Analysis of Potential Subgroups in Vaes ME/CFS Patient Clusters

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Abstract

Background: Vaes et al. (2023)[1] identified 13 symptom clusters in a large cohort of ME/CFS patients. Symptom intensity is broadly correlated with post-exertional malaise (PEM) severity, with variation across clusters that seems disorganized. Despite this research, no broadly accepted organizing principle has emerged from this paper or other attempts at phenotyping ME/CFS.

Objective: To identify and characterize potential subgroups defined by symptom domain severity relative to PEM within the original Vaes symptom clusters.

Methods: We analyzed the Vaes cluster summary data[2], calculated geometric means for each symptom domain within each cluster, and plotted these means against PEM severity to identify patterns and subgroups.

Results: Our analysis identified two groups of patient clusters with distinct symptom-domain profiles. The first group showed a consistent amplification pattern across all symptom domains as PEM increased, whereas the second group has selective amplification: pain and neurocognitive symptoms escalated more rapidly with PEM, while immune and sleep symptoms remained relatively flat. This subgroup's symptom trends could align with those of fibromyalgia. Further exploration with individual patient data is needed to validate these findings.

Keywords: Myalgic Encephalomyelitis, ME/CFS, patient clustering, fibromyalgia, post-exertional malaise, PEM

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1 Introduction

Dr. Anouk W. Vaes and her collaborators at CIRO performed one of the largest systematic surveys of ME/CFS symptom patterns, applying clustering methods to patient-reported outcomes to identify recurring constellations of symptoms[1]. They used symptom surveys based on the DePaul Symptom Questionnaire version 2 (DSQ-2) from 337 patients to identify a total of 45 patient clusters, of which 13 were of size ≥ 10 while the remaining were mostly of size ≤ 2 and approximately 9.4% of the total cohort. We used the publicly available data of their final 13 clusters for most of the analysis[2] but we discuss the small clusters in Section 4.

While Vaes described each cluster, and noted some differences among symptom severity, an overall organizing principle was not proposed. Here we examine whether one might emerge from that dataset.

Our analysis suggests that the Vaes ME/CFS clusters can be organized into two overarching families defined by their relationship to PEM. This reduces the complexity of the cluster symptom intensities and highlights a subgroup within the Vaes clusters that has not been explicitly recognized in prior work. Once separated and profiled, this subgroup has pain and neurocognitive trends that could meet fibromyalgia criteria if the data extended along the regression lines. This paper invites further investigation with patient-level symptom data to validate or refute these findings and clarify their clinical and biological relevance.

2 Symptom Domains Relative to PEM

The strong weight of PEM symptoms makes the Vaes dataset tricky to interpret if you only look at raw intensity, so instead we compared overall symptom levels to PEM severity. From this comparison we identified two groups of clusters that maintain their integrity across most symptom domains.

Because these analyses are exploratory and based on cluster-level data, the patterns described below should be interpreted as hypothesis-generating.

We first examine the areas of most similarity before highlighting how they differ. We add an "all" category to gauge overall symptom severity to the original symptom domains. Our use of the terms "high-intensity" and "low-intensity" does not reproduce the clusters described by Vaes or others but reflects a new organization based on how symptom domains scale with PEM. Vaes used the DePaul symptom groupings which we keep as-is.

Notes: The DSQ-2 uses a single symptom for fatigue. Also, for the "Overall" chart we used the arithmetic mean instead of geometric mean for the Y axis. All other graphs in this study use geometric means exclusively. Shaded bands represent the 95% confidence interval of the linear models when used. While this section focuses on visual inspection, we provide more statistical descriptions in Appendix B.

2.1 Similarities

In terms of fatigue, the two groups overlap completely, as ME/CFS patient clusters are expected to. See Figure 1, below. While the lines have a similar slope and offset we note the R^2 value is significantly weaker in the low-intensity group (0.54 vs. 0.86).

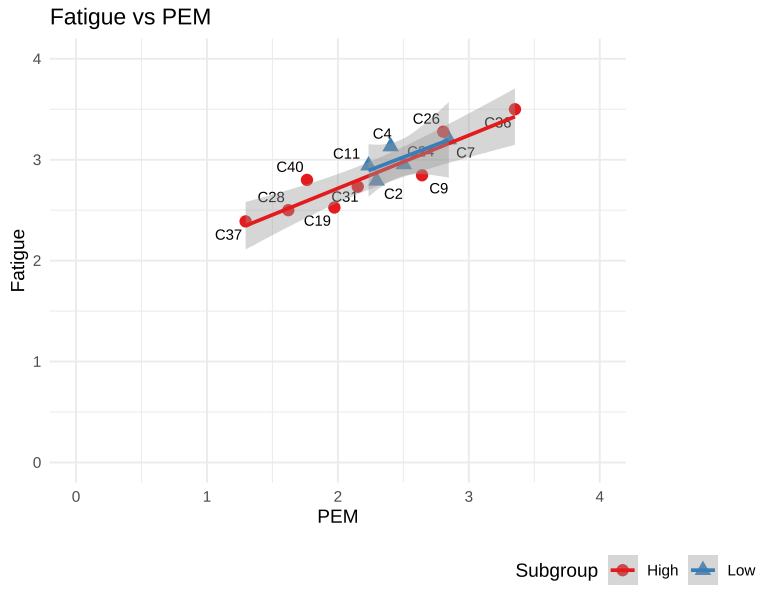

Figure 1: Fatigue vs. PEM

While fatigue shows complete overlap in our groups the next charts (Figure 2) show how they stand out. In the first chart (top left) we compare the average of all symptoms to PEM. In this chart one can easily discern two tiers of clusters. One has a higher overall symptom burden at any given PEM level compared to the other and this relationship is where we derive the names for our groups: high and low-intensity.

Autonomic, Neuroendocrine and Other also follow a similar pattern: Parallel but lower than the high-intensity group.

We note that the limited range of PEM severity in the low-intensity group (~2.2 to 2.9) may have otherwise caused it to remain undetected.

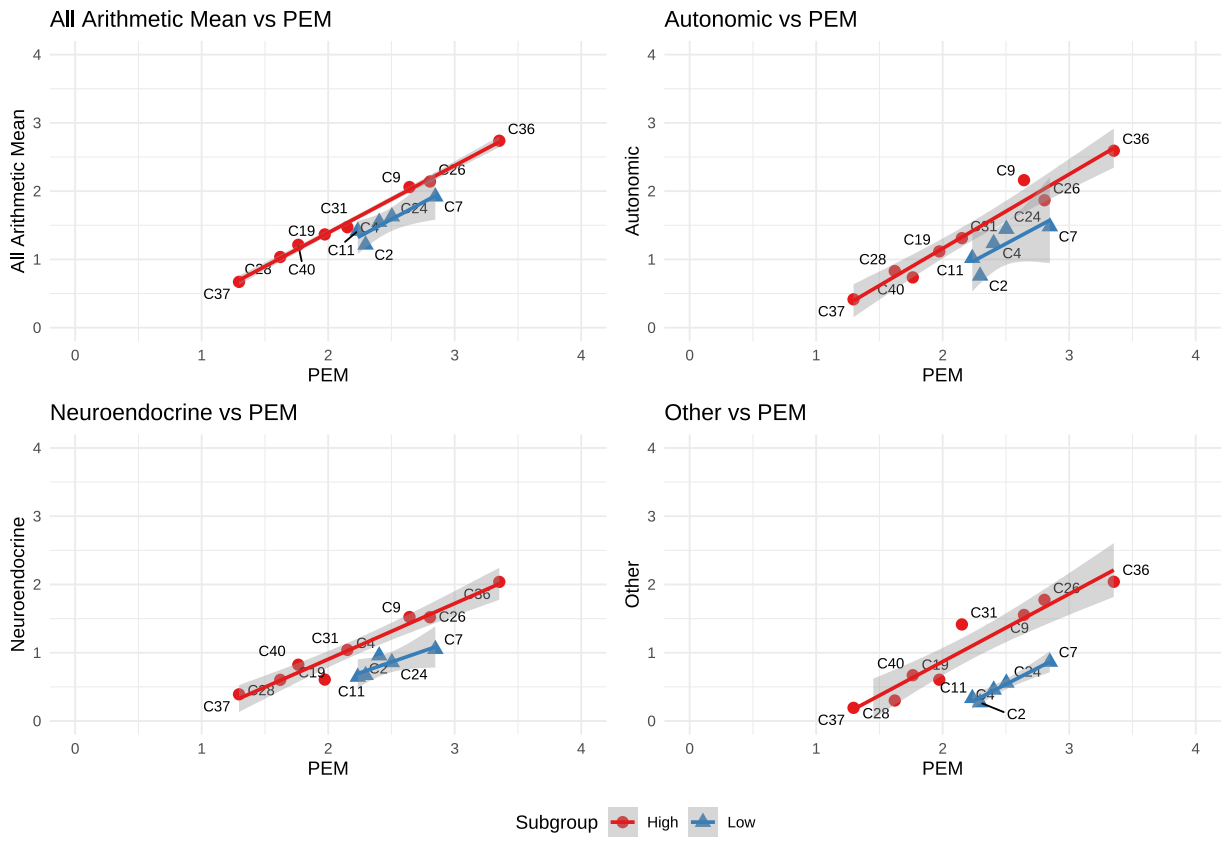

Figure 2: Similar Symptom Domains vs. PEM

Groupings: From visual inspection of the charts above we group clusters C9, C19, C26, C28, C31, C36, C37, C40 as high-intensity, C2, C4, C7, C11, C24 as low-intensity. We'll keep these groupings for all plots that follow. The high and low groups are approximately 54.5% and 45.5% of the total cohort, respectively.

The last chart (Other vs. PEM) shows a markedly lower offset and is perhaps the most visibly obvious difference between the two groups. The two symptoms in 'Other' are sensitivity to mold and vibration.

While the groups maintain integrity, the parallelism observed in the charts above does not hold in the next charts and suggests a more complex physiological cause for the differences.

2.2 Separate Amplification Patterns Relative to PEM

Pain and neurocognitive domains show that while the low-intensity group remains offset, symptom domain amplification is increased. As PEM increases to 3, C7 comes very close to the high-intensity line.

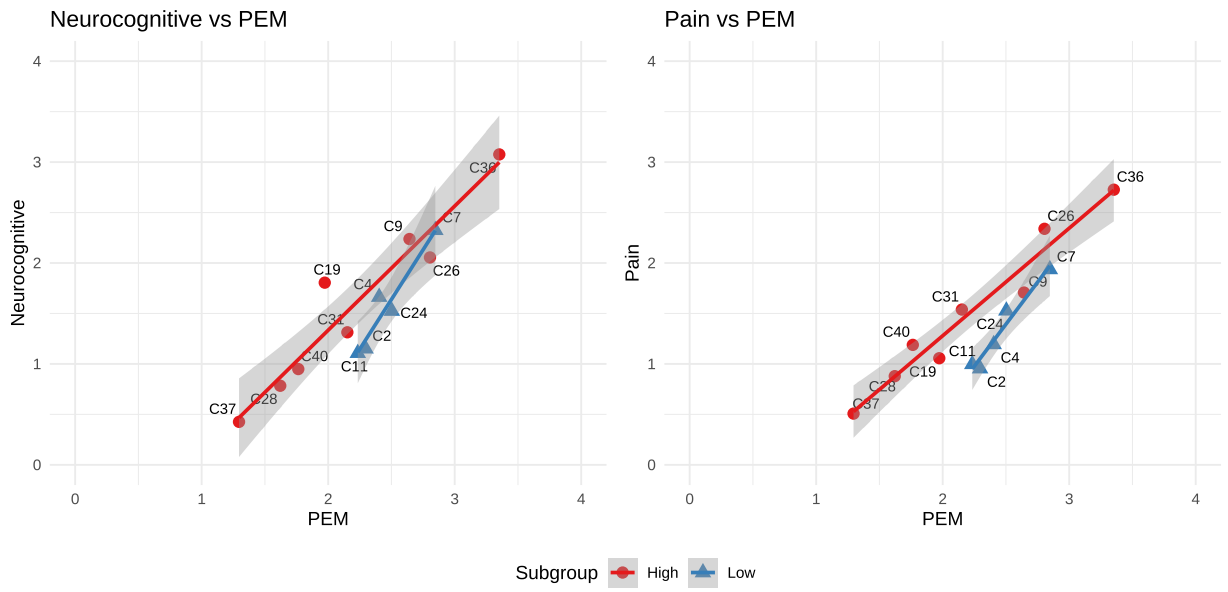

Figure 3: Pain and Neurocognitive vs. PEM

The regression lines for the low-intensity group suggest that if the data extended further they could meet fibromyalgia criteria. We discuss this more fully in Section 3.

2.3 Weak Correlations Relative to PEM

In all of the previous charts we've shown that both groups maintain a strong but distinct relationship to PEM. By contrast, the immune and sleep domains show little to no correlation with PEM severity in the low-intensity group. Notably, despite this lack of correlation, sleep disturbance can be pronounced in the low-intensity group.

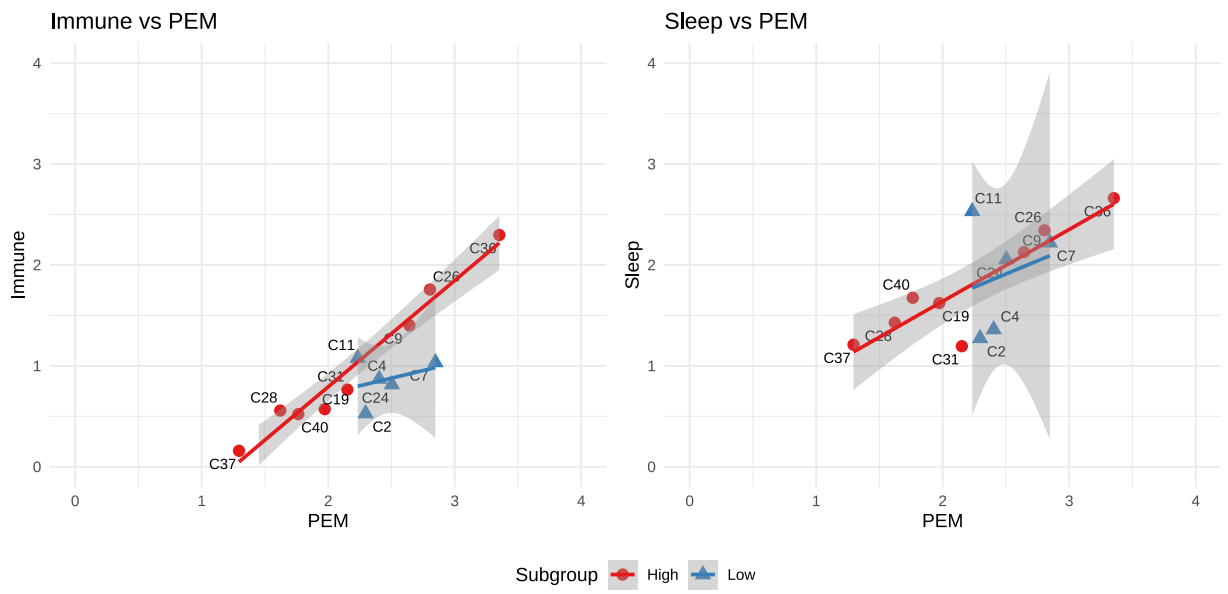

Figure 4: Immune and Sleep vs. PEM

2.4 Summary of Differences

High-intensity: Has a consistent amplification pattern: as PEM increases, all other symptom domains rise together.

Low-intensity: Presents a more selective profile. Pain, neurocognitive, and neuroendocrine symptoms escalate more rapidly with PEM, while sleep and immune symptoms show little or no correlation. Additionally, the low-intensity group is notably nestled in the middle of the overall PEM range which could allow it to hide among the other clusters if not actively searched for.

We summarize these differences in the table below.

Table 1: Summary of Group Differences

Feature	High group	Low group
Overall vs. PEM	Higher	Lower - esp. “Other”
Fatigue	R^2 - 0.86	Weaker R^2 - 0.54
PEM range	~1.3 – 3.4	Narrower ~2.2 – 2.9
Neurocognitive	Tracks PEM	Elevated amplification
Pain	Tracks PEM	Elevated amplification
Immune	Tracks PEM	Flat at ~1
Sleep	Tracks PEM	~1.4 – 2.5, but uncorrelated

Symptom Severity: In terms of overall symptom range the two groups largely overlap. The high-intensity group, by symptom intensity alone, could be considered a superset of the low-intensity group. It is the differing relationships to PEM and the selective amplification of pain and neurocognitive symptom domains in the low-intensity group that set the two groups apart. We discuss this further in Section 3, below.

3 Exploring the Fibromyalgia–trending Subgroup

Based on the trends noticed in Section 2.2, a discussion of whether the low-intensity group could be related to fibromyalgia is warranted. The question of whether we have identified a clinically useful subgroup in the Vaes data remains an important but separate debate.

To be clear, we do not claim this subgroup is clinically fibromyalgia, but that the pain and neurocognitive symptom trends suggest that group members might meet fibromyalgia criteria if additional group members appeared along the low-intensity regression lines. Further, if this subgroup represents a continuum of the same physiological processes responsible for fibromyalgia it would have implications for future diagnostic criteria.

Vaes[1] described the selection criteria for their cohort as follows:

> Almost 90% of the participants fulfilled the Fukuda case definition, compared to 80%, 59% and 39% fulfilling the IOM, CCC and ME-ICC case definitions, respectively. More than a quarter of the participants met the criteria for all four different case definitions, whilst 5% of the participants met none of the abovementioned case definitions,…

This seems like a potentially wide net and may help explain two potential subgroups which suggest a different pathophysiology. An examination of the proposed low-intensity subgroup shows it occupies a very narrow range of PEM intensity and then stops abruptly when PEM reaches ~2.9. We see no reason for a PEM-specific boundary but the charts in Figure 3 suggest that our group is not PEM-limited but pain- and neurocognitive-limited within this dataset. In those charts, pain and neurocognitive symptoms trend upward and then the clusters stop at C7 just before they would cross the high-intensity group trend lines. This truncation could be due to diagnostic criteria that would classify these individuals as fibromyalgia when pain and neurocognitive scores are above the high-intensity lines.

This would align with published DSQ-2 studies showing that ME/CFS patients who meet fibromyalgia criteria exhibit amplified pain and more severe post-exertional malaise compared with ME/CFS alone[3–5] and would also explain why our subgroup occupies such a narrow range of PEM scores compared to the overall cohort.

Table 2: ME/CFS vs. Fibromyalgia

Domain (DSQ / DSQ-PEM items)	ME/CFS (no FM)	ME/CFS + FM	FM (alone)
Post-Exertional Malaise (PEM)	Core feature; DSQ captures frequency & severity well	**Higher** PEM frequency/severity than ME/CFS alone (incl. “General” and “Muscle” PEM factors)	Not a defining criterion of FM; DSQ-PEM shows variable/typically lower PEM signal vs ME/CFS cohorts
Pain	Common, variable; not primary diagnostic driver	**Higher overall pain burden**; amplifies illness severity	**Defining**; widespread musculoskeletal pain central to diagnosis

Domain (DSQ / DSQ-PEM items)	ME/CFS (no FM)	ME/CFS + FM	FM (alone)
Neurocognitive	Frequent (attention, memory, processing speed)	Often **worse** with FM comorbidity	Present in many FM cohorts; DSQ-SF cognitive items used in FM studies
Sleep	Non-restorative sleep common	Often **worse** with FM comorbidity	Common in FM, often prominent
Autonomic / Immune / Neuroendo	Captured across DSQ domains; heterogeneous patterns documented	Can be **more burdensome** with FM comorbidity	Less emphasized in FM criteria; may appear but not core

It is possible that current pain and neurocognitive thresholds for a fibromyalgia diagnosis may focus too heavily on symptom severity to capture the nuanced patterns revealed by our subgroup analysis. If these groups are commingled, then properly separating the two groups may require a more complex approach that relies less on absolute pain scores and more on the ratio of symptom domain scores to PEM.

Recent work in fibromyalgia adds weight to this possibility. Maurel et al. (2023) applied hierarchical clustering to a population-based FM cohort and identified distinct clinical–neuropsychological phenotypes, separating pain-dominant and maladaptive-coping profiles from more resilient ones[6]. Although FM lacks a PEM requirement, these findings align with our two-subgroup model: they suggest that the low-intensity ME/CFS group—marked by elevated pain and neurocognitive amplification—may represent a fibromyalgia-like subtype that retains post-exertional features.

While the hypothesis that this subgroup is related to fibromyalgia is attractive, other explanations may also be true. At the very least, these data need direct comparison to fibromyalgia cohorts before it can be used to formally claim a fibromyalgia link.

4 Characteristics of the Smallest Clusters

Vaes[1] identified 32 additional clusters[7]. These were mostly of size 1, with 4 clusters of size 2 and represent approximately 9.4% of the cohort. We examined these smaller clusters in light of our two-group hypothesis.

Taken as a whole the 13 large clusters appear to have captured the broad trends of the entire dataset with the remaining clusters spread across and beyond our two subgroups. This may suggest that the two groups are not distinct but artifacts of the clustering constraints when applied to data that is widespread.

Specifically however, the small clusters mostly had sleep scores above the high-intensity group's regression lines and showed similar upper pain and neurocognitive limits as described in Section 3.

Finally, the small clusters, when added to the large clusters, did not significantly move their location or regression lines.

5 Discussion

We have shown that the Vaes clusters can be organized into two subgroups defined by symptom domains relative to PEM, and that these subgroups remain consistent across most domains. Grouping them by their relationship to PEM simplifies the original complexity at both the domain and cluster levels. From this perspective, the 13 Vaes clusters can be reduced to two groups of linear models, where membership in one group combined with PEM intensity allows inference of the intensity of other symptom domains.

While these patterns are striking, our analysis is a secondary exploration based on symptom-domain summaries of the Vaes patient data. The small subgroup sizes (eight and five clusters) and the original clustering approach may distort some relationships, and the plots can suggest trends that do not necessarily exist at the patient level. The apparent high/low separation may therefore reflect artifacts of the clustering algorithm averaging a broad underlying distribution rather than true biological subgroups, which would render our suggested distinctions essentially coincidental.

These considerations highlight the need for validation before our findings can be clinically relevant. We see challenges in at least three areas: whether the groupings hold when tested with individual patient data; whether symptom-domain analysis can reliably identify the group to which a given patient belongs; and how these subgroups relate—or fail to relate—to fibromyalgia.

As an example, Vaes[1] provided cluster-specific summaries of standout symptoms; some of those align with our domain-based charts while others show a different emphasis, demonstrating the challenge of reconciling symptom-level observations with domain-level analyses.

Despite these limitations, we believe further steps are justified. Analyses using the original patient scores could validate, refine, or repudiate these subgroups and help identify a short list of differentiating symptoms. If that proves useful, examining biological markers across these subgroups could help reveal underlying pathophysiological differences. In addition, applying similar analyses to fibromyalgia and long-COVID could clarify relationships among these overlapping conditions or even help refine how ME/CFS and fibromyalgia are classified.

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6 Conclusion

Our secondary analysis suggests that the 13 clusters identified by Vaes can be organized into two broader structures defined by overall symptom intensity relative to PEM severity. These two groups remain coherent across symptom domains and display distinct, largely linear relationships with PEM. Although exploratory and limited to cluster-level data, these findings offer a reproducible framework for further research, specifically for validating these potential subgroups and exploring their clinical and biological relevance.

7 Appendices

7.1 Appendix A: Cluster Summary Data

We use the Vaes cluster spreadsheet[2] exclusively as our data source. We use R for significant cleaning and transformation from Excel to CSV. After removing everything but the mean severity of the symptoms in the 13 clusters we:

- Group each cluster by symptom domain and calculate geometric means for each symptom domain within each cluster.
- Rotate the table
- Add a new column, "all_mean" which is the arithmetic mean of all symptoms within each cluster.
- Save this file as cluster_grouped_tidy.csv

This file is what we then used for our analysis, and the plots. We present the final data used below. Values were rounded and abbreviations were used for formatting. See the Code Availability section for links to the code used to generate this file.

Table 3: Cluster Summary Data with Geometric Means by Symptom Domain

Cluster	All	PEM	Fatigue	Auto	Immune	NC	NE	Pain	Other
C2	1.21	2.30	2.79	0.76	0.53	1.15	0.67	0.95	0.27
C4	1.55	2.40	3.13	1.24	0.87	1.66	0.96	1.19	0.46
C7	1.92	2.85	3.20	1.48	1.03	2.32	1.05	1.94	0.86
C9	2.06	2.64	2.85	2.16	1.40	2.24	1.52	1.71	1.55
C11	1.42	2.23	2.94	1.02	1.08	1.11	0.64	1.00	0.33
C19	1.37	1.97	2.53	1.12	0.57	1.81	0.61	1.06	0.60
C24	1.63	2.50	2.95	1.44	0.82	1.52	0.86	1.53	0.56
C26	2.14	2.80	3.28	1.87	1.76	2.06	1.52	2.34	1.77
C28	1.03	1.62	2.50	0.83	0.56	0.78	0.60	0.88	0.30
C31	1.47	2.15	2.73	1.31	0.77	1.31	1.04	1.54	1.41
C36	2.74	3.35	3.50	2.59	2.30	3.08	2.04	2.73	2.04
C37	0.67	1.30	2.39	0.41	0.16	0.43	0.39	0.51	0.19
C40	1.21	1.76	2.80	0.74	0.52	0.95	0.82	1.19	0.67

Appendix B: Statistical Descriptions

In the body of the text, we used visual analysis to identify two subgroups of ME/CFS patient clusters defined by their symptom domain profiles relative to PEM severity. Here we present statistical analyses that may help clarify the data in the plots.

Given the exploratory nature of this study and the small number of clusters, these results should be interpreted as descriptive rather than confirmatory.

R^2 by Group

We compare the coefficient of determination (R^2) of the symptom domains in three levels:

- Combined (all clusters)
- High-intensity group
- Low-intensity group

Table 4: R^2 by Domain

Domain	Combined R^2	High-intensity R^2	Low-intensity R^2
All_mean	0.91	1.00	0.84
Autonomic	0.79	0.96	0.62
Fatigue	0.84	0.86	0.54
Immune	0.78	0.96	0.11
Neurocognitive	0.85	0.92	0.93
Neuroendocrine	0.73	0.96	0.75
Other	0.52	0.92	0.96
Pain	0.81	0.95	0.95
Sleep	0.53	0.81	0.05

Table 4 shows that for almost every symptom domain, the high-intensity group has a higher R^2 than the combined group, which in turn has a higher R^2 than the low-intensity group. In some cases – such as neurocognitive, neuroendocrine, and Other – the combined R^2 is significantly weaker than either of the subgroups.

Leave-one-out Analysis

We discovered our two groups by visual analysis, but we can re-identify the set using leave-one-out analysis. In this analysis we go through each domain and examine what happens when we remove one cluster at a time. The goal is to identify which clusters, when removed, improve R^2 the most.

We summarize the mean R^2 differences, in order of greatest improvement, in Table 5.

Of the top 6 clusters only C31 is not part of the low-intensity group. While this does not prove the existence of two subgroups it does support the idea that the members of the low-intensity group are, overall, responsible for most of the R^2 degradation in the combined group.

Table 5: Leave-one-out Analysis

Cluster	Group	Domains Improved	Mean ΔR^2
C2	low	9	0.0379
C11	low	9	0.0271
C4	low	8	0.0191
C31	high	7	0.0163
C7	low	5	0.0129
C24	low	8	0.0126

ANCOVA

We performed an analysis of covariance (ANCOVA) to identify the domains which could be most statistically significant and discriminatory. We compare our two groups for each symptom domain. We arrange the results by descending F-statistic and show the p-value rounded to three decimal places.

Table 6: ANCOVA Analysis

Domain	F statistic	p-value	Significance
Other	33.3	< 0.001	***
Neuroendocrine	11.6	0.007	**
Fatigue	5.2	0.046	*
Autonomic	3.8	0.08	.
Pain	3.1	0.108	NA
Immune	1.4	0.268	NA
All_mean	1.0	0.33	NA
Sleep	0.2	0.647	NA
Neurocognitive	0.0	0.849	NA

Table 6 shows agreement with the charts that Other, Neuroendocrine and Autonomic domains show the most significant offsets between the two groups. The high F-statistic and low p-value for Fatigue, however, do not agree with the regression lines. As we note earlier in Section 2.1, the lines in Figure 1 are nearly identical but R^2 is significantly weaker in the low-intensity group. The high F-statistic and p-value in the ANCOVA analysis for fatigue is most likely an artifact of the wide spread in the low-intensity group.

Code Availability

All data manipulation and analysis scripts are available at
https://github.com/eriksquires/VaesSubgroups

References

1. Vaes, A. W. *et al.* Symptom-based clusters in people with ME/CFS: An illustration of clinical variety in a cross-sectional cohort. *Journal of Translational Medicine* **21**, 112 (2023).
2. Vaes, A. W. & collaborators. Supplementary dataset (excel): Additional file 3: Table S1 : 12967_2023_3946_MOESM3_ESM.xlsx. Journal of Translational Medicine, Springer Nature (2023).
3. Jason, L. A. *et al.* The DePaul symptom questionnaire-2: A validation study. *Fatigue: Biomedicine, Health & Behavior* **3**, 1–13 (2015).
4. McManimen, S. L., Jason, L. A. & Williams, Y. J. Post-exertional malaise in patients with myalgic encephalomyelitis/chronic fatigue syndrome with comorbid fibromyalgia. *Fatigue: Biomedicine, Health & Behavior* **5**, 102–117 (2017).
5. Almenar-Pérez, E. & al., et. MicroRNA profiles distinguish fibromyalgia, ME/CFS, and ME/CFS with fibromyalgia. *Scientific Reports* **13**, 28955 (2023).
6. Maurel, S., Giménez-Llort, L., Alegre-Martin, J. & Castro-Marrero, J. Hierarchical cluster analysis based on clinical and neuropsychological symptoms reveals distinct subgroups in fibromyalgia: A population-based cohort study. *Biomedicines* **11**, 2867 (2023).
7. Vaes, A. W. & collaborators. Supplementary dataset (excel): Additional file 4 table S2 : 12967_2023_3946_MOESM4_ESM.xlsx. Journal of Translational Medicine, Springer Nature (2023).

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Dr. Vaes had no role in the writing of this paper, and all errors or misinterpretations are the responsibility of the author.

Author Contributions

Erik K. Squires conceived the study, performed the analysis, and wrote the manuscript. This work presents an original investigative method and resulting framework which were both developed and first reported by the author in this preprint.

Competing Interests

The author declares no competing interests.

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Ethics Statement

This study reanalyzed publicly available reports and published symptom cluster data (Vaes 2023). No new patient data were collected.