants.

Full study protocol and methodology details are reported elsewhere [28].

2.3. Neuroimmune biomarkers

Blood/serum samples were collected at baseline and after 4 and 12 weeks of treatment to quantify changes in neuroimmune biomarkers, using standardized protocols and multiplex immunoenzymatic platforms [28]. Based on current evidence regarding the most reliable peripheral biomarkers for both CHR state and transition to psychosis, neuroimmune biomarkers were operationalized as peripheral indices of inflammation and hypothalamic-pituitary-adrenal (HPA) axis activity, including the following in final analyses: basophils, C-reactive protein (CRP), cortisol awakening response (CAR), eosinophils, erythrocyte sedimentation rate (ESR), interferon-γ (IFN-γ), IFN-γ-induced protein-10 (IP-10), interleukin-1β (IL-1β), IL-2-receptor-α (IL-2Rα), IL-6, IL-8, IL-10, IL-17a, monocyte/lymphocyte ratio, neutrophil/lymphocyte ratio, platelet/lymphocyte ratio, systemic immune inflammation (SII) index, tumor necrosis factor (TNF-α), and total white blood cell count (WBC) [4-12]. In line with previous research in this field, we also calculated the IL-10/IL-6 ratio to account for potential imbalance between anti-inflammatory and pro-inflammatory cytokines [6]. In total, 20 neuroimmune biomarkers were included in the exploratory analyses outlined below.

2.4. Ethics considerations

The study was conducted in accordance with the Declaration of Helsinki [29] and approved by the Institutional Review Board (IRB) of the Department of Medicine (DMED) at the University of Udine (protocol number: 93/2023; Clinicaltrials.gov Identifier: NCT06037993, accessed on 15 June 2025). The trial is reported in accordance with the Transparent Reporting of Evaluations with Nonrandomized Designs (TREND) guidelines.

2.5. Statistical analyses

A-priori power analyses were conducted to determine the minimum detectable effect size for within-subject effects over time, given the available sample size. Effects of Cohen's $f \geq 0.403$ (roughly corresponding to Kendall $W \geq 0.120$ in corresponding non-parametric test) could be reliably detected (with $\alpha=0.05$ and $\beta=0.20$) across three timepoints, assuming moderate correlations between repeated measures ($0.5 < r \leq 0.7$) and sphericity ($\varepsilon \geq 0.7$).

Linear mixed-effects models were used to evaluate longitudinal changes in biobehavioral outcomes over 12 weeks of PEA supplementation, with assessment time point (baseline, 4 weeks, 12 weeks) as a fixed effect and participant as a random effect. The Friedman test was used as non-parametric method to verify results. In subsequent exploratory analyses, for statistically significant models with

adequate effect size, baseline neuroimmune biomarkers (dichotomized as at or above vs. below the sample median) were included as fixed effects to assess their interactive effects (Biomarker × Assessment) with assessment time on symptom trajectories. Finally, to preliminarily examine the dynamic neuroimmune influences on treatment response, changes in biomarker levels over time were considered as potential moderators of symptom trajectories (time-varying Biomarker × Assessment). In analyses for longitudinal changes, isolated single-assessment missing values were substituted with spline-interpolated values using adjacent time points. List-wise deletion was subsequently adopted, when incorporating further fixed factors or covariates in the models. Sensitivity analyses were performed on complete cases. All analyses were conducted in R 4.5.1 (https:// www.R-project.org).

3. Results

Overall, sixteen subjects at CHR for psychosis with APS were enrolled. Participants were help-seeking young adults (21.6 ±3.34 years), equally distributed between males and females, typically complaining of anxiety (87.5%) and depressive (62.5%) symptoms, but with limited previous contacts with mental health (25.0%), or social (12.5%) services. Only a minority reported impairments in daily functioning (18.8%) or underlying medical conditions (12.5%). Most participants came from urban areas (60.0%), had completed high school or higher education (56.3%), and were single (75.0%), unemployed (87.5%), and living with their families of origin (68.8%). Symptoms reported at the CAARMS are detailed in Table 1.

Fifteen subjects completed the 12-week protocol and were considered adherent to the treatment, defined as taking more than 50% of the prescribed intake. A single subject withdrew after one month due to difficulties in complying with the scheduled protocol visits (Figure 1).

Following PEA treatment, linear mixed effects models showed significant reductions in symptom severity for the CAARMS Total APS ($\Delta_{(FU-1 - BL)}$=-2.7, with 95% confidence interval in [-4.1, -1.3] range, -21.7% [-32.8%, -10.5%]; $\Delta_{(FU-2 - BL)}$=-3.8 [-5.2, -2.3], -30.0% [-41.7%, -18.4%]; $F_{2,30}$=14.0, p<0.001; Cohen's f=0.484; Supplementary Figure 1) and Unusual Thought Content (UTC; $\Delta_{(FU-1 - BL)}$=-1.2 [-2.0, -0.4], -32.1% [-53.0%, -11.3%]; $\Delta_{(FU-2 - BL)}$=-2.0 [-2.8, -1.2], -52.5% [-74.0%, -31.0%]; $F_{2,30}$=11.4, p<0.001; f= 0.554; Supplementary Figure 2). Statistically significant effects were also observed for Non-Bizarre Ideas (NBI; $\Delta_{(FU-1 - BL)}$=-0.9 [-1.5, -0.2], -22.3% [-40.4%, -4.2%]; $\Delta_{(FU-2 - BL)}$=-1.1 [-1.9, -0.4], -29.5% [-48.8%, -10.2%]; $F_{2,30}$=5.6, p=0.009; f=0.336), self-reported anxiety (HADS-A; $\Delta_{(FU-1 - BL)}$=-2.4 [-3.6, -1.2], -22.2% [-33.4%, -10.9%]; $\Delta_{(FU-2 - BL)}$=-2.8 [-4.3, -1.3], -26.3% [-40.2%, -12.4%]; $F_{2,30}$=10.2, p<0.001; f=0.380), self-reported depression (HADS-D; $\Delta_{(FU-1 - BL)}$=-1.8 [-3.5, -0.1], -22.3% [-43.8%, -0.8%]; $\Delta_{(FU-2 - BL)}$=-2.5 [-4.5, -0.4], -31.0% [-56.4%, -5.5%]; $F_{2,30}$=4.7, p=0.017; f=0.252), and social functioning (GF: Social; $\Delta_{(FU-1 - BL)}$=+0.3 [<+0.1, +0.6], +5.4% [+0.6%,

+10.1%]; $\Delta_{(FU-2\ -\ BL)}$=+0.5 [+0.1, +0.9], +8.1% [+2.3%, +13.9%]; $F_{2,30}$=4.8, p=0.015; f=0.160). Nevertheless, the magnitude of these effects did not reach the predefined threshold for reliably detectable size (i.e., f≥0.403) and should be interpreted with caution. By contrast, no significant changes were observed for Perceptual Abnormalities (PA; $F_{2,30}$=1.8, p=0.176) and Disorganized Speech (DS; $F_{2,30}$=1.9, p=0.169), as well as for role functioning (GF: Role; $F_{2,30}$=1.4, p=0.272). Among neuroimmune biomarkers, IL-8 demonstrated a significant but insufficient increase over time ($F_{2,30}$=3.7, p=0.036; f=0.267). The effects for HADS-D and IL-8, which were nonetheless rejected, were not confirmed by the non-parametric test (Supplementary Table 1). No treatment-emergent side-effects were reported. No participants transitioned to psychosis during the 12-week trial. One participant who discontinued PEA afterwards developed a FEP within the following 6 months, requiring initiation of a dopamine D2 receptor partial agonist.

When median-split baseline levels of neuroimmune biomarkers were included as covariates in the mixed-effects models with statistically significant assessment effects (i.e., CAARMS total APS and UTC), CHR participants with lower IL-1β levels exhibited greater improvement over time in UTC ($F_{2,28}$=4.8, p=0.017; Figure 2). No other statistically significant Biomarkers × Assessment interactions were observed (Supplementary Table 2).

In supplementary analyses, when neuroimmune biomarkers were modeled as time-varying moderators, significant Biomarker × Assessment interactions emerged. Specifically, IL-10/IL-6 ratio moderated changes in CAARMS Total APS ($F_{2,23}$=5.21, p=0.014; Figure 3), and IFN-γ ($F_{2,31}$=3.5, p=0.044) and IL-6 ($F_{2,24}$=4.4, p=0.024) moderated changes in UTC (Figure 4, Figure 5). No other statistically significant Biomarker × Assessment interactions were observed (Supplementary Table 3). Sensitivity analyses on complete data were substantially consistent with the main conclusions, with only minor variations.

4. Discussion

To the best of our knowledge, this is the first study exploring potential biobehavioral changes associated with PEA supplementation in subjects at CHR for psychosis.

Over a 12-week treatment period, PEA was well tolerated and associated with clinical improvements in overall APS, particularly in UTC. Together with suspiciousness/paranoia, unusual thoughts (i.e., delusional mood and perplexity, ideas of reference, bizarre ideas) are among the most prevalent APS in CHR populations [30, 31] and demonstrated predictive validity for transition to psychosis [32-34]. The magnitude of improvement observed in our sample appeared greater that that reported in control groups from comparable studies [35], although our follow-up period was shorter than that generally recommended to establish clinical efficacy [36]. Therefore, although these findings require

replications, early reductions in UTC may hold promise for PEA's clinical utility in CHR state. Additionally, despite lower statistical power, improvements were also observed in affective symptoms and social functioning. These exploratory findings converge towards the potential of PEA as a safe adjunctive treatment for transdiagnostic neuropsychiatric symptoms, often contributing to poor outcomes in CHR populations [13, 20].

Notably, baseline levels and longitudinal changes in selected psychosis-related inflammatory biomarkers (i.e., IL-1β, IL-10/IL-6 ratio, IFN-γ, IL-6) were found to interact with APS trajectories in meaningful and differential ways. Although exploratory in nature, these findings may inform future investigations.

First, lower baseline levels of IL-1β were associated with greater reductions in UTC intensity throughout the observation, suggesting that PEA's immunomodulatory effects may be especially beneficial for subsets of CHR individuals with a relatively lower systemic inflammatory tone (i.e., reduced acute-phase/pro-inflammatory cytokine activation), consistent with greater therapeutic benefit in subjects at an earlier or less advanced inflammatory stage [20].

Second, IL-10/IL-6 ratio moderated total APS intensity during PEA treatment. Specifically, higher IL-10/IL-6 ratio was associated to greater symptom improvement in 4 weeks, partially subsiding by the end of treatment, whereas lower IL-10/IL-6 ratio corresponded to slower but more sustained APS reduction over 12 weeks. This temporal divergence may indicate that participants with unbalanced anti-inflammatory tone may respond quickly but lack long-term recalibration, while those with a more pro-inflammatory yet plastic immune profile may derive progressive benefit from PEA's homeostatic effects. Interestingly, both IL-1β and IL-10/IL-6 ratio may represent reliable markers of the at-risk state subjects who would later convert to psychosis [6, 9, 37].

Third, time-varying increases in IL-6 and IFN-γ during the treatment period were overall associated with greater improvement in UTC intensity. Particularly, IL-6 showed a more complex temporal pattern, with higher levels linked to initial UTC worsening between baseline and 4 weeks, and a subsequent reversal toward improvement between 4 and 12 weeks. Transient increases in these cytokines may again reflect immune flexibility rather than harmful inflammation, not only aligning with the previous standpoint, but also with the evidence that functional immune responsiveness—not just inflammation *per se*—might modulate risk trajectories and treatment response in psychopathology [38].

Undeniably, this study should be seen considering some limitations.

First, the small sample size, which was nevertheless powered to detect large effects and is considered appropriate for a proof-of-concept study aimed at evaluating mechanistic outcomes.

Second, the lack of randomization and blinding procedures, which could help minimizing potential biases. Accordingly, factors beyond the effect of PEA alone, such as the intensity of therapeutic contact and expectation effects, may have influenced the efficacy findings. However, an open-label, single-arm design incorporating both clinical and biological assessments was deemed appropriate for an early-phase understanding of the biobehavioral effects of PEA supplementation in people at CHR for psychosis [39].

5. Conclusions

Our findings suggest that PEA supplementation may contribute to the improvement of APS in individuals at CHR state for psychosis. The most remarkable changes were observed in reductions of UTC intensity, which appeared to be associated with the dynamic modulation of peripheral CHR- and psychosis-related neuroimmune biomarkers.

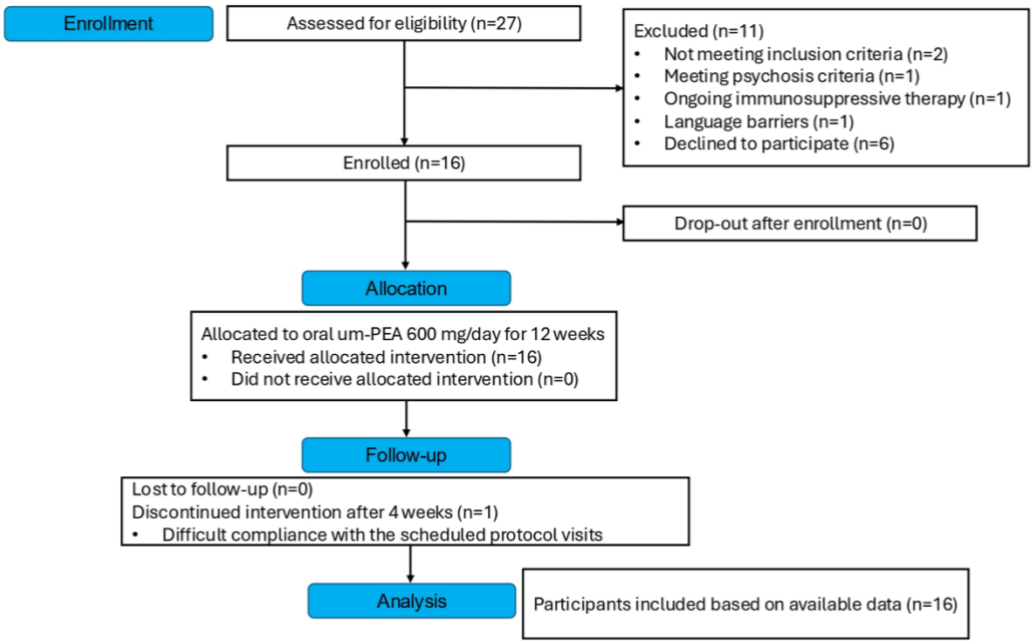
This evidence supports further investigation of PEA in subsequent phases of research to assess its clinical utility in this still therapy-orphan population.

Table 1. CAARMS Positive Symptoms in the sample: Longitudinal scores progression across assessments. Results of repeated measures analyses are presented for GRS (see also **Table S1** in Supplementary Materials for details). Scores were assessed at BL (beginning of PEA administration), at FU-1 (4 weeks), and at FU-2 (12 weeks).

		Mean ±SD				
Scale	**Score**	**BL** (n=16)	**FU-1** (n=15)	**FU-2** (n=15)	**Significance**	**ES**
P1, Total	[4×GRS]	12.6 ±2.56	9.7 ±3.83	8.9 ±3.86	*	Detectable
	[4×F&D]	13.3 ±4.24	11.02 ±3.396	2.1 ±1.94		
	[4×LoD]	358.7 ±35.83	327.5 ±86.66	304.0 ±97.75		
P1.1, UTC	[GRS]	3.8 ±1.33	2.6 ±1.81	1.9 ±1.64	*	Detectable
	[F&D]	3.4 ±1.55	2.5 ±1.81	2.8 ±1.90		
	[LoD]	69.7 ±34.56	43.5 ±39.91	36.0 ±40.50		
P1.2, NPI	[GRS]	3.8 ±1.56	2.9 ±1.06	2.7 ±1.71	*	Insufficient
	[F&D]	3.9 ±1.44	3.5 ±0.83	2.3 ±1.54		
	[LoD]	75.3 ±32.21	58.9 ±28.97	52.0 ±40.74		
P1.3, PA	[GRS]	3.1 ±1.00	2.4 ±1.40	2.8 ±1.42	-	-
	[F&D]	3.4 ±1.5	2.8 ±1.57	2.2 ±2.19		
	[LoD]	61.9 ±35.56	53.0 ±37.22	43.2 ±33.41		
P1.4, DS	[GRS]	1.9 ±1.59	1.8 ±1.42	1.5 ±1.41	-	-
	[F&D]	2.3 ±2.16	2.2 ±1.95	9.4 ±4.28		
	[LoD]	32.9 ±41.77	28.6 ±42.58	28.3 ±37.02		

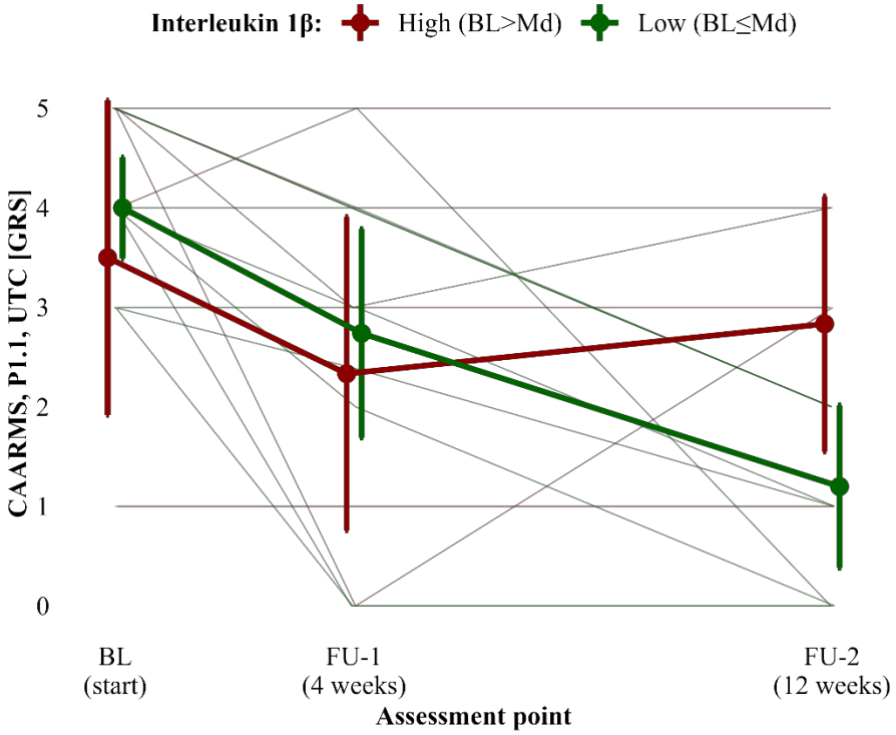
BL: Baseline assessment; CAARMS: Comprehensive Assessment of At-Risk Mental States; DS: Disorganised Speech (CAARMS-P1.4); ES: Effect-size; FU-1: First follow-up assessment (4 weeks); FU-2: Second follow-up assessment (12 weeks); GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); NBI: Non-Bizarre Ideas (CAARMS-P1.2); P1: CAARMS Positive Symptoms (UTC, NBI, PA, and DS); PA: Perceptual Abnormalities (CAARMS-P1.3); PEA: Palmitoylethanolamide (administered in its ultramicronized form); SD: Standard Deviation; UTC: Unusual Thought Content (CAARMS-P1.1); F&D: Frequency and Duration of CAARMS symptoms (ranging: 0-6); LoD: Level of Distress in relation to CAARMS symptoms (in percentage). *: Denotes a statistically significant effect of assessment time point ($p<0.050$), confirmed both with linear mixed effect model and with Friedman test. ES were considered detectable, with adequate power, for Cohen $f\geq0.401$ (insufficient elsewhere).

Figure 1. CONSORT Flow Diagram for Nonrandomized Clinical Trials.



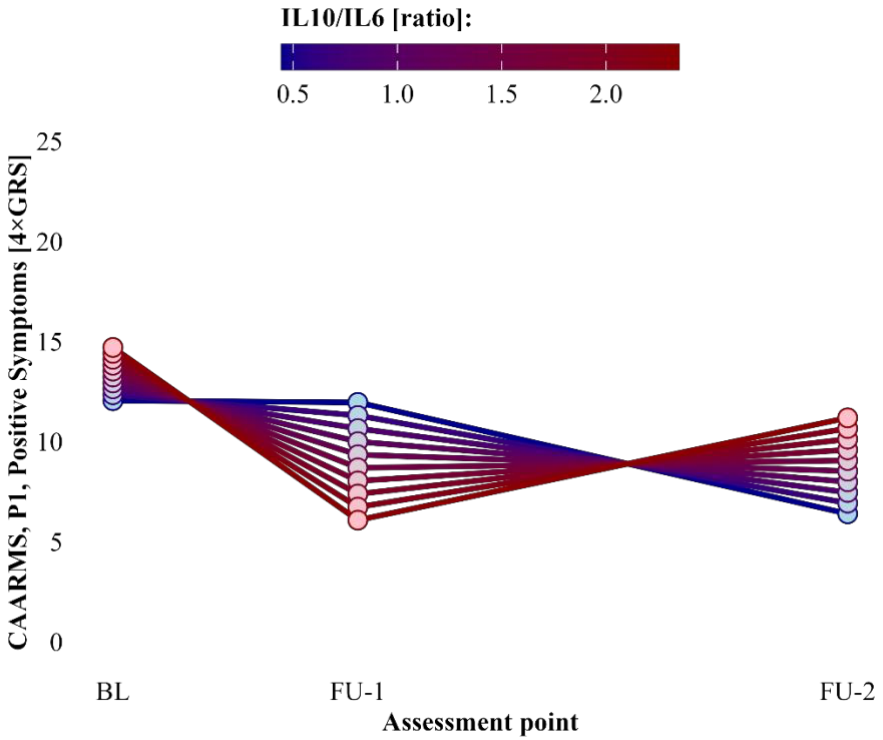
CAARMS: Comprehensive Assessment of At-Risk Mental States; um-PEA: Ultramicronized-Palmitoylethanolamide.

Figure 2. Trend of UTC score (P1.1, CAARMS) over time by IL-1β dichotomized at sample median. Linear mixed-effects model was fitted with the biomarker introduced as fixed effect. Dichotomization was based on BL measures.



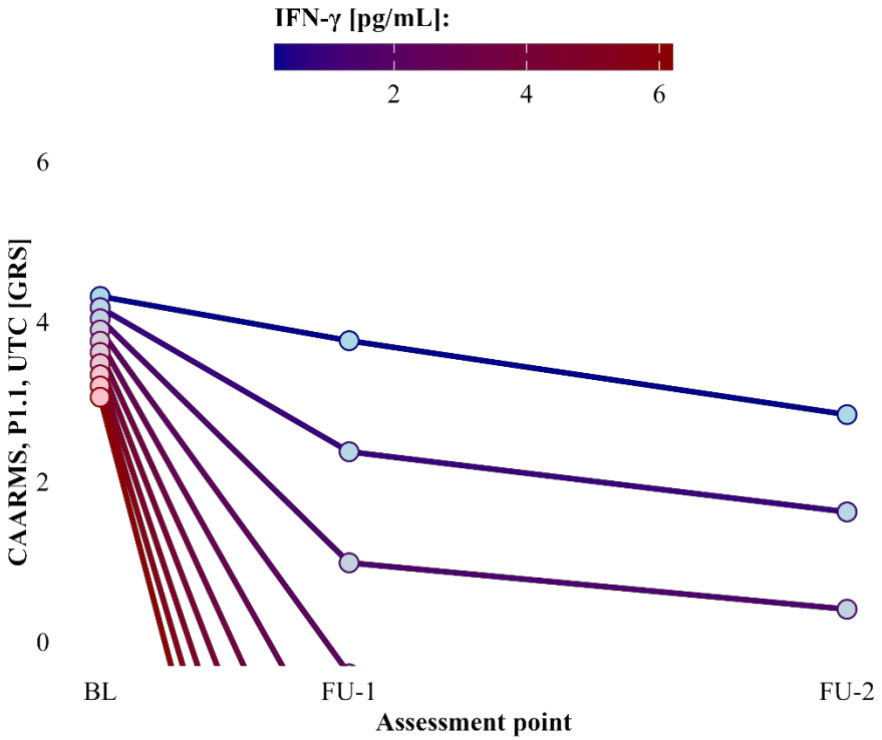
BL: Baseline; CAARMS: Comprehensive Assessment of At-Risk Mental States; FU-1: First follow-up assessment; FU-2: Second follow-up assessment; GRS: Global Rating Scale; IL: Interleukin; Md: median; UTC: Unusual Thought Content (CAARMS scale P1.1). Error bars indicate 95% confidence intervals of the mean.

Figure 3. Moderation of the temporal trajectory of CAARMS Total APS score as a function of IL-10/IL-6 ratio. Linear mixed-effects model was fitted with the biomarker introduced as a time-varying fixed effect. Model predictions are shown for values within the observed biomarker range of variation. Scores were assessed at BL, FU-1, and FU-2.



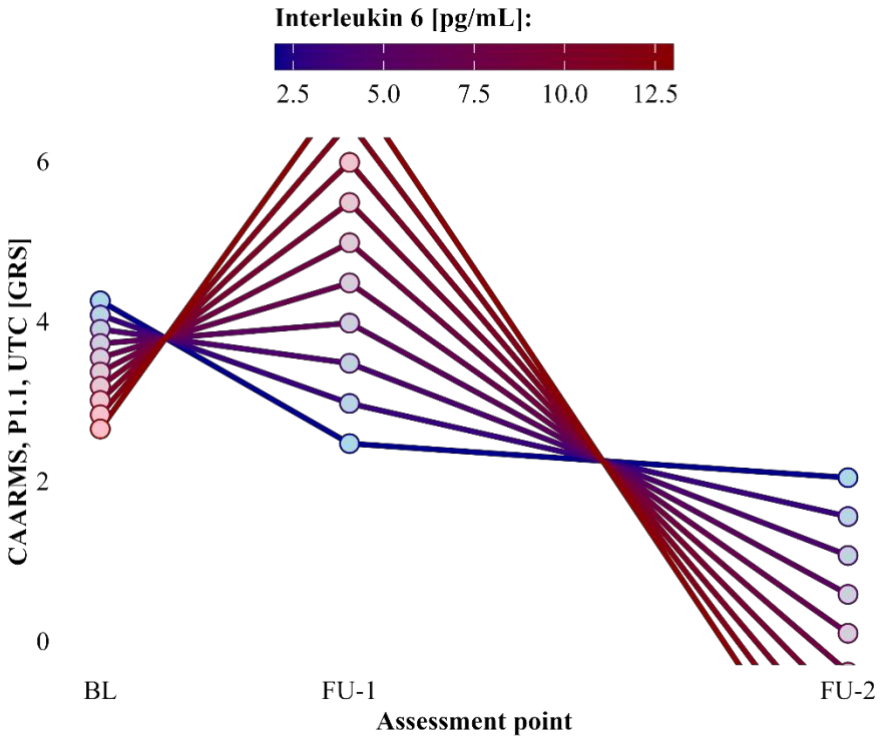
APS: Attenuated Psychotic Symptoms; BL: Baseline assessment (beginning of PEA administration); CAARMS: Comprehensive Assessment of At-Risk Mental States; FU-1: First follow-up assessment (4 weeks); FU-2: Second follow-up assessment (12 weeks); GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); IL: Interleukin; PEA: Palmitoylethanolamide (administered in its ultramicronized form); UTC: Unusual Thought Content (CAARMS-P1.1).

Figure 4. Moderation of the temporal trajectory of UTC score (P1.1, CAARMS) as a function of IFN-γ. Linear mixed-effects model was fitted with the biomarker introduced as a time-varying fixed effect. Model predictions are shown for values within the observed biomarker range of variation. Scores were assessed at BL, FU-1, and FU-2.



BL: Baseline assessment (beginning of PEA administration); CAARMS: Comprehensive Assessment of At-Risk Mental States; FU-1: First follow-up assessment (4 weeks); FU-2: Second follow-up assessment (12 weeks); GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); IFN-γ: Interferon-γ; PEA: Palmitoylethanolamide (administered in its ultramicronized form); UTC: Unusual Thought Content (CAARMS-P1.1).

Figure 5. Moderation of the temporal trajectory of UTC score (P1.1, CAARMS) as a function of IL-6. Linear mixed-effects model was fitted with the biomarker introduced as a time-varying fixed effect. Model predictions are shown for values within the observed biomarker range of variation. Scores were assessed at BL, FU-1, and FU-2.



BL: Baseline assessment (beginning of PEA administration); CAARMS: Comprehensive Assessment of At-Risk Mental States; FU-1: First follow-up assessment (4 weeks); FU-2: Second follow-up assessment (12 weeks); GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); IL: Interleukin; PEA: Palmitoylethanolamide (administered in its ultramicronized form); UTC: Unusual Thought Content (CAARMS-P1.1).

References

1. Yung, A.R., et al., *Mapping the onset of psychosis: the Comprehensive Assessment of At-Risk Mental States.* Aust N Z J Psychiatry, 2005. **39**(11-12): p. 964-71.
2. Davies, C., et al., *Lack of evidence to favor specific preventive interventions in psychosis: a network meta-analysis.* World Psychiatry, 2018. **17**(2): p. 196-209.
3. Fusar-Poli, P., et al., *The psychosis high-risk state: a comprehensive state-of-the-art review.* JAMA Psychiatry, 2013. **70**(1): p. 107-20.
4. Day, F.L., et al., *Blunted cortisol awakening response in people at ultra high risk of developing psychosis.* Schizophr Res, 2014. **158**(1-3): p. 25-31.
5. Misiak, B., et al., *Immune-inflammatory markers and psychosis risk: A systematic review and meta-analysis.* Psychoneuroendocrinology, 2021. **127**: p. 105200.
6. Mondelli, V., et al., *Serum immune markers and transition to psychosis in individuals at clinical high risk.* Brain Behav Immun, 2023. **110**: p. 290-296.
7. Nisha Aji, K., et al., *Interaction between peripheral and central immune markers in clinical high risk for psychosis.* Brain Behav Immun Health, 2023. **30**: p. 100636.
8. Park, S. and B.J. Miller, *Meta-analysis of cytokine and C-reactive protein levels in high-risk psychosis.* Schizophr Res, 2020. **226**: p. 5-12.
9. Zhang, T., et al., *Changes in inflammatory balance correlates with conversion to psychosis among individuals at clinical high-risk: A prospective cohort study.* Psychiatry Res, 2022. **318**: p. 114938.
10. Berger, M., et al., *Cortisol awakening response in patients with psychosis: Systematic review and meta-analysis.* Neurosci Biobehav Rev, 2016. **68**: p. 157-166.
11. Taylor, J.H., et al., *Immune and oxidative stress biomarkers in pediatric psychosis and psychosis-risk: Meta-analyses and systematic review.* Brain Behav Immun, 2024. **117**: p. 1-11.
12. Upthegrove, R., N. Manzanares-Teson, and N.M. Barnes, *Cytokine function in medication-naive first episode psychosis: a systematic review and meta-analysis.* Schizophr Res, 2014. **155**(1-3): p. 101-8.
13. Bortoletto, R., et al., *Questioning the role of palmitoylethanolamide in psychosis: a systematic review of clinical and preclinical evidence.* Front Psychiatry, 2023. **14**: p. 1231710.
14. Appiah-Kusi, E., et al., *Childhood trauma and being at-risk for psychosis are associated with higher peripheral endocannabinoids.* Psychol Med, 2020. **50**(11): p. 1862-1871.
15. Leweke, F.M., et al., *Cannabidiol enhances anandamide signaling and alleviates psychotic symptoms of schizophrenia.* Transl Psychiatry, 2012. **2**(3): p. e94.

16. Koethe, D., et al., *Familial abnormalities of endocannabinoid signaling in schizophrenia.* World Journal of Biological Psychiatry, 2019. **20**(2): p. 117-125.
17. Ibarra-Lecue, I., et al., *Cannabis use selectively modulates circulating biomarkers in the blood of schizophrenia patients.* Addiction Biology, 2022. **27**(6).
18. Parksepp, M., et al., *The Expanded Endocannabinoid System Contributes to Metabolic and Body Mass Shifts in First-Episode Schizophrenia: A 5-Year Follow-Up Study.* Biomedicines, 2022. **10**(2).
19. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.
20. Bortoletto, R., et al., *Palmitoylethanolamide supplementation for human health: A state-of-the-art systematic review of Randomized Controlled Trials in patient populations.* Brain Behav Immun Health, 2025. **43**: p. 100927.
21. Salazar de Pablo, G., et al., *Probability of Transition to Psychosis in Individuals at Clinical High Risk: An Updated Meta-analysis.* JAMA Psychiatry, 2021. **78**(9): p. 970-978.
22. Bhattacharyya, S., et al., *Effects of cannabidiol on symptoms in people at clinical high risk for psychosis.* World Psychiatry, 2024. **23**(3): p. 451-452.
23. Fusar-Poli, P., et al., *Heterogeneity of Psychosis Risk Within Individuals at Clinical High Risk: A Meta-analytical Stratification.* JAMA Psychiatry, 2016. **73**(2): p. 113-20.
24. Association, A.P., *Diagnostic and Statistical Manual of Mental Disorders*. 5th ed., text rev. ed. 2022: American Psychiatric Publishing.
25. Snaith, R.P. and A.S. Zigmond, *The hospital anxiety and depression scale.* Br Med J (Clin Res Ed), 1986. **292**(6516): p. 344.
26. Cornblatt, B.A., et al., *Preliminary findings for two new measures of social and role functioning in the prodromal phase of schizophrenia.* Schizophr Bull, 2007. **33**(3): p. 688-702.
27. Lingjaerde, O., et al., *The UKU side effect rating scale. A new comprehensive rating scale for psychotropic drugs and a cross-sectional study of side effects in neuroleptic-treated patients.* Acta Psychiatr Scand Suppl, 1987. **334**: p. 1-100.
28. Bortoletto, R., et al., *The Endocannabinoid Activity Remodulation for Psychosis Liability in Youth (EARLY) Study: An Open-Label Feasibility Trial of Ultramicronized-Palmitoylethanolamide Oral Supplementation in Clinical High-Risk State for Psychosis.* Brain Sci, 2024. **14**(12).

29. Shrestha, B. and L. Dunn, *The Declaration of Helsinki on Medical Research involving Human Subjects: A Review of Seventh Revision.* J Nepal Health Res Counc, 2020. **17**(4): p. 548-552.
30. Addington, J., et al., *North American Prodrome Longitudinal Study (NAPLS 2): The Prodromal Symptoms.* J Nerv Ment Dis, 2015. **203**(5): p. 328-35.
31. Alderman, T., et al., *Negative symptoms and impaired social functioning predict later psychosis in Latino youth at clinical high risk in the North American prodromal longitudinal studies consortium.* Early Interv Psychiatry, 2015. **9**(6): p. 467-75.
32. Kotlicka-Antczak, M., et al., *Short clinically-based prediction model to forecast transition to psychosis in individuals at clinical high risk state.* Eur Psychiatry, 2019. **58**: p. 72-79.
33. Addington, J., et al., *Progression from being at-risk to psychosis: next steps.* npj Schizophrenia, 2020. **6**(1): p. 27.
34. Perkins, D.O., et al., *Severity of thought disorder predicts psychosis in persons at clinical high-risk.* Schizophrenia Research, 2015. **169**(1): p. 169-177.
35. van der Gaag, M., et al., *Cognitive behavioral therapy for subjects at ultrahigh risk for developing psychosis: a randomized controlled clinical trial.* Schizophr Bull, 2012. **38**(6): p. 1180-8.
36. Nelson, B., et al., *Long-term follow-up of a group at ultra high risk ("prodromal") for psychosis: the PACE 400 study.* JAMA Psychiatry, 2013. **70**(8): p. 793-802.
37. Khoury, R. and H.A. Nasrallah, *Inflammatory biomarkers in individuals at clinical high risk for psychosis (CHR-P): State or trait?* Schizophr Res, 2018. **199**: p. 31-38.
38. Goldsmith, D.R., et al., *Inflammation-Related Functional and Structural Dysconnectivity as a Pathway to Psychopathology.* Biol Psychiatry, 2023. **93**(5): p. 405-418.
39. Jones, N.E., *Pathways to Treatment Development*, in *Encyclopedia of Behavioral Neuroscience, 2nd edition (Second Edition)*, S. Della Sala, Editor. 2022, Elsevier. p. 226-239.

Acknowledgement: The authors would like to thank the Biobehavioral Investigation in Neuropsychiatric and neurodevelopmental Disorders (BIND) team for support with participants' identification and data collection as well as acknowledge infrastructure from the University of Udine and the Friuli Centrale Health University Authority.

Funding: This work received no external funding.

CRediT authorship contribution statement:

RB: Conceptualization, Methodology, Writing-original draft, Writing – review and editing

MG: Conceptualization, Methodology, Formal analysis, Writing-original draft, Writing – review and editing

MBas: Conceptualization, Methodology, Writing – review and editing

CS: Conceptualization, Methodology, Writing – review and editing

OS: Conceptualization, Methodology, Writing-original draft, Writing – review and editing

MF: Conceptualization, Methodology, Writing – review and editing

FC: Conceptualization, Methodology, Writing – review and editing

MBal: Conceptualization, Methodology, Writing – review and editing

MC: Conceptualization, Methodology, Writing-original draft, Writing – review and editing, Supervision, Project administration

CHAPTER VI

General Discussion and Future Directions

The concluding aim of this doctoral research project was to explore the therapeutic potential of the dietary supplement palmitoylethanolamide (PEA) in young individuals identified as being at clinical high-risk (CHR) for psychosis [1, 2], focusing on both clinical and biological correlates. The CHR state is not only characterized by core attenuated or brief psychotic symptoms, but also by diverse concurrent psychopathology and psychiatric comorbidities severely impacting on the individuals' daily functioning [3, 4]. Providing innovative and reasonably safe therapeutic strategies in the CHR population is therefore crucial, considering, on the one hand, the lack of evidence-based gold-standard interventions to alleviate the burden of distressing psychopathological and the iatrogenic risks linked to the use of conventional antipsychotic medications [5-9], and, on the other hand, the inherent prognostic uncertainty that characterizes this condition [10].

The focus on PEA was primarily based on its natural mechanism of action targeting altered immune cell function and increased microglial activation leading to reduction of glutamate excitotoxicity via the endocannabinoid (eCB) system modulation [11, 12]. This pathway potentially intersects with early neurobiological processes implicated in the final dopaminergic dysregulation characterizing the natural course of psychosis [13, 14].

In this final section, we summarize and discuss the findings presented in the previous chapters, alongside additional observations from biobehavioral mechanistic studies of PEA in individuals at CHR state and other conditions of mental health vulnerability. Finally, we address the study's limitations, highlight remaining gaps in the literature, and outline potential directions for future research.

1. Summary of findings

In Chapter II, to achieve a more comprehensive understanding of the therapeutic applications of PEA across diverse medical conditions, we conducted a systematic review of 47 available Randomized-Controlled Trials (RCTs) assessing PEA in clinical populations [15]. Consistent with prior literature, the most robust efficacy findings emerged from studies on pain management [16-19]. However, a growing body of research pointed towards evidence of benefits over neuropsychiatric symptoms, either primary or secondary, suggesting a transdiagnostic potential of PEA in improving overall patient wellbeing, particularly for small-particle formulations (e.g., micronized-/ultramicronized-PEA). Notably, nearly all RCTs addressing neuropsychiatric conditions examined PEA as an add-on treatment to conventional psychopharmacological agents such as dopamine D2 receptor antagonists, selective serotonin reuptake inhibitors (SSRIs), or lithium, typically in moderate-to-severe clinical presentations (e.g., schizophrenia, bipolar disorder, autism spectrum disorders with significant behavioral dysregulation). By contrast, no studies to date have systematically assessed PEA as

adjunctive or stand-alone therapy in individuals with milder forms, early-stage mental illness, or nonspecific subthreshold psychic distress, despite its favorable safety, tolerability, and accessibility profile. Furthermore, converging clinical evidence suggests that PEA may exert disease-modifying effects, potentially through normalization of inflammatory biomarkers that are early disrupted in the course of several medical conditions, although such findings are still lacking for neuropsychiatric disorders [20-34].

In Chapter III, we extended our focus to the biobehavioral correlates of PEA in psychosis. To this aim, we conducted a second systematic review encompassing both clinical and preclinical findings in the field [35], in line with previous research exploring the role of PEA in other neuropsychiatric disorders [36-38]. Overall, while preclinical studies yielded no significant or consistent results [39, 40], evidence from clinical investigations was more robust, including both observational and interventional findings. Specifically, current literature converges on the notion that PEA signaling in peripheral blood and in the central nervous system (CNS) may mirror the stage of illness across different psychotic disorders and phenotypes [e.g., CHR for psychosis, first-episode psychosis (FEP), schizophrenia, schizophreniform disorder, bipolar disorder], being elevated during the early stages of illness, potentially as an innate homeostatic mechanism, before subsiding with disease progression [41-48]. Furthermore, evidence from clinical trials indicates that PEA add-on to treatment-as-usual (TAU) may mitigate specific symptom domains commonly observed in psychotic disorders (i.e., negative symptoms, manic symptoms), while not affecting positive psychotic symptoms (i.e., delusions, hallucinations). These findings highlight the need for further studies to assess the therapeutic potential of PEA supplementation and confirm its safety and tolerability profile [20, 27, 49]. Notably, the absence of clinical investigations on the effects of PEA supplementation as a standalone intervention in CHR states or as an adjunct to antipsychotics in early psychosis, together with the lack of data on its target biological pathways, provided the rationale for the investigator-initiated clinical trial detailed in the following chapters.

In Chapter IV, we detailed the methodology adopted for a proof-of-concept study of PEA in individuals at CHR for psychosis [50]. To ensure methodological rigor and transparency, the study was approved by the Institutional Review Board (IRB) and registered in a dedicated clinical trial repository. The study was intentionally designed as a feasibility-oriented, exploratory investigation, rather than a hypothesis-testing trial, with the primary aim of generating preliminary clinical and biological signals to inform future confirmatory research. Accordingly, we conducted a 12-week, open-label, non-randomized, single-arm trial of ultramicronized-PEA (um-PEA, 600 mg/day) to

assess the feasibility of enrolling 20 subjects at CHR for psychosis. Eligible individuals were young adults aged 18-35 years, referred to the Unit of Psychiatry and Eating Disorders at the Udine University Hospital for the treatment of attenuated psychotic symptoms (APS). Participants could then enter a subsequent 24-week open-label extension phase to further monitor clinical outcomes and biological trajectories under continued PEA supplementation. Throughout the treatment period, participants underwent assessments at multiple timepoints using standardized psychometric tools [2, 51, 52], alongside biological sample collection (i.e., blood/serum, stools) for mechanistic purposes.

In Chapter V, we described the main findings from the 12-week feasibility phase of our proof-of-concept study, the first to investigate the biobehavioral correlates of dietary PEA supplementation in individuals at CHR for psychosis [53]. Sixteen participants were evaluated at multiple timepoints through the Comprehensive Assessment of At-Risk Mental States (CAARMS) [2], together with peripheral neuroimmune biomarkers relevant to psychosis risk. Over the course of treatment, we observed significant reductions in total APS and unusual thought content (UTC), with no treatment-emergent side effects. Given the exploratory, non-comparator design of the study, these findings should be interpreted as preliminary clinical signals rather than definitive evidence of therapeutic efficacy. Exploratory analyses suggested that individual inflammatory profiles might shape treatment response, as lower baseline interleukin (IL)-1β and dynamic changes in IL-10/IL-6 ratio, interferon (IFN)-γ, and IL-6 appeared to modulate symptom improvement.

2. Reflections and advancing knowledge

2.1. Palmitoylethanolamide: a dietary supplement between disease modification and precision psychiatry

Altogether, findings from our proof-of-concept study suggest that PEA supplementation may modulate immune-inflammatory pathways relevant to psychosis course, leading to improvements in APS among individuals at CHR [53]. Although exploratory and requiring future confirmation, these observations offer several considerations. First, a subset of individuals at CHR with UTC may be more responsive to PEA, particularly those with lower peripheral IL-1β levels. Given that IL-1β is a pro-inflammatory cytokine primarily released by innate immune cells (e.g., monocytes/macrophages) during the early phases of the inflammatory cascade [54] and has been proposed as a relatable biomarker of progression to psychosis [55-57], it could be speculated that earlier intervention with PEA could yield better outcomes. IL-1β would therefore serve not only as a prognostic biomarker for identifying individuals most eligible for early interventions, but also as specifically predictive of differential responsiveness to PEA [58]. This would also align with the endogenous homeostatic role

of PEA described in Chapter III, characterized by an early increase and subsequent decline of its peripheral and central levels throughout the psychosis trajectory, suggesting that exogenous supplementation may help prevent such inherent biological depletion [35].

Second, consistent with findings outlined in Chapter II, it was not possible to isolate direct effects of PEA on single inflammatory biomarkers, not only suggesting that PEA may operate through different, multiple pathways in individuals at CHR, but also reinforcing its pro-homeostatic rather than solely anti-inflammatory properties [15]. To this extent, the observation of more sustained APS intensity improvements in participants with lower time-varying IL-10/IL-6 ratio, alongside more pronounced UTC reductions in those showing higher time-varying IFN-γ and IL-6 levels, should be interpreted as hypothesis-generating associations rather than pathway-specific effects. Such pattern aligns with the notion that PEA's modulatory effects emerge more clearly when counter-regulatory mechanisms are insufficient, but the immune-inflammatory system remains biologically plastic and receptive.

Finally, no treatment-emergent adverse effects were observed in our sample, a finding of particular ethical relevance given the young age of CHR individuals and the unpredictable nature of their clinical trajectories, which call for the safest possible therapeutic approaches [10]. A single participant reported mild appetite loss after study completion, which did not require medical intervention and may reflect the involvement of eCBs and eCB-like compounds such as anandamide (AEA) and oleoylethanolamide (OEA) in the physiological regulation of appetite and energy balance [59]. The favorable safety profile of PEA as a dietary supplement further supports its potential as a minimally intrusive and acceptable preventive intervention for youth patients and their caregivers.

2.2. Beyond attenuated psychotic symptoms: exploring the potential of palmitoylethanolamide on further symptom dimensions in clinical high-risk

Alongside APS, individuals at CHR for psychosis often present with other clinically relevant manifestations, particularly negative symptoms (i.e., alogia, apathy/avolition, anhedonia), impaired cognition, broader psychopathology, and decline in role and social functioning, which are closely related to each other [60-63]. These domains are disabling aspects of the CHR condition contributing substantially to subjective stress and reduced quality of life, also independently from progression to psychosis [61]. Although our proof-of-concept study primarily focused on APS, which operationally define the CHR state [2], further analyses were conducted on negative/cognitive, general psychopathology, and functional dimensions to gain a more comprehensive understanding of PEA's potential effects [50]. Over the 12-week supplementation period, participants showed a significant improvement in total negative symptoms [$F(2,28)=7.93$, $p=0.002$; Cohen's $f=0.284$, medium; $\eta^2_p=0.075$] and general psychopathology [$F(2,30)=12.40$, $p<0.001$; Cohen's $f=0.422$, large;

η^2_p=0.151], along with a trend toward improvement in total cognitive symptoms [F(2,28)=2.53, p=0.098], as measured using the CAARMS [2]. In parallel, social functioning improved [F(2,30)=4.81, p=0.015; Cohen's f=0.160, small; η^2_p=0.025], while role functioning did not show significant change [F(2,30)=1.36, p=0.272], according to the Global Functioning (GF)-Role and Social scales [64]. When baseline levels of neuroimmune biomarkers were included as covariates in mixed-effects models, participants with higher IL-1β [Assessment × Biomarker interaction: F(2,28)=3.38, p=0.049] and Neutrophils/Lymphocytes ratio [Assessment × Biomarker interaction: F(2,28)=4.98, p=0.014] exhibited a sharp improvement in social functioning at 4 weeks, that plateaued thereafter, whereas those with lower baseline levels displayed a steadier but gradual improvement over time. Similarly, higher baseline levels of 2-arachidonoylgylcerol (2-AG) predicted greater reductions in general psychopathology levels over time [Assessment × Biomarker interaction: F(2,20)=4.56, p=0.023].

While exploratory, these analyses were directionally consistent with APS reductions and their biological correlates, suggesting a convergent clinical effect of PEA across multiple psychopathological dimensions of the CHR state. Importantly, these improvements emerged in a population typically characterized by low baseline functioning and poor motivation, further supporting the opportunity of PEA supplementation in such a vulnerable group [63, 65].

Negative symptoms, psychopathology burden, and functional impairment are recognized as major predictors of long-term outcomes in CHR individuals, often exceeding the prognostic power of APS for transition to psychosis, poorer response to psychosocial interventions, and higher risk of chronic impairment even among non-converters [60, 61, 65-70]. Yet, current interventions remain largely ineffective for these domains, underscoring the need for novel therapeutic approaches [5, 9].

From a translational standpoint, such evidence aligns with research implicating inflammation and oxidative stress across the negative, cognitive, and functional dimensions of psychosis [71-73], as well as with the proven benefits of PEA supplementation on these domains in schizophrenia [27, 35]. Additionally, preliminary associations with higher 2-AG levels further suggest that individuals with a more responsive eCB tone, or preserved endogenous PEA availability typical of early illness stages, may exhibit faster clinical improvement, emphasizing the importance of early intervention [12, 35]. Within this framework, functional improvements may reflect indirect effects on immune–glial–eCB-dependent processes such as stress resilience, affective regulation, and neurocognitive efficiency [74-76]. Aligning with recent models of the CHR state [62, 77], recovery is likely to depend not only on interventions targeting APS, but also on those addressing motivational, affective, and cognitive deficits that share overlapping pathophysiological mechanisms.

2.3. Shared pathways, different faces: palmitoylethanolamide in autistic adults

An additional reflection emerging from this doctoral work concerns the broader applicability of PEA supplementation to other symptom-orphan conditions, characterized by underpinning immune system abnormalities, altered microglial activation, and glutamate excitotoxicity [78-82]. Among these, autism represents a paradigmatic example for increasing challenges in social functioning, occupational difficulties, and liability to psychic distress and mental health comorbidities in adulthood [83-88]. Despite increasing referrals to adult mental health services, particularly autistic individuals without intellectual disability often remain undertreated, as conventional psychotropic medications show limited efficacy and tolerability, particularly for affective or anxiety-related symptoms [89-98]. In previous review of the literature, we outlined the biobehavioral role of PEA in autism spectrum disorder (ASD), including a clinical study in children and adolescents showing improvements in core ASD symptoms and good tolerability [37]. By contrast, evidence on PEA supplementation in autistic adults remains scarce and mostly anecdotal, with conflicting findings across severity levels. Notably, we reported two cases in which mood and social functioning improved in a level 1 autistic adult, but not in a level 2/3 individual, accompanied by reduced inflammatory activity detected through Raman spectroscopy and enhanced AEA signaling. [99]. Subsequently, within a parallel 12-week proof-of-concept study, we assessed changes in symptoms of psychic distress among fifteen level 1 autistic adults without intellectual disability during treatment with PEA [100]. Our study yielded significant improvements in perceived anxiety, overall psychic distress, and adaptive functioning, with no treatment-emergent adverse effects. These effects were particularly evident among participants exhibiting an imbalance toward acute pro-inflammatory activity, including higher baseline IFN-γ, IL-8, and IL-17A, but lower IL-10 and erythrocyte sedimentation rate (ESR). These preliminary findings mirror those observed in the CHR sample and reinforce the hypothesis that PEA may exert beneficial effects across diagnostic boundaries by targeting shared pathophysiological processes [15]. Indeed, in both CHR and adult ASD, individuals exhibiting a more reactive or pro-inflammatory biological profile appeared to benefit the most from treatment. This pattern supports a model in which PEA acts as a homeostatic modulator, most effective when endogenous regulatory systems are disrupted but still biologically plastic, representing a transdiagnostic therapeutic tool for early or atypical neurodevelopmental conditions characterized by psychic distress, functional impairment, and immune-glial imbalance.

3. Limitations and emerging questions

3.1. Methodological rigor and design considerations

Despite the encouraging findings, this doctoral project is not devoid of methodological limitations. First, while the two systematic reviews presented in Chapter II and III [15, 35] were performed rigorously according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines [101], the heterogeneity of the available literature in terms of sample characteristics, study design, and outcome measures, prevented a meta-analytic synthesis. As a result, it was not possible to draw quantitative conclusions about the magnitude and consistency of PEA effects across neuropsychiatric symptoms and psychosis-related conditions.

Regarding the empirical components presented in Chapters IV and V [50, 53], the open-label, non-randomized, and non-comparator design precludes firm causal inferences and limits control over expectancy effects and investigator bias. Although this approach was appropriate for a proof-of-concept study with mechanistic objectives, intended to explore potential biological correlates of clinical response, future research should prioritize randomized controlled designs with placebo or active comparators to validate the PEA's efficacy and specificity.

3.2. Recruitment challenges and sample characteristics

Recruiting individuals at CHR for psychosis proved challenging. First, the number of referrals to the Unit of Psychiatry and Eating Disorders over a two-year period was slightly below the estimated 19.2% epidemiological prevalence of the CHR state in clinical samples [102]. This rate included patients referred both to the internal transition-age mental health prevention service and to the adult neurodevelopment service, possibly reflecting difficulties in continuity of care between tertiary centers and community mental health services [92]. Second, as shown in Figure 1 of Chapter V, among twenty-seven screened individuals, approximately one third did not entirely meet eligibility criteria for study inclusion, had already transitioned to FEP, or declined participation due to reluctance to engage in clinical research involving experimental interventions. Consequently, the feasibility objective of enrolling at least 20 participants at CHR state during the observation period was not achieved, despite power analysis confirmed the adequacy of the obtained sample for mechanistic purposes. On the other hand, retention among allocated participants exceeded 93% across the 12-week observation period, supporting the acceptability of PEA supplementation in this population. Future studies involving underage participants and other CHR subgroups will be crucial to enhance the representativeness and generalizability of findings on PEA's effects in CHR state.

Although every effort was made to characterize the sample comprehensively at baseline, including levels of functional impairment and self-reported anxiety and depressive symptoms, future studies should systematically employ the 16-item version of the Prodromal Questionnaire (PQ-16) alongside the CAARMS to also delineate the subjective features and distress levels associated with APS [103].

Additionally, incorporating standardized, repeated assessments of substance use and stressful life events [104, 105] would allow for a more accurate evaluation of factors that may influence treatment responsiveness, adherence, and long-term outcomes.

3.3. Frequency, dosing, and clinical heterogeneity

The 12-week phase of the proof-of-concept study employed a single daily dosing regimen of um-PEA (600 mg/day in the morning), selected for its favorable safety and mechanistic rationale.

The small-particle formulation was chosen due to its superior penetrance in the CNS and therapeutic efficacy across transdiagnostic symptom domains involving neuropsychiatric and somatic conditions sustained by neuroinflammatory processes [11, 15]. However, it remains unclear whether alternative dosing schedules (e.g., 1200 mg/day, divided into two doses), to be explored in the trial extension phase, or different formulations (e.g., Co-ultramicronized PEA plus Luteolin, LipiSperse PEA) could yield differential outcomes or improve bioavailability, thereby partially limiting the generalizability of the present findings to um-PEA.

3.4. What next? Long-term outcomes and integrative mechanisms

Another limitation concerns the relatively short observation period (12 weeks), which provides only a glimpse of early clinical and biological changes and does not allow conclusions to be drawn regarding outcomes typically explored in CHR samples, including transition to psychosis [106]. Longer-term follow-up studies are needed to evaluate the durability of improvements in APS, negative and cognitive symptoms, and functional outcomes in PEA-treated CHR individuals, as well as potential delayed or cumulative effects on inflammatory and neuroimmune pathways.

Furthermore, emerging evidence highlights intricate crosstalk between the eCB system, peripheral inflammation, and the gut–brain axis, with potential implications for PEA's mechanism of action. Alterations in microbiota composition can influence eCB tone, immune activation, and stress reactivity, thereby shaping the therapeutic response to PEA [107]. The microbiome analyses and complete eCB profiling currently underway on blood and stool samples collected during this proof-of-concept study are expected to provide deeper insights into how PEA exerts its biobehavioral effects and to help identify biological signatures of treatment responsiveness.

Together with the forthcoming findings from the six-month clinical extension phase, these analyses will complement the present results and contribute to a more comprehensive understanding of PEA's clinical and biological effects. Such data will help clarify whether the effects of PEA in this populations are merely symptomatic or reflect disease-modifying properties.

4. Conclusions

This doctoral project explored the biobehavioral potential of PEA, an eCB-like lipid mediator, as a safe and mechanistically grounded intervention for mental health vulnerability states. Through a combination of systematic reviews of current literature and proof-of-concept studies, the research aimed to establish a translational framework linking immune-glial modulation to clinical improvement in therapy-orphan conditions, primarily including the CHR state for psychosis [15, 35, 50, 53]. The systematic reviews identified converging evidence supporting emerging benefits of PEA for neuropsychiatric symptoms and psychosis. Building upon this foundation, the empirical study indicated improvements in APS among CHR individuals during PEA supplementation, without treatment-emergent adverse effects. Exploratory analyses suggested that individuals with more reactive or pro-inflammatory immune profiles may benefit the most, reinforcing the hypothesis that PEA acts as a homeostatic modulator where endogenous regulation is compromised but biological plasticity persists. These findings provide preliminary support for the use of PEA as a transdiagnostic and biologically informed early intervention, targeting shared neuroinflammatory mechanisms across distinct vulnerability conditions. Methodologically, the project demonstrated the feasibility and acceptability of dietary supplement-based, biomarker-informed designs in young help-seeking populations at CHR for psychosis.

In conclusion, this work contributes to a growing paradigm emphasizing the role of neuroimmune modulation as a key target for early mental health intervention. By bridging clinical and biological perspectives, it posits PEA as a model compound for developing safe, personalized, and mechanism-driven strategies in precision psychiatry.

References

1. Yung, A.R. and B. Nelson, *Young people at ultra high risk for psychosis: research from the PACE clinic.* Braz J Psychiatry, 2011. **33 Suppl 2**: p. s143-60.
2. Yung, A.R., et al., *Mapping the onset of psychosis: the Comprehensive Assessment of At-Risk Mental States.* Aust N Z J Psychiatry, 2005. **39**(11-12): p. 964-71.
3. Fusar-Poli, P., et al., *The psychosis high-risk state: a comprehensive state-of-the-art review.* JAMA Psychiatry, 2013. **70**(1): p. 107-20.
4. Solmi, M., et al., *Meta-analytic prevalence of comorbid mental disorders in individuals at clinical high risk of psychosis: the case for transdiagnostic assessment.* Mol Psychiatry, 2023. **28**(6): p. 2291-2300.
5. Davies, C., et al., *Lack of evidence to favor specific preventive interventions in psychosis: a network meta-analysis.* World Psychiatry, 2018. **17**(2): p. 196-209.
6. Davies, C., et al., *Efficacy and Acceptability of Interventions for Attenuated Positive Psychotic Symptoms in Individuals at Clinical High Risk of Psychosis: A Network Meta-Analysis.* Front Psychiatry, 2018. **9**: p. 187.
7. Zhang, T., et al., *Further evidence that antipsychotic medication does not prevent long-term psychosis in higher-risk individuals.* Eur Arch Psychiatry Clin Neurosci, 2022. **272**(4): p. 591-602.
8. Bastiampillai, T., et al., *Antipsychotic treatment in clinical high risk for psychosis: Iatrogenesis related to dopamine supersensitivity psychosis?* Aust N Z J Psychiatry, 2022. **56**(1): p. 97.
9. Minichino, A., et al., *Preventing psychosis in people at clinical high risk: an updated meta-analysis by the World Psychiatric Association Preventive Psychiatry section.* Mol Psychiatry, 2025. **30**(6): p. 2773-2782.
10. Polari, A., et al., *Clinical trajectories in the ultra-high risk for psychosis population.* Schizophr Res, 2018. **197**: p. 550-556.
11. Petrosino, S. and V. Di Marzo, *The pharmacology of palmitoylethanolamide and first data on the therapeutic efficacy of some of its new formulations.* Br J Pharmacol, 2017. **174**(11): p. 1349-1365.
12. Petrosino, S., et al., *The anti-inflammatory mediator palmitoylethanolamide enhances the levels of 2-arachidonoyl-glycerol and potentiates its actions at TRPV1 cation channels.* Br J Pharmacol, 2016. **173**(7): p. 1154-62.
13. Basavaraju, R., et al., *Hippocampal Glutamate and Positive Symptom Severity in Clinical High Risk for Psychosis.* JAMA Psychiatry, 2022. **79**(2): p. 178-179.

14. Bojesen, K.B., et al., *Cerebral glutamate levels over two years in initially antipsychotic-naïve first-episode patients with psychosis are related to clinical symptoms and cognition.* Molecular Psychiatry, 2025.
15. Bortoletto, R., et al., *Palmitoylethanolamide supplementation for human health: A state-of-the-art systematic review of Randomized Controlled Trials in patient populations.* Brain Behav Immun Health, 2025. **43**: p. 100927.
16. Lang-Illievich, K., et al., *Palmitoylethanolamide in the Treatment of Chronic Pain: A Systematic Review and Meta-Analysis of Double-Blind Randomized Controlled Trials.* Nutrients, 2023. **15**(6).
17. Scuteri, D., et al., *Effects of Palmitoylethanolamide (PEA) on Nociceptive, Musculoskeletal and Neuropathic Pain: Systematic Review and Meta-Analysis of Clinical Evidence.* Pharmaceutics, 2022. **14**(8).
18. Schweiger, V., et al., *Extended Treatment with Micron-Size Oral Palmitoylethanolamide (PEA) in Chronic Pain: A Systematic Review and Meta-Analysis.* Nutrients, 2024. **16**(11).
19. Varrassi, G., et al., *A Decades-Long Journey of Palmitoylethanolamide (PEA) for Chronic Neuropathic Pain Management: A Comprehensive Narrative Review.* Pain Ther, 2024.
20. Abedini, T., et al., *Efficacy and safety of palmitoylethanolamide as an adjunctive treatment for acute mania: A randomized, double-blind, placebo-controlled trial.* Psychiatry Clin Neurosci, 2022. **76**(10): p. 505-511.
21. Campolo, M., et al., *Co-Ultra PEALut Enhances Endogenous Repair Response Following Moderate Traumatic Brain Injury.* Int J Mol Sci, 2021. **22**(16).
22. Evangelista, M., et al., *Ultra-micronized Palmitoylethanolamide Effects on Sleep-wake Rhythm and Neuropathic Pain Phenotypes in Patients with Carpal Tunnel Syndrome: An Open-label, Randomized Controlled Study.* CNS Neurol Disord Drug Targets, 2018. **17**(4): p. 291-298.
23. Ghazizadeh-Hashemi, M., et al., *Palmitoylethanolamide as adjunctive therapy in major depressive disorder: A double-blind, randomized and placebo-controlled trial.* J Affect Disord, 2018. **232**: p. 127-133.
24. Khalaj, M., et al., *Palmitoylethanolamide as adjunctive therapy for autism: Efficacy and safety results from a randomized controlled trial.* J Psychiatr Res, 2018. **103**: p. 104-111.
25. Lunardelli, M.L., et al., *Co-ultraPEALut: Role in Preclinical and Clinical Delirium Manifestations.* CNS Neurol Disord Drug Targets, 2019. **18**(7): p. 530-554.
26. Rao, A., et al., *Palmitoylethanolamide for sleep disturbance. A double-blind, randomised, placebo-controlled interventional study.* Sleep Sci Pract, 2021. **5**(1): p. 12.

27. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.
28. Versace, V., et al., *Co-ultramicronized palmitoylethanolamide/luteolin normalizes GABA(B)-ergic activity and cortical plasticity in long COVID-19 syndrome.* Clin Neurophysiol, 2023. **145**: p. 81-88.
29. Andresen, S.R., et al., *Ultramicronized palmitoylethanolamide in spinal cord injury neuropathic pain: a randomized, double-blind, placebo-controlled trial.* Pain, 2016. **157**(9): p. 2097-2103.
30. Gagliano, C., et al., *Ocular hypotensive effect of oral palmitoyl-ethanolamide: a clinical trial.* Invest Ophthalmol Vis Sci, 2011. **52**(9): p. 6096-100.
31. Orefice, N.S., et al., *Oral Palmitoylethanolamide Treatment Is Associated with Reduced Cutaneous Adverse Effects of Interferon-beta1a and Circulating Proinflammatory Cytokines in Relapsing-Remitting Multiple Sclerosis.* Neurotherapeutics, 2016. **13**(2): p. 428-38.
32. Pickering, E., et al., *A randomized controlled trial assessing the safety and efficacy of palmitoylethanolamide for treating diabetic-related peripheral neuropathic pain.* Inflammopharmacology, 2022. **30**(6): p. 2063-2077.
33. Steels, E., et al., *A double-blind randomized placebo controlled study assessing safety, tolerability and efficacy of palmitoylethanolamide for symptoms of knee osteoarthritis.* Inflammopharmacology, 2019. **27**(3): p. 475-485.
34. Bonzanino, M., et al., *PEALut in the Dietary Management of Patients with Acute Ischemic Stroke: A Prospective Randomized Controlled Clinical Trial.* J Clin Med, 2024. **13**(2).
35. Bortoletto, R., et al., *Questioning the role of palmitoylethanolamide in psychosis: a systematic review of clinical and preclinical evidence.* Front Psychiatry, 2023. **14**: p. 1231710.
36. Colizzi, M., et al., *Therapeutic effect of palmitoylethanolamide in cognitive decline: A systematic review and preliminary meta-analysis of preclinical and clinical evidence.* Frontiers in Psychiatry, 2022. **13**.
37. Colizzi, M., et al., *Palmitoylethanolamide and Its Biobehavioral Correlates in Autism Spectrum Disorder: A Systematic Review of Human and Animal Evidence.* Nutrients, 2021. **13**(4).
38. Bortoletto, R., et al., *Is It Time to Test the Antiseizure Potential of Palmitoylethanolamide in Human Studies? A Systematic Review of Preclinical Evidence.* Brain Sciences, 2022. **12**(1).

39. Stark, T., et al., *Peripubertal cannabidiol treatment rescues behavioral and neurochemical abnormalities in the MAM model of schizophrenia.* Neuropharmacology, 2019. **146**: p. 212-221.
40. Di Bartolomeo, M., et al., *Crosstalk between the transcriptional regulation of dopamine D2 and cannabinoid CB1 receptors in schizophrenia: Analyses in patients and in perinatal Δ9-tetrahydrocannabinol-exposed rats.* Pharmacol Res, 2021. **164**: p. 105357.
41. Leweke, F.M., et al., *Elevated endogenous cannabinoids in schizophrenia.* NeuroReport, 1999. **10**(8): p. 1665-1669.
42. Leweke, F.M., et al., *Cannabidiol enhances anandamide signaling and alleviates psychotic symptoms of schizophrenia.* Transl Psychiatry, 2012. **2**(3): p. e94.
43. Muguruza, C., et al., *Quantification of endocannabinoids in postmortem brain of schizophrenic subjects.* Schizophrenia Research, 2013. **148**(1-3): p. 145-150.
44. Koethe, D., et al., *Familial abnormalities of endocannabinoid signaling in schizophrenia.* World Journal of Biological Psychiatry, 2019. **20**(2): p. 117-125.
45. Appiah-Kusi, E., et al., *Childhood trauma and being at-risk for psychosis are associated with higher peripheral endocannabinoids.* Psychol Med, 2020. **50**(11): p. 1862-1871.
46. Ibarra-Lecue, I., et al., *Cannabis use selectively modulates circulating biomarkers in the blood of schizophrenia patients.* Addiction Biology, 2022. **27**(6).
47. Parksepp, M., et al., *The Expanded Endocannabinoid System Contributes to Metabolic and Body Mass Shifts in First-Episode Schizophrenia: A 5-Year Follow-Up Study.* Biomedicines, 2022. **10**(2).
48. Topuz, R.D., Y. Gorgulu, and M.K. Uluturk, *Could serum endocannabinoid and N-acylethanolamine levels be important in bipolar disorder?* World Journal of Biological Psychiatry: p. 7.
49. Brotini, S., C. Schievano, and L. Guidi, *Ultra-micronized Palmitoylethanolamide: An Efficacious Adjuvant Therapy for Parkinson's Disease.* CNS Neurol Disord Drug Targets, 2017. **16**(6): p. 705-713.
50. Bortoletto, R., et al., *The Endocannabinoid Activity Remodulation for Psychosis Liability in Youth (EARLY) Study: An Open-Label Feasibility Trial of Ultramicronized-Palmitoylethanolamide Oral Supplementation in Clinical High-Risk State for Psychosis.* Brain Sci, 2024. **14**(12).
51. Snaith, R.P. and A.S. Zigmond, *The hospital anxiety and depression scale.* Br Med J (Clin Res Ed), 1986. **292**(6516): p. 344.

52. Lingjaerde, O., et al., *The UKU side effect rating scale. A new comprehensive rating scale for psychotropic drugs and a cross-sectional study of side effects in neuroleptic-treated patients.* Acta Psychiatr Scand Suppl, 1987. **334**: p. 1-100.
53. Bortoletto, R., et al., *Effects of palmitoylethanolamide in clinical high-risk for psychosis: A nonrandomized open-label trial.* Brain, Behavior, & Immunity - Health, 2025. **50**: p. 101141.
54. Italiani, P., et al., *Profiling the Course of Resolving vs. Persistent Inflammation in Human Monocytes: The Role of IL-1 Family Molecules.* Front Immunol, 2020. **11**: p. 1426.
55. Zhang, T., et al., *Changes in inflammatory balance correlates with conversion to psychosis among individuals at clinical high-risk: A prospective cohort study.* Psychiatry Res, 2022. **318**: p. 114938.
56. Mondelli, V., et al., *Serum immune markers and transition to psychosis in individuals at clinical high risk.* Brain Behav Immun, 2023. **110**: p. 290-296.
57. Khoury, R. and H.A. Nasrallah, *Inflammatory biomarkers in individuals at clinical high risk for psychosis (CHR-P): State or trait?* Schizophr Res, 2018. **199**: p. 31-38.
58. Cagney, D.N., et al., *The FDA NIH Biomarkers, EndpointS, and other Tools (BEST) resource in neuro-oncology.* Neuro-Oncology, 2017. **20**(9): p. 1162-1172.
59. Di Marzo, V. and I. Matias, *Endocannabinoid control of food intake and energy balance.* Nat Neurosci, 2005. **8**(5): p. 585-9.
60. Devoe, D.J., et al., *Negative Symptoms and Functioning in Youth at Risk of Psychosis: A Systematic Review and Meta-analysis.* Harv Rev Psychiatry, 2020. **28**(6): p. 341-355.
61. Carrión, R.E., et al., *Prediction of Functional Outcome in Individuals at Clinical High Risk for Psychosis.* JAMA Psychiatry, 2013. **70**(11): p. 1133-1142.
62. Fusar-Poli, P., et al., *Prevention of Psychosis: Advances in Detection, Prognosis, and Intervention.* JAMA Psychiatry, 2020. **77**(7): p. 755-765.
63. McGorry, P.D. and C. Mei, *Ultra-high-risk paradigm: lessons learnt and new directions.* Evid Based Ment Health, 2018. **21**(4): p. 131-133.
64. Cornblatt, B.A., et al., *Preliminary findings for two new measures of social and role functioning in the prodromal phase of schizophrenia.* Schizophr Bull, 2007. **33**(3): p. 688-702.
65. Yung, A.R., et al., *Persistent negative symptoms in individuals at Ultra High Risk for psychosis.* Schizophr Res, 2019. **206**: p. 355-361.
66. Hauke, D.J., et al., *Multimodal prognosis of negative symptom severity in individuals at increased risk of developing psychosis.* Translational Psychiatry, 2021. **11**(1): p. 312.

67. Glenthøj, L.B., et al., *Experiential negative symptoms are more predictive of real-life functional outcome than expressive negative symptoms in clinical high-risk states.* Schizophr Res, 2020. **218**: p. 151-156.
68. Schlosser, D.A., et al., *Modeling the role of negative symptoms in determining social functioning in individuals at clinical high risk of psychosis.* Schizophr Res, 2015. **169**(1-3): p. 204-208.
69. Devoe, D.J., A. Peterson, and J. Addington, *Negative Symptom Interventions in Youth at Risk of Psychosis: A Systematic Review and Network Meta-analysis.* Schizophr Bull, 2018. **44**(4): p. 807-823.
70. Devoe, D.J., et al., *Interventions and social functioning in youth at risk of psychosis: A systematic review and meta-analysis.* Early Interv Psychiatry, 2019. **13**(2): p. 169-180.
71. Kogan, S., et al., *The impact of inflammation on neurocognition and risk for psychosis: a critical review.* Eur Arch Psychiatry Clin Neurosci, 2020. **270**(7): p. 793-802.
72. Juchnowicz, D., et al., *The usefulness of a complete blood count in the prediction of the first episode of schizophrenia diagnosis and its relationship with oxidative stress.* PLoS One, 2023. **18**(10): p. e0292756.
73. Howes, O.D. and R. McCutcheon, *Inflammation and the neural diathesis-stress hypothesis of schizophrenia: a reconceptualization.* Transl Psychiatry, 2017. **7**(2): p. e1024.
74. Klawonn, A.M., et al., *Microglial activation elicits a negative affective state through prostaglandin-mediated modulation of striatal neurons.* Immunity, 2021. **54**(2): p. 225-234.e6.
75. Chung, H.Y., et al., *Microglia mediate neurocognitive deficits by eliminating C1q-tagged synapses in sepsis-associated encephalopathy.* Sci Adv, 2023. **9**(21): p. eabq7806.
76. Zhang, J., et al., *IL4-driven microglia modulate stress resilience through BDNF-dependent neurogenesis.* Sci Adv, 2021. **7**(12).
77. Fusar-Poli, P., et al., *Heterogeneity of Psychosis Risk Within Individuals at Clinical High Risk: A Meta-analytical Stratification.* JAMA Psychiatry, 2016. **73**(2): p. 113-20.
78. Bjorklund, G., et al., *Immune dysfunction and neuroinflammation in autism spectrum disorder.* Acta neurobiologiae experimentalis, 2016. **76**(4): p. 257-268.
79. Ferencova, N., et al., *Peripheral inflammatory markers in autism spectrum disorder and attention deficit/hyperactivity disorder at adolescent age.* International journal of molecular sciences, 2023. **24**(14): p. 11710.
80. Masi, A., et al., *Cytokine aberrations in autism spectrum disorder: a systematic review and meta-analysis.* Mol Psychiatry, 2015. **20**(4): p. 440-6.

81. Morgan, J.T., et al., *Microglial activation and increased microglial density observed in the dorsolateral prefrontal cortex in autism.* Biological psychiatry, 2010. **68**(4): p. 368-376.
82. Blaylock, R. and A. Strunecka, *Immune-glutamatergic dysfunction as a central mechanism of the autism spectrum disorders.* Current medicinal chemistry, 2009. **16**(2): p. 157-170.
83. Bennett, M., et al., *Establishing social inclusion the autism way: denying the "They Don't Want Friends" myth*, in *Life on the autism spectrum: Translating myths and misconceptions into positive futures*. 2019, Springer. p. 173-193.
84. Black, M.H., et al., *Multi-informant international perspectives on the facilitators and barriers to employment for autistic adults.* Autism Research, 2020. **13**(7): p. 1195-1214.
85. Bortoletto, R., et al., *Risk of psychosis in autism spectrum disorder individuals exposed to psychosocial stressors: A 9-year chart review study.* Autism Research, 2023. **16**(11): p. 2139-2149.
86. Hollocks, M.J., et al., *Anxiety and depression in adults with autism spectrum disorder: a systematic review and meta-analysis.* Psychol Med, 2019. **49**(4): p. 559-572.
87. O'Nions, E., et al., *Diagnosis of common health conditions among autistic adults in the UK: evidence from a matched cohort study.* The Lancet Regional Health - Europe, 2024. **41**: p. 100907.
88. Nimmo-Smith, V., et al., *Anxiety Disorders in Adults with Autism Spectrum Disorder: A Population-Based Study.* J Autism Dev Disord, 2020. **50**(1): p. 308-318.
89. Joshi, G., et al., *Psychiatric comorbidity and functioning in a clinically referred population of adults with autism spectrum disorders: a comparative study.* J Autism Dev Disord, 2013. **43**(6): p. 1314-25.
90. Joshi, G., et al., *The heavy burden of psychiatric comorbidity in youth with autism spectrum disorders: a large comparative study of a psychiatrically referred population.* J Autism Dev Disord, 2010. **40**(11): p. 1361-70.
91. Pehlivanidis, A., et al., *Lifetime co-occurring psychiatric disorders in newly diagnosed adults with attention deficit hyperactivity disorder (ADHD) or/and autism spectrum disorder (ASD).* BMC Psychiatry, 2020. **20**(1): p. 423.
92. Colizzi, M., et al., *Lessons Learnt From Running a Transition-Age Youth Mental Health Outpatient Clinic in Italy: The PRecocity of Intervention in Adolescent Medicine (PRIMA) Experience.* Early Interv Psychiatry, 2024.
93. Odalovic, M., et al., *Psychotropic medicines' prevalence, patterns and effects on cognitive and physical function in older adults with intellectual disability in Ireland: longitudinal cohort study, 2009-2020.* BJPsych Open, 2024. **10**(2): p. e39.

94. Alfageh, B.H., et al., *Psychotropic Medication Prescribing for Neuropsychiatric Comorbidities in Individuals Diagnosed with Autism Spectrum Disorder (ASD) in the UK.* J Autism Dev Disord, 2020. **50**(2): p. 625-633.
95. Houghton, R., C. Liu, and F. Bolognani, *Psychiatric Comorbidities and Psychotropic Medication Use in Autism: A Matched Cohort Study with ADHD and General Population Comparator Groups in the United Kingdom.* Autism Res, 2018. **11**(12): p. 1690-1700.
96. Jobski, K., et al., *Use of psychotropic drugs in patients with autism spectrum disorders: a systematic review.* Acta Psychiatr Scand, 2017. **135**(1): p. 8-28.
97. Deb, S., et al., *Randomised controlled trials of antidepressant and anti-anxiety medications for people with autism spectrum disorder: systematic review and meta-analysis.* Bjpsych Open, 2021. **7**(6).
98. Curnow, E., et al., *Mental health in autistic adults: A rapid review of prevalence of psychiatric disorders and umbrella review of the effectiveness of interventions within a neurodiversity informed perspective.* PLoS One, 2023. **18**(7): p. e0288275.
99. Bortoletto, R., et al., *Assessing the biobehavioral effects of ultramicronized-palmitoylethanolamide monotherapy in autistic adults with different severity levels: a report of two cases.* Frontiers in Psychiatry, 2024. **Volume 15 - 2024**.
100. Bortoletto, R., et al., *The Supplementation Therapy in Autism and Response to Treatment (START) Study: An Open-Label Feasibility Trial of Ultramicronized Palmitoylethanolamide Potential to Alleviate Psychic Distress among Autistic Adults.* Clinical and Translational Neuroscience, 2024. **8**(2).
101. Parums, D.V., *Editorial: Review Articles, Systematic Reviews, Meta-Analysis, and the Updated Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 Guidelines.* Med Sci Monit, 2021. **27**: p. e934475.
102. Salazar de Pablo, G., et al., *Prevalence of Individuals at Clinical High-Risk of Psychosis in the General Population and Clinical Samples: Systematic Review and Meta-Analysis.* Brain Sci, 2021. **11**(11).
103. Ising, H.K., et al., *The validity of the 16-item version of the Prodromal Questionnaire (PQ-16) to screen for ultra high risk of developing psychosis in the general help-seeking population.* Schizophr Bull, 2012. **38**(6): p. 1288-96.
104. Tognin, S., et al., *Association between Adverse Childhood Experiences and long-term outcomes in people at Clinical High-Risk for Psychosis.* Schizophrenia (Heidelb), 2025. **11**(1): p. 23.

105. Birnbaum, M.L., et al., *Factor structure of the Cannabis Experiences Questionnaire in a first-episode psychosis sample.* Early Interv Psychiatry, 2019. **13**(3): p. 495-501.
106. Nelson, B., et al., *Long-term follow-up of a group at ultra high risk ("prodromal") for psychosis: the PACE 400 study.* JAMA Psychiatry, 2013. **70**(8): p. 793-802.
107. Minichino, A., et al., *Endocannabinoid system mediates the association between gut-microbial diversity and anhedonia/amotivation in a general population cohort.* Mol Psychiatry, 2021. **26**(11): p. 6269-6276.

APPENDIX

Appendix A – Supplementary material for Chapter II

Supplementary Table 1. Eligibility criteria of Randomized Controlled Trials investigating Palmitoylethanolamide supplementation across human diseases.

Study ID	Inclusion criteria	Exclusion criteria
Abedini et al., 2022 (Iran) [1]	1. Inpatients 2. 18 to 54 years of age 3. Moderate to severe manic episode (YMRS ≥ 20)	1. IQ < 70 2. Allergy to lth, rsp, or PEA 3. Substance dependence (except nicotine and caffeine) 4. Ongoing manic-inducing medications 5. Current metabolic disorders 6. Current severe hepatic disease
Albanese et al., 2022 (Italy) [2]	1. 18 to 80 years of age 2. Past 24 h confirmed virological COVID-19 with no symptoms or mild-to-moderate infection	1. Pregnant or breastfeeding women 2. Severe respiratory failure requiring invasive mechanic ventilation 3. Allergy or hypersensitivity to PEA or to ≥ 1 of its excipients
Andresen et al., 2016 (Denmark, Norway, United Kingdom) [3]	1. ≥ 18 years of age 2. ≥ 6 month-lasting SCI 3. ≥ 3 month-lasting at- and/or below-level SCI neuropathic pain 4. 4 ≤ average pain intensity ≤ 9, during a 7-day baseline period	1. Concomitant severe cerebral damage, terminal illness 2. Planned surgery 3. Alcohol, or substance abuse 4. Hypersensitivity to umPEA or its excipients 5. Current psychiatric disease (except reactive depression) 6. Pregnant or breastfeeding women, insufficient contraceptives 7. Impossibility to differentiate neuropathic SCI pain from other types of pain
Bacci et al., 2011 (Italy) [4]	1. 18 to 30 years of age 2. No signs of inflammation or tooth decay 3. Lower third molars identical in position, orientation, depth, and class of impaction	1. Patients needing monolateral extractions 2. Past 2 months other clinical studies involvement 3. Systemic diseases and taking chronic pharmacotherapy 4. Unlikelihood of compliance 5. Allergies to NSAIDs 6. Impossibility to take NSAIDs
Bonzanino et al., 2024 (Italy) [5]	1. ≥ 18 and ≤ 90 years of age 2. 5 ≤ NIHSS score ≤ 20 3. No significant pre-stroke disabilities	1. Hemorrhagic stroke 2. Concomitant diseases interfering with evaluation
Briskey et al., 2023 (Australia) [6]	1. ≥ 18 years of age 2. Good general health	1. Using any allergy relief supplements or products during the study 2. Pregnant or breastfeeding women, insufficient contraceptives

	3. Providing written informed consent 4. Experiencing seasonal allergic rhinitis	
Briskey et al., 2024 (Australia) [7]	1. ≥ 18 years of age 2. Good general health 3. At least five attacks fulfilling ICHD3 criteria B-D 4. Headache attacks lasting 4–72 h (untreated or unsuccessfully treated) 5. Headaches having ≥ 2/4 following: (a) unilateral location, (b) pulsating quality, (c) moderate or severe pain intensity, (d) aggravated by or causing avoidance of routine physical activity 6. During headaches, ≥ 1 following: (a) nausea and/or vomiting, (b) photophobia, (c) phonophobia 7. Migraine not better accounted for by another ICHD3 diagnosis	1. Chronic past and/or current alcohol use (>14 alcoholic drinks per week) 2. Using long-term medication (unless for controlled medical conditions as above) 3. Pregnant or breastfeeding women, insufficient contraceptives 4. Allergic, or hypersensitive to any of the ingredients in the active or placebo formula 5. Used preventative medication 6. Migraines to occur on ≥ 15 days of the month for > 3 months, which, on ≥ 8 days per month, had the features of a migraine headache, a debilitating attack lasting > 72 h, or a seizure
Campolo et al., 2021 (Italy) [8]	1. All consecutive admitted patients 2. Moderate TBI (GCS 9-13)	X
Cantone et al., 2024 (Italy) [9]	1. 18 to 60 years of age 2. Confirmed history of COVID-19 (positive nasopharyngeal swab for SARS-CoV-2) 3. Smell disturbances resulting from the COVID-19 episode 4. Persistent olfactory disfunction at time of recruitment (smell loss and parosmia)	1. Known neuroinflammatory or neurodegenerative diseases 2. Stroke or head trauma within the past 5 years 3. Prior or current chemo-radiotherapy 4. Prior or current sinonasal cancer or chronic rhinosinusitis 5. Prior or current nasal allergy 6. Prior or current nasal polyps 7. Anatomical conditions reducing nasal airflow 8. Ongoing medications interacting with olfactory function 9. Current severe psychiatric disorders.
Cobellis et al., 2011 (Italy) [10]	EMS diagnosis	X
Coraci et al., 2018 (Italy) [11]	1. Minimum/mild idiopathic CTS (Padua's classification) 2. Providing written informed consent	1. Other neuropathies 2. Orthopedic, endocrinological and rheumatological diseases
Costagliola et al., 2014 (Italy) [12]	1. NTG diagnosis (optic disc abnormalities consistent with glaucomatous optic neuropathy with visual field loss) 2. IOP ≤ 18 mmHg without topical hypotensive therapy	1. Active ocular disease 2. Use of other ocular medications or therapies that might have a substantial effect on IOP 3. History of ocular surgery

	3. ≥ 18 years of age 4. logMAR BCVA ≥ 0.70 5. Progression of anatomic and functional damage as determined by computerized perimetry and serial stereophotographs of the optic nerve head 6. No history of amblyopia 7. No history of ocular disease or previous eye surgery	4. Use of other similar systemic medications 5. Vasoactive systemic therapies (e.g., Ca-antagonists, oral beta-blocker, etc.) 6. Current tobacco smoker 7. Pregnant or breastfeeding women
Cremon et al., 2016 (Italy, Spain, France, Croatia, Bosnia) [13]	1. All IBS subtypes (IBS-C, IBS-D, IBS-M) 2. 18 to 70 years of age 3. Previous 5 years negative colonoscopy or barium enema examination 4. Negative relevant additional screening or consultation	1. Pregnant or breastfeeding women, insufficient contraceptives 2. Current use of NSAIDs, corticosteroids, and mast cell stabilizers 3. Past month use of topical or systemic antibiotics 4. Continuous use of stimulant laxatives 5. Major abdominal surgery 6. Medical history, appropriate consultations, and laboratory tests confirmation of IBD, infectious diarrhoea or diverticular disease, coeliac disease, allergic diseases (including asthma), other organic or psychiatric disorders 7. Impossibility to take NSAIDs
D'Ascanio et al., 2021 (Italy) [14]	1. Outpatients 2. 18 to 90 years of age 3. Confirmed history of COVID-19 (positive nasopharyngeal swab for SARS-CoV-2) 4. Confirmed anosmia/hyposmia (Sniffin' Sticks psychophysical test, TDI < 31) persisting ≥ 90 days after subsequent negative COVID-19 nasopharyngeal swab	1. History of olfactory-gustatory disorders 2. Impaired cognitive function 3. History of NDD (AD and PDis) 4. Medical therapy with known detrimental effects on olfactory function 5. Active rhinological disorders 6. History or chemo-radiotherapy of the head/neck region 7. History of stroke or neurotrauma 8. Severe nasal blockage from stenosis or deformity 9. Severe psychiatric illness 10. Previous sinonasal or nasopharyngeal tumors
Di Nardo et al., 2024 (Italy) [15]	1. 10 to 17 years of age 2. All IBS subtypes (IBS-C, IBS-D, IBS-M) 3. Negative findings for fecal calprotectin and anti-transglutaminase antibodies	1. Current use of NSAIDs, corticosteroids, and mast cells stabilizers 2. Past month use of topical or systemic antibiotics 3. Continuous use of stimulant laxatives 4. Major abdominal surgery 5. Medical history, appropriate consultations, and laboratory tests confirmation of IBD, infectious diarrhoea, allergic diseases,

		other organic or psychiatric disorders
Di Stadio et al., 2022 (Italy) [16]	1. Outpatients 2. 18 to 65 years of age 3. ≥ 180 days olfactory impairment post negative COVID-19 nasopharyngeal swab	1. History of olfactory-gustatory disorders 2. Active chemotherapy or estrogen inhibitors-therapy (aromatase) 3. Impaired cognitive function 4. History of NDD 5. Medical therapy with detrimental effects on olfactory function 6. Active rhinological disorders 7. History or chemo-radiotherapy of the head/neck region 8. History of stroke or head trauma 9. Severe nasal blockage from stenosis or deformity 10. Severe psychiatric illness 11. History of sinonasal or nasopharyngeal tumors 12. Past 30 days corticosteroid therapy to treat olfactory dysfunction 13. Current anti-inflammatory or immunomodulant therapy
Di Stadio et al., 2023a (Italy) [17]	1. Outpatients 2. 18 to 65 years of age 3. ≥ 180 days olfactory impairment post negative COVID-19 nasopharyngeal swab	1. History of olfactory-gustatory disorders 2. Active chemotherapy or estrogen inhibitors-therapy (aromatase) 3. Impaired cognitive function 4. History of NDD 5. Active NDD/neuroinflammatory diseases 6. Medical therapy with detrimental effects on olfactory function 7. Active rhinological disorders 8. History or chemo-radiotherapy of the head and neck region 9. History of removal of sinonasal or nasopharyngeal tumors 10. < 3 years stroke or moderate/severe head trauma, nasal septal deviation, or turbinate hypertrophy 11. Previous 30 days corticosteroid therapy to treat olfactory dysfunction 12. Current anti-inflammatory or immunomodulant therapy
Di Stadio et al., 2023b (Italy) [18]	1. Outpatients 2. 18 to 60 years of age 3. ≥ 180 days olfactory impairment post negative COVID-19 nasopharyngeal swab	1. History of olfactory-gustatory disorders 2. Active chemotherapy or estrogen inhibitors-therapy (aromatase) 3. Impaired cognitive function 4. History of NDD 5. Medical therapy with detrimental effects on olfactory function 6. Active rhinological disorders

		7. History or chemo-radiotherapy of the head and neck region 8. History of stroke or head trauma 9. Severe nasal blockage from stenosis or deformity 10. Severe psychiatric illness 11. History of sinonasal or nasopharyngeal tumors 12. Previous 30 days corticosteroid therapy to treat olfactory dysfunctic 13. Current anti-inflammatory or immunomodulant therapy
Evangelista et al., 2018 (Italy) [19]	1. ≥ 18 years of age 2. ≥ 6 months painful symptoms 3. Sleep disturbance due to nocturnal numbness and/or tingling and sustained arm or hand pain and mitigated by change in posture 4. PSQI score ≥ 8 5. 5 ≤ NRS ≤ 9 6. Never surgically treated unilateral CTS	1. Concomitant clinical conditions interfering with CTS painful symptoms evaluation 2. Previous surgically treated unilateral CTS
Faig-Marti and Martìnez-Catassùs, 2017 (Spain) [20] Faig-Martì and Martìnez-Catassùs, 2020 (Spain) [21]	1. 18 to 75 years of age 2. ≥ 3 months mild to moderate CTS (ENG)	1. History of previous upper extremity surgery 2. Active treatment with steroids 3. Use of night splinting 4. Food allergies
Gagliano et al., 2011 (Italy) [22]	1. ≥ 18 years of age 2. POAG/OH diagnosis 3. ≥ 3 months timolol 0.5% eyedrops twice daily 4. 19 mmHg < IOP < 24 mmHg 5. Able to perform a reliable visual field (a minimum of 3 tests) 6. < 0.6 C/D ratio	1. Need for glaucoma surgical or laser therapy 2. Past year ocular surgery 3. No tolerability to product under evaluation 4. Visual acuity 8/10 with refractive error 3 diopters 5. Pupillary diameter 2.5 mm 6. Concomitant systemic or ocular pathologies 7. Pregnant or breastfeeding women 8. Vasoactive systemic therapies 9. Current tobacco smoker
Germini et al., 2017 (Italy) [23]	1. ≥ 65 years of age 2. Back, joints, or limbs chronic pain for ≥ 6 months	1. Chronic cancer pain 2. Subacute or chronic ischemic pain 3. Recently started new pharmacological or non-pharmacological treatment for pain

Ghazizadeh-Hashemi et al., 2018 (Iran) [24]	1. 18 to 50 years of age 2. HDRS Total ≥ 19; HDRS Item 1 ≥ 2	1. Past month antidepressant and psychotherapy 2. Past 2 months ECT 3. Psychosis or other mental disorders diagnosis 4. Alcohol or substance abuse or dependence (except nicotine) 5. Risk of suicide or suicidal ideation 6. Any uncontrolled medical problem
Giammusso et al., 2017 (Italy) [25]	1. ≥ 6 months complaints of chronic pelvic pain 2. IPSS score > 13 at visit 1 3. Pain domain of NIH-CPSI > 1 at visit 1 4. Total PSA < 4 ng/ml 5. Sign and symptoms of category III CP/CPPS (NIH)	1. Subjects < 22 and > 61 years of age 2. Major comorbidities 3. Known anatomical abnormalities of the urinary tract 4. Evidence of other urological diseases 5. Residual urine volume > 50 ml resulting from bladder outlet obstruct
Guida et al., 2010 (Italy) [26]	1. 18 to 75 years of age 2. VAS ≥ 5	1. Unconfirmed lumbosciatalgia diagnosis at baseline 2. Pregnant women 3. Concomitant use of medications causing drug-induced peripheral neuropathies 4. Comorbidities interfering with efficacy assessment 5. Past 4 weeks participation to a clinical study 6. Unlikelihood of compliance
Isola et al, 2021 (Italy) [27]	1. Good general health 2. ≥ 6 teeth per quadrant 3. PD ≥ 4 mm and CAL ≥ 2 mm in ≥ 40% of the analyzed sites 4. ≥ 2 teeth in each quadrant with PD ≥ 5 mm 5. ≥ 40% sites with BOP 6. No furcation involvement	1. Past 6 months periodontal therapy 2. Past 6 months use of antibiotics, anti-inflammatory, or Immunosuppressant medication 3. Pregnant women 4. Any systemic condition which might affect the study 5. Past 3 months use of mouthwash containing antimicrobials 6. Use of hormonal contraceptives 7. Anti-inflammatory and immunosuppressive medications 8. History of excessive drinking 9. Current tobacco smoker 10. Class II and III tooth mobility
Kadanangode Narayanaswam et al., 2023 (India) [28]	1. 18 to 60 years of age 2. DAS28 score ≥ 3 3. Providing written informed consent 4. Ability to comprehend the nature and objectives of the study and demonstrate a willingness to adhere to study	1. Current use of NSAIDs/painkillers other than standard drug 2. Abnormal results on liver function test 3. Current diabetic neuropathy 4. Current severe renal, hepatic, cardiac, gastrointestinal, neurological, hematological, or respiratory disorder

	procedures	5. Current psychiatric disorder 6. BMI > 35 kg/m^2 or < 20 kg/m^2 7. Need for surgery during the study period 8. Past 12-week participation in any clinical study or trial 9. Known hypersensitivity to the study drugs 10. Patients with severe infection 11. History of intake of any ayurvedic/herbal/homeopathic/dietary supplements in the last two months 12. Pregnant or breastfeeding women, insufficient contraceptives
Khalaj et al., 2018 (Iran) [29]	1. 4 to 12 years of age 2. Irritability symptoms of at least moderate severity (scores ≥ 12 on the ABC-C Irritability subscale)	1. Symptoms not pronounced enough to be considered for treatment with rsp 2. Concomitant psychiatric disorder 3. Preexisting medical condition 4. Severe intellectual disability 5. Alcohol/drug abuse 6. Dyskinesia 7. Past 6 months AP medication or behavior treatment
Lunardelli et al., 2019 (Italy) [30]	1. > 75 years of age 2. Proximal hip fracture eligible for early surgery	1. Severe dementia 2. Delirium at the time of admission 3. Patients receiving AP medications 4. Surgery cannot be performed within 48 h from admission
Marini et al., 2013 (Italy) [31]	Presence of arthralgia (TMJ pain at rest and during function, and on palpation, and crepitus) or OA (TMJ pain at rest and during function, and on palpation)	1. Presence of myogenic pain 2. Presence of musculoskeletal pain 3. Co-occurring depressive disorders 4. Presence of odontogenic pain 5. Pregnant women 6. Malignancy 7. Systemic rheumatologic diseases
Masek et al., 1974 (Czech Republic) [32]	X	X
Murina et al., 2013 (Italy) [33]	X	X
Orefice et al., 2016 (Italy) [34]	1. 18 to 55 years of age 2. < 1 year disease duration	1. MS-related conditions, such as current relapse 2. Past 30 days steroid therapy

	3. 1.0 < EDSS < 3.5 4. ≥ 6 months IFN-β1a treatment 5. Experiencing IFN-β1a-related AEs	3. Concomitant diseases precluding IFN-β1a 4. Pregnant or breastfeeding women 5. Cognitive decline 6. Pathological conditions interfering MS evolution 7. Allergy to NSAIDs 8. IFN-β1a intolerance
Ottaviani et al., 2019 (Italy) [35]	1. 35 to 80 years of age 2. Non-smokers 3. NRS > 4	1. Signs or symptoms of injury or detectable pathologies in the oral cavity 2. Periodontal disease 3. Past 3 months pain medications (anti-inflammatory or therapy inducing xerostomia)
Palma et al., 2016 (Italy) [36]	1. ≥ 18 years of age 2. ALSFRS-R score > 20 3. FVC > 30% 4. Treatment with rlz	1. Other diseases of motor neurons 2. Participation to experimental treatments in the previous 3 months 3. Pregnant or breastfeeding women 4. Contraindications to the use of rlz 5. Patients undergoing tracheostomy, enteral or parenteral supply 6. Severe psychiatric disorders
Pickering et al., 2022 (Australia) [37]	1. ≥ 18 years of age 2. Type 1 or type 2 diabetes with PNP 3. DN4 > 4 4. S-LANSS > 12 5. Ongoing anti-diabetic medications	1. PNP due to hereditary sensory neuropathy, vitamin B12 or folate deficiency, paraneoplastic diseases, advanced liver disease, kidney disease, hypothyroidism, prolonged phenytoin, warfarin, or immunosuppressive drug use 2. Ongoing herbal medications for pain relief 3. Pregnant or breastfeeding women, insufficient contraceptives 4. Alcohol or substance abuse 5. Allergy or sensitivity to PEA or any ingredients 6. Any clinically relevant abnormal findings

Rao et al., 2021 (Australia) [38]	1. ≥ 18 years of age 2. PSQI > 5 3. Males and females 4. Females with child-bearing potential on a prescribed form of birth control 5. Agree not to change current diet or exercise or use other supplements for sleep disturbances for the study period	1. Unstable or serious illness 2. Past 2 years malignancy or treatment for malignancy 3. Clinically significant inflammation connective tissue disease or arthr 4. Current mood disorders 5. Current neurological disorders 6. OTC sleep medication or aid during the trial 7. Diagnosed sleep apnea 8. Diagnosed or consistent gastrointestinal issues disrupting sleep 9. Active smoking or nicotine, prescribed drug, or illegal substances intake 10. Chronic past and/or present alcohol use (>14 alcoholic drinks per week) 11. Regular stimulants intake (e.g. coffee, caffeine supplements, or caffeine containing beverages) from midday onwards 12. Diagnosed clinical sleep disorder 13. Night-shift employment or other situations leaving one unable to have a normal night's sleep 14. Disturbed sleeping patterns caused by external factors (e.g. children, partner, noise) 15. Allergic to the PEA or placebo formula 16. Pregnant or breastfeeding women or any condition or non-medicated supplement making the participant unsuitable for inclusion

Rao et al., 2023 (Australia) [39]	1. Providing written informed consent 2. Patients agreeing not to take other supplements or medications aimed at preventing URTIs for the duration of the trial	1. Cognitive damage 2. Serious mood disorders or neurological disorders 3. Unstable or serious illness 4. Acute sickness in the previous 2 months 5. Active smokers or nicotine/drugs abusers 6. Chronic alcohol use (>14 alcoholic drinks per week) 7. Allergy to any of the ingredients in the active or placebo formula 8. Pregnant or breastfeeding women 9. Medically prescribed medications that could affect the immune and/or inflammatory response 10. Participated in a related clinical trial in the 1 month prior 11. Treatment for cancer, HIV, or the chronic use of steroids in the past year
Rao et al., 2024 (Australia) [40]	1. ≥ 18 years of age 2. Atopic dermatitis with redness, dry skin, scaling, and/or itchiness on their hands or arm 3. Providing written informed consent	1. Active allergic skin responses 2. Pregnant or breastfeeding women 3. Current tobacco smoker 4. Chronic past and/or current alcohol use 5. Unstable or serious illness including renal, hepatic, gastro-intestinal, cardiovascular, diabetes, mood disorders, cancer, 6. Using or used immunosuppressive medication within the last 3 months 7. Allergies to any of the ingredients in the Levagen+ or comparator formula
Rossi et al., 2020 (Italy) [41]	1. ≥ 18 years of age 2. Diagnosis of POAG/NTG 3. Controlled IOP (< 18 mmHg) with prostaglandin analogues monotherapy 4. Past 2 years stable IOP < 18 mmHg 5. Past 2 years stable disease (≤ -1 dB/year at MD of visual field)	1. Ocular hypertension with normal optic nerve and visual field 2. Contraindication to PEA 3. Brimonidine eye drops use 4. Limiting conditions 5. Other causes of visual field changes

	6. Past 6 months no ocular surgery	
Salaffi et al., 2023 (Italy) [42]	1. Providing written informed consent 2. 12-week dlx and pgb treatment	Other ongoing medications (except dlx and pgb)
Salehi et al., 2022 (Iran) [43]	1. 18 to 60 years of age 2. Illness duration ≥ 2 years 3. PANSS negative ≥ 15 4. HDRS < 14 5. Clinical stability on rsp (PANSS total change ≤ 20% on 2 subsequent assessments within 2 weeks)	1. IQ < 70 2. History of head trauma 3. Past 3 months history of ECT 4. Past 6 months substance or alcohol dependence 5. Pregnant or breastfeeding women 6. Suicidal ideation 7. History of neurosurgery 8. Current acute or chronic medical disease 9. History of allergy to rsp or PEA
Steels et al., 2018 (Australia) [44]	1. NRS ≥ 4 2. Abstaining from any knee OA continuative medications	1. Other forms of arthritis 2. Past 6 months joint injury 3. Past 30 days use of knee OA medications 4. BMI > 35 5. Pregnant or breastfeeding women 6. Uncontrolled diabetes 7. High cholesterol 8. Hypertension 9. Receiving anti-coagulation therapy 10. History of cerebrovascular accidents, stroke, or transient ischemia 11. MDD 12. Past 6 months unintended weight loss of > 15% body weight 13. Active substance abuse
Strobbe et al., 2013 (Italy) [45]	1. Baseline IOP ≥ 22 mmHg for ≥ two measurements 2. Open anterior chamber angle at gonioscopy 3. C/D ratio < 0.4 4. Normal visual field parameters (MD < 3 dB and PSD < 2.5 dB) 5. Normal Glaucoma Hemifield Test 6. Normal corneal central thickness (530–560 lm)	1. Cardiovascular disease or known cardiovascular risk factors 2. Taking vasoactive medications
Tartaglia et al., 2015 (Italy) [46]	1. Untreated patients 2. 16 to 24 years of age	1. Secondary dysmenorrhea 2. Chronic pelvic pain

Versace et al., 2023 (Italy) [47]	1. Previous SARS-CoV-2 infection diagnosis (PCR testing of nasopharyngeal swab) 2. Subsequent infection recovery (2 consecutive negative PCR tests separated by ≥ 1 day) 3. Mild form of COVID-19, not necessitating inpatient admission 4. Complaints of fatigue and/or cognitive difficulties persisting after COVID-19 (FSS > 36, PCDS ≥ 1)	1. Prior or concurrent neurological, psychiatric, endocrine, metabolic, or cardiopulmonary conditions 2. Clinical and/or radiological evidence of active COVID-19 related pneumonia 3. Anemia 4. Ongoing treatment with corticosteroids, antihistamines, antihypertensives, diuretics, antidepressants, anxiolytics, hypnotics
Yuan et al., 2014 (China) [48]	1. Mild to moderate Asteatotic eczema on the lower leg 2. Mild to moderate erythema, scaling, or dryness	1. Active psoriasis or a history of psoriasis 2. Active allergic skin responses 3. Severe eczema

<, less/smaller than; >, more/greater than; ≤, less/smaller than or equal to; ≥, more/greater than or equal to; ABC-C, Aberrant Behavior Checklist-Community; AD, Alzheimer's disease; AEs, Adverse Events; ALSFRS-R, ALS Functional Rating Scale-Revised; AP, Antipsychotic; BCVA, Best Corrected Visual Acuity; BMI, Body Mass Index; BOP, Bleeding on probing; C/D, Cup-to-disc; CAL, Clinical attachment level; COVID-19, SARS-CoV-2 infection; CP/CPPS, Chronic Prostatitis/Chronic Pelvic Pain syndrome; CPSI, Chronic Prostatitis Symptom Index; CTS, Carpal Tunnel Syndrome; DAS, Disease Activity Score; dlx, Duloxetine; DN4, Neuropathic pain diagnostic questionnaire; ECT, Electroconvulsive Therapy; EDSS, Expanded Disability Status Scale; EMS, Endometriosis; ENG, Electroneurography; FSS, Fatigue Severity Scale; FVC, Forced Vital Capacity; GCS, Glasgow Coma Scale; h, hours; HDRS, Hamilton Depression Rating Scale; IBD, Inflammatory Bowel Disease; IBS, Irritable Bowel Syndrome; IBS-C, IBS with predominant constipation; IBS-D, IBS with predominant diarrhoea; IBS-M, IBS with mixed symptoms; ICHD, The International Classification of Headache Disorders; IFN-β1a, Interferon-Beta1a; IOP, Intra-ocular pressure; IQ, Intelligence quotient; logMAR, Logarithm of the Minimal Angle of Resolution; lth, Lithium; MD, Mean Deviation; MDD, Major Depressive Disorder; NDD, Neurodegenerative Disorder; NIH, National Institutes of Health; NIHSS, National Institutes of Health Stroke Scale; NRS, Numeric Pain Rating Scale; NSAIDs, Nonsteroidal anti-inflammatory drugs; NTG, Normal Tension Glaucoma; OA, Osteoarthritis; OH, Ocular hypertension; OTC, Over-the-counter; PANSS, Positive and Negative Syndrome Scale; PCDS, Perceived Cognitive Difficulties Scale; PCR, Polymerase Chain Reaction; PD, Probing depth; PDis, Parkinson's disease; PEA, Palmitoylethanolamide; pgb, Pregabalin; POAG, Primary open glaucoma; PSD, Pattern standard deviation; PSQI, Pittsburgh Sleep Quality Index; rlz, Riluzole; rsp, Risperidone; S-LANSS, Self-reported Leeds Assessment of Neuropathic Symptoms and Signs; SCI, Spinal Cord Injury; TBI, Traumatic Brain Injury; TDI, Threshold, Discrimination and Identification subtests; TMJ, Temporo-mandibular joint; umPEA, Ultramicronized-PEA; URTI, Upper Respiratory Tract Infection; VAS, Visual Analogue Scale; YMRS, Young Mania Rating Scale.

Appendix B – Supplementary material for Chapter V

Supplementary Table 1. Results of repeated measures analyses for longitudinal score progression across assessments (parametric, non-parametric, and ES are reported). Scores were assessed at baseline (beginning of PEA administration), at 4 weeks, and at 12 weeks. Results are reported for both clinical measures (i.e., CAARMS, HADS, and GF) and for all biomarkers under consideration.

Measure	**Score**	**LME** F (p)	**Friedman test** χ^2, p	**ES** Cohen f
CAARMS-P1, Total	[4×GRS]	13.97 (<0.001*)	11.25 (0.004*)	0.484§
CAARMS-P1.1, UTC	[GRS]	11.38 (<0.001*)	14.30 (<0.001*)	0.554§
CAARMS-P1.2, NPI	[GRS]	5.55 (0.009*)	7.35 (0.025*)	0.336
CAARMS-P1.3, PA	[GRS]	1.84 (0.176)	3.11 (0.212)	0.212
CAARMS-P1.4, DS	[GRS]	1.88 (0.169)	6.33 (0.042*)	0.102
HADS-A, Anxiety scale	[0, 28]	10.21 (<0.001*)	9.25 (0.010*)	0.380
HADS-D, Depression scale	[0, 28]	4.68 (0.017*)	4.51 (0.105)	0 .252
GF-S, Social scale	[0, 10]	4.81 (0.015*)	7.94 (0.019*)	0.160
GF-R, Role scale	[0, 10]	1.36 (0.272)	3.81 (0.149)	0.067
Basophils	[×10³/µl]	0.11 (0.892)	0.91 (0.633)	0.057
CRP	[mg/L]	1.52 (0.234)	1.00 (0.607)	0.178
CAR	[nMol/L]	0.10 (0.904)	0.13 (0.936)	0.031
Eosinophils	[×10³/µl]	0.68 (0.515)	1.66 (0.437)	0.078
ESR	[mm/h]	0.17 (0.841)	0.52 (0.771)	0.042
IFN-γ	[pg/mL]	1.99 (0.155)	1.37 (0.504)	0.277
IP10	[pg/mL]	0.17 (0.841)	0.13 (0.936)	0.082
IL-1β	[pg/mL]	1.24 (0.304)	2.50 (0.287)	0.208
IL-2Rα	[pg/mL]	0.95 (0.400)	9.73 (0.008*)	0.109
IL-6	[pg/mL]	0.75 (0.483)	0.24 (0.887)	0.147
IL-8	[pg/mL]	3.72 (0.036*)	5.73 (0.057)	0.267
IL-10	[pg/mL]	0.07 (0.934)	1.93 (0.381)	0.043
IL-17A	[pg/mL]	0.21 (0.816)	0.58 (0.747)	0.087

Measure	**Score**	**LME** F (p)	**Friedman test** χ^2, p	**ES** Cohen f
Monocytes/Lymph	[ratio]	0.47 (0.628)	1.73 (0.420)	0.091
Neutrophils/Lymph	[ratio]	0.24 (0.789)	0.53 (0.766)	0.074
Platelets/Lymph	[ratio]	2.28 (0.119)	0.93 (0.627)	0.117
SII	[ratio]	0.62 (0.546)	0.93 (0.627)	0.101
TNF α	[pg/mL]	0.11 (0.900)	1.66 (0.436)	0.039
WBC	[$\times 10^3/\mu l$]	0.39 (0.683)	0.53 (0.766)	0.096
IL-10/IL-6	[ratio]	1.65 (0.210)	2.68 (0.262)	0.191

CAARMS: Comprehensive Assessment of At-Risk Mental States; CAR: Cortisol Awakening Response; CRP: C-reactive protein; DS: Disorganised Speech (CAARMS-P1.4); ES: Effect-size; ESR: Erythrocyte Sedimentation Rate; GF: Global Functioning; GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); HADS: Hospital Anxiety and Depression Scale; IFN-γ: Interferon-γ; IL: Interleukin; IP10: IFN-γ-induced protein-10; LME: Linear Mixed Effect model; Lymph: Lymphocyte; NBI: Non-Bizarre Ideas (CAARMS-P1.2); P1: CAARMS Positive Symptoms (UTC, NBI, PA, and DS); PA: Perceptual Abnormalities (CAARMS-P1.3); PEA: Palmitoylethanolamide (administered in its ultramicronized form); SII: Systemic Immune Inflammation index (Neutrophils×Platelets/Lymphocyte;); TNF α: Tumor Necrosis Factor; UTC: Unusual Thought Content (CAARMS-P1.1); WBC: White Blood Cell total count. *: Denotes a statistically significant effect of assessment time point ($p<0.050$). §: Denotes detectable ES, with adequate power (Cohen $f\geq0.401$).

Supplementary Table 2. Effect of median-split baseline biomarker in longitudinal score progression across assessments (Assessment main-effect, Biomarker main-effect, and Biomarker × Assessment interaction are reported). Changes in scores were assessed between baseline (beginning of PEA administration), 4 weeks, and 12 weeks (Assessment). Biomarker values were included as time-varying moderators (Biomarkers). The analyses pertain to: (**a**) CAARMS Positive Symptoms global rating and (**b**) CAARMS UTC severity.

S2.a. CAARMS Positive Symptoms global rating (UTC+NBI+PA+DS).

Baseline value	**LME** – F (p)		
≤Md vs >Md	**Assessment**	**Biomarker**	**Biomarker × Assessment**
Basophils	6.15 (0.006*)	0.45 (0.513)	0.01 (0.988)
CRP	8.39 (0.002*)	0.02 (0.878)	0.41 (0.666)
CAR	11.70 (<0.001*)	1.01 (0.331)	1.23 (0.307)
Eosinophils	6.00 (0.007*)	0.03 (0.860)	0.03 (0.972)
ESR	5.07 (0.013*)	0.38 (0.546)	1.35 (0.275)
IFN-γ	5.96 (0.007*)	4.88 (0.044*)	0.52 (0.598)
IP10	5.96 (0.007*)	0.25 (0.624)	0.35 (0.709)
IL-1β	1.81 (0.183)	0.00 (0.979)	1.88 (0.171)
IL-2Rα	6.51 (0.005*)	0.00 (0.977)	0.30 (0.744)
I IL-6	4.36 (0.024*)	0.16 (0.696)	0.96 (0.399)
IL-8	7.44 (0.003*)	6.01 (0.029*)	0.37 (0.693)
IL-10	7.37 (0.003*)	0.48 (0.499)	0.11 (0.898)
IL-17A	5.23 (0.012*)	0.20 (0.660)	0.56 (0.575)
Monocytes/Lymph	9.23 (<0.001*)	0.48 (0.499)	1.98 (0.157)
Neutrophils/Lymph	9.88 (<0.001*)	0.75 (0.402)	0.86 (0.433)
Platelets/Lymph	5.73 (0.008*)	0.06 (0.811)	0.06 (0.937)
SII	3.63 (0.039*)	0.75 (0.402)	1.23 (0.308)
TNF α	4.03 (0.029*)	0.05 (0.827)	0.73 (0.491)
WBC	3.13 (0.060)	2.11 (0.169)	2.14 (0.136)
IL-10/IL-6	8.24 (0.002*)	0.58 (0.462)	2.78 (0.082)

S2.b. CAARMS Unusual Thought Content severity.

Baseline value	**LME** – F (p)		
≤Md vs >Md	**Assessment**	**Biomarker**	**Biomarker × Assessment**
Basophils	6.24 (0.006*)	0.00 (0.969)	0.66 (0.522)
CRP	4.56 (0.020*)	0.04 (0.838)	0.04 (0.963)
CAR	7.62 (0.002*)	0.39 (0.542)	0.48 (0.627)
Eosinophils	5.64 (0.009*)	0.68 (0.423)	0.74 (0.484)
ESR	5.74 (0.008*)	0.42 (0.526)	1.38 (0.269)
IFN-γ	5.60 (0.009*)	7.22 (0.018*)	2.05 (0.147)
IP10	5.55 (0.009*)	1.67 (0.217)	0.66 (0.522)
IL-1β	1.80 (0.184)	0.14 (0.716)	4.77 (0.017*)
IL-2Rα	4.90 (0.015*)	0.99 (0.338)	0.13 (0.875)
IL-6	3.54 (0.045*)	0.25 (0.625)	0.31 (0.734)
IL-8	3.92 (0.033)	3.34 (0.091)	0.13 (0.877)
IL-10	7.84 (0.002*)	0.50 (0.490)	1.85 (0.176)
IL-17A	4.57 (0.019*)	0.11 (0.748)	0.88 (0.426)
Monocytes/Lymph	7.12 (<0.003*)	1.83 (0.197)	0.83 (0.446)
Neutrophils/Lymph	8.67 (0.001*)	0.99 (0.338)	0.69 (0.511)
Platelets/Lymph	5.31 (0.011*)	0.00 (0.969)	0.45 (0.642)
SII	3.77 (0.036*)	0.50 (0.490)	2.02 (0.152)
TNF α	2.92 (0.070)	0.08 (0.785)	0.51 (0.606)
WBC	4.14 (0.027*)	1.10 (0.312)	1.76 (0.190)
IL-10/IL-6	6.36 (0.006*)	6.43 (0.026*)	0.33 (0.723)

CAARMS: Comprehensive Assessment of At-Risk Mental States; CAR: Cortisol Awakening Response; CRP: C-reactive protein; DS: Disorganised Speech (CAARMS-P1.4); ESR: Erythrocyte Sedimentation Rate; IFN-γ: Interferon-γ; IL: Interleukin; IP10: IFN-γ-induced protein-10; LME: Linear Mixed Effect model; Lymph: Lymphocyte; Md: Median; NBI: Non-Bizarre Ideas (CAARMS-P1.2); P1: CAARMS Positive Symptoms (UTC, NBI, PA, and DS); PA: Perceptual Abnormalities (CAARMS-P1.3); PEA: Palmitoylethanolamide (administered in its ultramicronized form); SII: Systemic Immune Inflammation index (Neutrophils×Platelets/Lymphocyte;); TNF α: Tumor Necrosis Factor; UTC: Unusual Thought Content (CAARMS-P1.1); WBC: White Blood Cell total count. *: Denotes a statistically significant effect of assessment time point (p<0.050).

Supplementary Table 3. Effect of time-varying biomarker in longitudinal score progression across assessments (Assessment main-effect, Biomarker main-effect, and Biomarker × Assessment interaction are reported). Changes in scores were assessed between baseline (beginning of PEA administration), 4 weeks, and 12 weeks (Assessment). The analyses pertain to: (**a**) CAARMS Positive Symptoms global rating and (**b**) CAARMS UTC severity.

S3.a. CAARMS Positive Symptoms global rating (UTC+NBI+PA+DS).

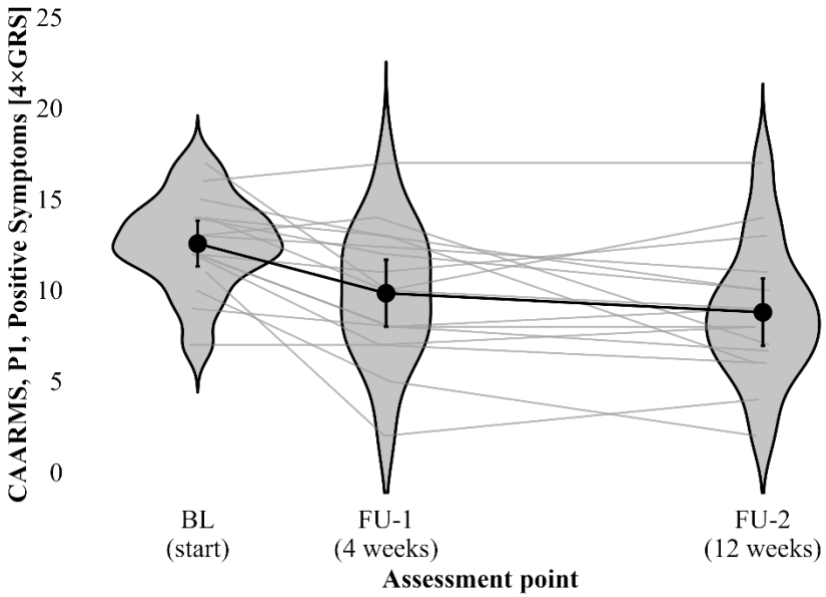
	LME – F (p)		
Covariate	**Assessment**	**Biomarker**	**Biomarker × Assessment**
Basophils	1.19 (0.320)	0.61 (0.442)	1.14 (0.335)
CRP	14.66 (<0.001*)	1.36 (0.264)	1.21 (0.317)
CAR	0.15 (0.859)	0.32 (0.574)	0.86 (0.435)
Eosinophils	0.80 (0.462)	1.33 (0.258)	0.18 (0.836)
ESR	5.76 (0.010*)	1.26 (0.272)	0.63 (0.539)
IFN-γ	11.60 (<0.001*)	2.48 (0.126)	1.09 (0.350)
IP10	7.49 (0.003*)	6.33 (0.017*)	3.18 (0.058)
Interleukin 1β	7.13 (0.003*)	4.64 (0.040*)	1.82 (0.183)
Interleukin 2Rα	0.73 (0.492)	0.06 (0.807)	0.42 (0.659)
Interleukin 6	9.85 (<0.001*)	0.20 (0.657)	2.12 (0.143)
Interleukin 8	0.45 (0.645)	0.24 (0.628)	0.08 (0.927)
Interleukin 10	5.13 (0.014*)	0.08 (0.774)	0.17 (0.845)
Interleukin 17A	1.48 (0.251)	1.01 (0.327)	0.10 (0.907)
Monocytes/Lymph	1.10 (0.349)	1.16 (0.289)	0.97 (0.394)
Neutrophils/Lymph	8.06 (0.002*)	0.23 (0.633)	0.89 (0.421)
Platelets/Lymph	1.85 (0.179)	0.17 (0.682)	0.09 (0.914)
SII	5.49 (0.011*)	0.18 (0.674)	1.16 (0.330)
TNF α	2.97 (0.072)	0.20 (0.659)	1.51 (0.242)
WBC	3.06 (0.066)	0.80 (0.378)	0.79 (0.465)
IL-10/IL-6	20.47 (<0.001*)	0.07 (0.793)	5.21 (0.014*)

S3.b. CAARMS Unusual Thought Content severity.

	LME – F (p)		
Covariate	**Assessment**	**Biomarker**	**Biomarker × Assessment**
Basophils	0.11 (0.895)	1.65 (0.208)	0.09 (0.914)
CRP	12.79 (<0.001*)	0.86 (0.369)	1.13 (0.340)
CAR	0.50 (0.610)	0.94 (0.343)	0.27 (0.766)
Eosinophils	1.05 (0.365)	0.25 (0.623)	0.57 (0.575)
ESR	5.06 (0.015*)	0.17 (0.682)	1.17 (0.327)
IFN-γ	14.60 (<0.001*)	8.29 (0.007*)	3.45 (0.044*)
IP10	6.12 (0.006*)	7.08 (0.012*)	1.64 (0.210)
Interleukin 1β	3.95 (0.031*)	1.58 (0.217)	1.99 (0.154)
Interleukin 2Rα	0.70 (0.506)	0.06 (0.812)	0.08 (0.926)
Interleukin 6	9.04 (0.001*)	0.09 (0.760)	4.36 (0.024*)
Interleukin 8	1.38 (0.272)	0.24 (0.630)	0.57 (0.573)
Interleukin 10	5.81 (0.008*)	0.03 (0.869)	0.66 (0.523)
Interleukin 17A	0.84 (0.442)	1.69 (0.206)	0.57 (0.571)
Monocytes/Lymph	1.39 (0.267)	6.35 (0.017*)	0.96 (0.396)
Neutrophils/Lymph	4.57 (0.020*)	1.54 (0.223)	0.20 (0.822)
Platelets/Lymph	3.63 (0.041*)	1.14 (0.299)	2.54 (0.099)
SII	4.37 (0.023*)	1.79 (0.189)	1.42 (0.260)
TNF α	1.99 (0.160)	0.97 (0.332)	0.41 (0.666)
WBC	3.33 (0.052)	0.01 (0.939)	1.04 (0.368)
IL-10/IL-6	13.95 (<0.001*)	<0.01 (0.968)	2.77 (0.082)

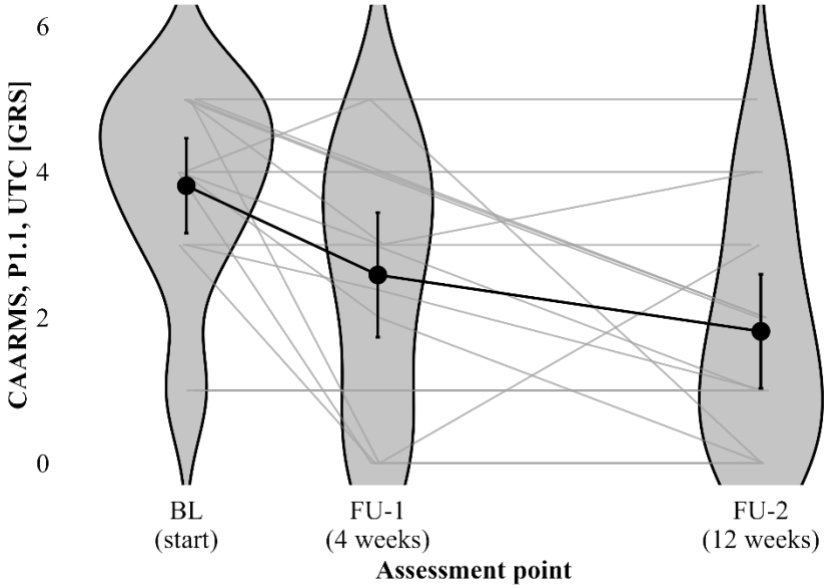
CAARMS: Comprehensive Assessment of At-Risk Mental States; CAR: Cortisol Awakening Response; CRP: C-reactive protein; DS: Disorganised Speech (CAARMS-P1.4); FDR-p: Corrected value of p for multiple comparisons, with False Discovery Rate method; ESR: Erythrocyte Sedimentation Rate; IFN-γ: Interferon-γ; IL: Interleukin; IP10: IFN-γ-induced protein-10; LME: Linear Mixed Effect model; Lymph: Lymphocyte; NBI: Non-Bizarre Ideas (CAARMS-P1.2); P1: CAARMS Positive Symptoms (UTC, NBI, PA, and DS); PA: Perceptual Abnormalities (CAARMS-P1.3); PEA: Palmitoylethanolamide (administered in its ultramicronized form); SII: Systemic Immune Inflammation index (Neutrophils×Platelets/Lymphocyte;); TNF α: Tumor Necrosis Factor; UTC: Unusual Thought Content (CAARMS-P1.1); WBC: White Blood Cell total count. *: Denotes a statistically significant effect of assessment time point (p<0.050).

Supplementary Figure 1. Longitudinal progression of CAARMS Positive Symptoms scale (sum of GRS scores for: UTC, NBI, PA, and DS) from the beginning of PEA administration.



BL: Baseline assessment; CAARMS: Comprehensive Assessment of At-Risk Mental States; DS: Disorganised Speech (CAARMS-P1.4); FU-1: First follow-up assessment; FU-2: Second follow-up assessment; GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); NBI: Non-Bizarre Ideas (CAARMS-P1.2); PA: Perceptual Abnormalities (CAARMS-P1.3); PEA: Palmitoylethanolamide (administered in its ultramicronized form); UTC: Unusual Thought Content (CAARMS-P1.1). Error bars indicate 95% confidence intervals of the mean.

Supplementary Figure 2. Longitudinal progression of CAARMS Unusual Thought Content scale (GRS score) from the beginning of PEA administration.



BL: Baseline assessment; CAARMS: Comprehensive Assessment of At-Risk Mental States; FU-1: First follow-up assessment; FU-2: Second follow-up assessment; GRS: Global Rating Scale of CAARMS symptoms (ranging: 0-6); PEA: Palmitoylethanolamide (administered in its ultramicronized form); UTC: Unusual Thought Content (CAARMS-P1.1). Error bars indicate 95% confidence intervals of the mean.

References

1. Abedini, T., et al., *Efficacy and safety of palmitoylethanolamide as an adjunctive treatment for acute mania: A randomized, double-blind, placebo-controlled trial.* Psychiatry Clin Neurosci, 2022. **76**(10): p. 505-511.
2. Albanese, M., et al., *Effects of Ultramicronized Palmitoylethanolamide (um-PEA) in COVID-19 Early Stages: A Case-Control Study.* Pharmaceuticals (Basel), 2022. **15**(2).
3. Andresen, S.R., et al., *Ultramicronized palmitoylethanolamide in spinal cord injury neuropathic pain: a randomized, double-blind, placebo-controlled trial.* Pain, 2016. **157**(9): p. 2097-2103.
4. Bacci, C., et al., *Randomized split-mouth study on postoperative effects of palmitoylethanolamide for impacted lower third molar surgery.* ISRN Surg, 2011. **2011**: p. 917350.
5. Bonzanino, M., et al., *PEALut in the Dietary Management of Patients with Acute Ischemic Stroke: A Prospective Randomized Controlled Clinical Trial.* J Clin Med, 2024. **13**(2).
6. Briskey, D., P. Ebelt, and A. Rao, *The Effect of Levagen+ (Palmitoylethanolamide) Supplementation on Symptoms of Allergic Rhinitis-A Double-Blind Placebo-Controlled Trial.* Nutrients, 2023. **15**(23).
7. Briskey, D., et al., *Effectiveness of Palmitoylethanolamide (Levagen+) Compared to a Placebo for Reducing Pain, Duration, and Medication Use during Migraines in Otherwise Healthy Participants-A Double-Blind Randomised Controlled Study.* Pharmaceuticals (Basel), 2024. **17**(2).
8. Campolo, M., et al., *Co-Ultra PEALut Enhances Endogenous Repair Response Following Moderate Traumatic Brain Injury.* Int J Mol Sci, 2021. **22**(16).
9. Cantone, E., et al., *Persistent COVID-19 parosmia and olfactory loss post olfactory training: randomized clinical trial comparing central and peripheral-acting therapeutics.* Eur Arch Otorhinolaryngol, 2024. **281**(7): p. 3671-3678.
10. Cobellis, L., et al., *Effectiveness of the association micronized N-Palmitoylethanolamine (PEA)-transpolydatin in the treatment of chronic pelvic pain related to endometriosis after laparoscopic assessment: a pilot study.* Eur J Obstet Gynecol Reprod Biol, 2011. **158**(1): p. 82-6.
11. Coraci, D., et al., *Carpal tunnel syndrome treatment with palmitoylethanolamide: neurophysiology and ultrasound show small changes in the median nerve.* Rheumatol Int, 2018. **38**(7): p. 1307-1309.

12. Costagliola, C., et al., *Effect of palmitoylethanolamide on visual field damage progression in normal tension glaucoma patients: results of an open-label six-month follow-up.* J Med Food, 2014. **17**(9): p. 949-54.
13. Cremon, C., et al., *Randomised clinical trial: the analgesic properties of dietary supplementation with palmitoylethanolamide and polydatin in irritable bowel syndrome.* Aliment Pharmacol Ther, 2017. **45**(7): p. 909-922.
14. D'Ascanio, L., et al., *Randomized clinical trial "olfactory dysfunction after COVID-19: olfactory rehabilitation therapy vs. intervention treatment with Palmitoylethanolamide and Luteolin": preliminary results.* Eur Rev Med Pharmacol Sci, 2021. **25**(11): p. 4156-4162.
15. Di Nardo, G., et al., *Palmitoylethanolamide and polydatin in pediatric irritable bowel syndrome: A multicentric randomized controlled trial.* Nutrition, 2024. **122**: p. 112397.
16. Di Stadio, A., et al., *Ultramicronized Palmitoylethanolamide and Luteolin Supplement Combined with Olfactory Training to Treat Post-COVID-19 Olfactory Impairment: A Multi-Center Double-Blinded Randomized Placebo- Controlled Clinical Trial.* Curr Neuropharmacol, 2022. **20**(10): p. 2001-2012.
17. Di Stadio, A., et al., *Parosmia COVID-19 Related Treated by a Combination of Olfactory Training and Ultramicronized PEA-LUT: A Prospective Randomized Controlled Trial.* Biomedicines, 2023. **11**(4).
18. Di Stadio, A., et al., *Treatment of COVID-19 olfactory dysfunction with olfactory training, palmitoylethanolamide with luteolin, or combined therapy: a blinded controlled multicenter randomized trial.* Eur Arch Otorhinolaryngol, 2023. **280**(11): p. 4949-4961.
19. Evangelista, M., et al., *Ultra-micronized Palmitoylethanolamide Effects on Sleep-wake Rhythm and Neuropathic Pain Phenotypes in Patients with Carpal Tunnel Syndrome: An Open-label, Randomized Controlled Study.* CNS Neurol Disord Drug Targets, 2018. **17**(4): p. 291-298.
20. Faig-Marti, J. and A. Martinez-Catassus, *Use of palmitoylethanolamide in carpal tunnel syndrome: a prospective randomized study.* J Orthop Traumatol, 2017. **18**(4): p. 451-455.
21. Faig-Marti, J. and A. Martinez-Catassus, *Measuring the placebo effect in carpal tunnel syndrome.* J Orthop Traumatol, 2020. **21**(1): p. 1.
22. Gagliano, C., et al., *Ocular hypotensive effect of oral palmitoyl-ethanolamide: a clinical trial.* Invest Ophthalmol Vis Sci, 2011. **52**(9): p. 6096-100.
23. Germini, F., et al., *N-of-1 Randomized Trials of Ultra-Micronized Palmitoylethanolamide in Older Patients with Chronic Pain.* Drugs Aging, 2017. **34**(12): p. 941-952.

24. Ghazizadeh-Hashemi, M., et al., *Palmitoylethanolamide as adjunctive therapy in major depressive disorder: A double-blind, randomized and placebo-controlled trial.* J Affect Disord, 2018. **232**: p. 127-133.
25. Giammusso, B., R. Di Mauro, and R. Bernardini, *The efficacy of an association of palmitoylethanolamide and alpha-lipoic acid in patients with chronic prostatitis/chronic pelvic pain syndrome: A randomized clinical trial.* Arch Ital Urol Androl, 2017. **89**(1): p. 17-21.
26. Guida, G.D.M., M.; De Fabiani, A.; Cantieri, L.; Alexandre, A.; Vassallo, G. M.; Rogai, M.; Lanaia, F.; Petrosino, S., *Palmitoylethanolamide (Normast®) in chronic neuropathic pain by compressive type lumbosciatalgia: Multicentric clinical study.* DOLOR, 2010(25): p. 35-42.
27. Isola, G., et al., *Effectiveness of a nutraceutical agent in the non-surgical periodontal therapy: a randomized, controlled clinical trial.* Clin Oral Investig, 2021. **25**(3): p. 1035-1045.
28. Kadanangode Narayanaswam, N., et al., *A randomized interventional clinical trial assessing the safety and effectiveness of PeaNoc XL tablets in managing joint pain and inflammation in arthritis patients.* F1000Res, 2023. **12**: p. 895.
29. Khalaj, M., et al., *Palmitoylethanolamide as adjunctive therapy for autism: Efficacy and safety results from a randomized controlled trial.* J Psychiatr Res, 2018. **103**: p. 104-111.
30. Lunardelli, M.L., et al., *Co-ultraPEALut: Role in Preclinical and Clinical Delirium Manifestations.* CNS Neurol Disord Drug Targets, 2019. **18**(7): p. 530-554.
31. Marini, I., et al., *Palmitoylethanolamide versus a nonsteroidal anti-inflammatory drug in the treatment of temporomandibular joint inflammatory pain.* J Orofac Pain, 2012. **26**(2): p. 99-104.
32. Masek, K., et al., *Prophylactic efficacy of N-2-hydroxyethyl palmitamide (impulsin) in acute respiratory tract infections.* Eur J Clin Pharmacol, 1974. **7**(6): p. 415-9.
33. Murina, F., et al., *Vestibulodynia: synergy between palmitoylethanolamide + transpolydatin and transcutaneous electrical nerve stimulation.* J Low Genit Tract Dis, 2013. **17**(2): p. 111-6.
34. Orefice, N.S., et al., *Oral Palmitoylethanolamide Treatment Is Associated with Reduced Cutaneous Adverse Effects of Interferon-beta1a and Circulating Proinflammatory Cytokines in Relapsing-Remitting Multiple Sclerosis.* Neurotherapeutics, 2016. **13**(2): p. 428-38.
35. Ottaviani, G., et al., *Efficacy of ultramicronized palmitoylethanolamide in burning mouth syndrome-affected patients: a preliminary randomized double-blind controlled trial.* Clin Oral Investig, 2019. **23**(6): p. 2743-2750.

36. Palma, E., et al., *Acetylcholine receptors from human muscle as pharmacological targets for ALS therapy.* Proc Natl Acad Sci U S A, 2016. **113**(11): p. 3060-5.
37. Pickering, E., et al., *A randomized controlled trial assessing the safety and efficacy of palmitoylethanolamide for treating diabetic-related peripheral neuropathic pain.* Inflammopharmacology, 2022. **30**(6): p. 2063-2077.
38. Rao, A., et al., *Palmitoylethanolamide for sleep disturbance. A double-blind, randomised, placebo-controlled interventional study.* Sleep Sci Pract, 2021. **5**(1): p. 12.
39. Rao, A., R. Skinner, and D. Briskey, *The Efficacy of Palmitoylethanolamide (Levagen+) on the Incidence and Symptoms of Upper Respiratory Tract Infection-A Double Blind, Randomised, Placebo-Controlled Trial.* Nutrients, 2023. **15**(20).
40. Rao, A., et al., *Efficacy of Topical Palmitoylethanolamide (Levagen+) for the Management of Eczema Symptoms: A Double-Blind, Comparator-Controlled, Randomized Clinical Trial.* Skin Pharmacol Physiol, 2023. **36**(6): p. 288-295.
41. Rossi, G.C.M., et al., *Effect of palmitoylethanolamide on inner retinal function in glaucoma: a randomized, single blind, crossover, clinical trial by pattern-electroretinogram.* Sci Rep, 2020. **10**(1): p. 10468.
42. Salaffi, F., et al., *Palmitoylethanolamide and acetyl-L-carnitine act synergistically with duloxetine and pregabalin in fibromyalgia: results of a randomised controlled study.* Clin Exp Rheumatol, 2023. **41**(6): p. 1323-1331.
43. Salehi, A., et al., *Adjuvant palmitoylethanolamide therapy with risperidone improves negative symptoms in patients with schizophrenia: A randomized, double-blinded, placebo-controlled trial.* Psychiatry Res, 2022. **316**: p. 114737.
44. Steels, E., et al., *A double-blind randomized placebo controlled study assessing safety, tolerability and efficacy of palmitoylethanolamide for symptoms of knee osteoarthritis.* Inflammopharmacology, 2019. **27**(3): p. 475-485.
45. Strobbe, E., M. Cellini, and E.C. Campos, *Effectiveness of palmitoylethanolamide on endothelial dysfunction in ocular hypertensive patients: a randomized, placebo-controlled cross-over study.* Invest Ophthalmol Vis Sci, 2013. **54**(2): p. 968-73.
46. Tartaglia, E., et al., *Effectiveness of the Association N-Palmitoylethanolamine and Transpolydatin in the Treatment of Primary Dysmenorrhea.* J Pediatr Adolesc Gynecol, 2015. **28**(6): p. 447-50.
47. Versace, V., et al., *Co-ultramicronized palmitoylethanolamide/luteolin normalizes GABA(B)-ergic activity and cortical plasticity in long COVID-19 syndrome.* Clin Neurophysiol, 2023. **145**: p. 81-88.

48. Yuan, C., et al., *N-palmitoylethanolamine and N-acetylethanolamine are effective in asteatotic eczema: results of a randomized, double-blind, controlled study in 60 patients*. Clin Interv Aging, 2014. **9**: p. 1163-9.