

Autonomic Indices Reveal Physiological Emotional Phenotypes during Virtual Reality Elicitation

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Abstract

Conventional emotion-recognition approaches rely on group-averaged features, overlooking meaningful inter-individual variability in emotion regulation.

We investigate within-emotion autonomic heterogeneity using heart rate variability (HRV) instantaneous Sympathetic and Parasympathetic Activity Indices (SAI, PAI) from a point-process electrocardiographic (ECG) framework in 38 participants undergoing a validated virtual reality protocol eliciting four primary emotions. Building on previously identified phenotypes, we quantify SAI/PAI temporal dynamics via cluster-based permutation.

Fear showed the greatest heterogeneity: a fight-or-flight phenotype (24%) with progressive SAI increase and PAI collapse, versus a freeze phenotype (76%) with SAI suppression and preserved parasympathetic tone. Happiness, Sadness, and Relax each showed a majority (90–97%) and minority phenotype. A cluster-based permutation test revealed a sustained divergence in both SAI and PAI between the Fear phenotypes over most of the scene.

These results show that autonomic phenotypes are distinguished not only by their average level but by a specific, temporally structured divergence, supporting phenotype-aware affective computing.

1. Introduction

Emotion-recognition systems conventionally characterize affective states from physiological signals averaged across groups of subjects, implicitly treating autonomic reactivity as homogeneous within an emotion category. This assumption is at odds with growing evidence that individuals engage markedly different regulation strategies when exposed to the same affective stimulus, and that this inter-individual variability is not merely noise but may carry physiologically meaningful, subject-specific information. Static, epoch-averaged heart rate variability (HRV) metrics are poorly suited to

resolving this variability, since they compress an entire emotional episode into a handful of summary statistics and cannot separate sympathetic and parasympathetic contributions with adequate temporal resolution.

Point-process models of heartbeat dynamics overcome these limitations by providing instantaneous, beat-to-beat estimates of autonomic outflow [1]. Building on this framework, the Sympathetic and Parasympathetic Activity Indices (SAI and PAI) [2] disentangle the two branches of the autonomic nervous system with a temporal resolution unattainable by frequency-domain HRV. In the same emotion elicitation protocol used in this study, dynamic features capturing the temporal coordination between skin conductance and heart rate have recently been shown to reveal emotion-related autonomic dynamics that traditional, static HRV features fail to capture [3], reinforcing the case that a temporal, rather than purely averaged, characterization of the autonomic response can reveal not only whether phenotypes diverge, but when and for how long — information a single scene-average necessarily discards. We hypothesize that the temporal evolution of instantaneous SAI and PAI, tracked continuously rather than summarized as a single value, is sufficient to reveal distinct autonomic phenotypes within a single emotion category.

Autonomic phenotypes for the four primary emotions of this protocol — Fear, Happiness, Sadness, and Relax — were identified through an unsupervised, multimodal feature-selection and clustering pipeline, yielding two phenotypes per emotion. In this work, we quantify the temporal evolution of instantaneous SAI and PAI separately for each phenotype using a cluster-based permutation approach, providing convergent physiological evidence for the clustering solutions and directly testing whether phenotype-defining differences are detectable from the dynamics of cardiac autonomic indices alone.

2. Methods

2.1. Experimental protocol

Thirty-eight healthy volunteers took part in an immersive virtual reality (VR) emotion-elicitation protocol. Each participant was exposed to a set of validated 150-second audiovisual scenes designed to elicit seven distinct emotional states, of which four — Fear, Happiness, Sadness, and Relax — are primary emotions spanning the valence-arousal space and are the focus of the present analysis. Precise temporal markers were logged at every scene transition to support accurate signal segmentation.

2.2. Physiological data acquisition

Five physiological signals were recorded simultaneously as part of the broader VR protocol. ECG, blood volume pulse (BVP), galvanic skin response (GSR), and respiration were acquired with an 8-channel USB peripheral system (ProComp Infiniti, Thought Technology): ECG through a 3-lead Einthoven configuration, respiration via a thoracic extensometer, GSR from two finger electrodes, and BVP with a reflectance pulse oximeter. A 32-channel wireless gel-based EEG system (Emotiv FLEX, 10-20 layout, Ag/AgCl electrodes) was recorded in parallel. ECG and BVP were sampled at 2048 Hz, and GSR, respiration, and EEG at 256 Hz. The present analysis specifically exploits the ECG-derived instantaneous autonomic indices described below.

2.3. Estimation of HRV Sympathetic and Parasympathetic Activity indices

Instantaneous sympathetic and parasympathetic activity was estimated from the R-R interval series derived from a point-process probabilistic framework that models heartbeat generation as an inhomogeneous, history-dependent process [1]. Within this framework, two structurally distinct linear models of the instantaneous RR mean based on a combination of Laguerre functions capture the fast, parasympathetic (vagal) and the slower, sympathetic contributions to heart-period dynamics, respectively. The two sets of time-varying coefficients are combined into continuous-time SAI and PAI series, respectively, updated at every heartbeat and interpolated onto a uniform time base for direct comparison across subjects and conditions [2].

2.4. Autonomic phenotype identification

Two physiologically distinct autonomic phenotypes per primary emotion were identified in a preliminary multimodal clustering analysis (feature selection, Principal

component analysis, and within-emotion K-means; $K \in \{2, \dots, 5\}$, selected by maximizing the mean silhouette coefficient), not detailed further here. For all four primary emotions considered, this procedure converged to a silhouette-optimal two-cluster solution, with prevalence reported in Table 1; the present work builds on these phenotype labels to provide independent, time-resolved validation based on the instantaneous cardiac autonomic indices alone.

2.5. Temporal dynamics analysis

For each subject and primary emotion, the SAI and PAI series were time-locked to scene onset and resampled onto a common 1 Hz time base spanning the 150-second scene duration, matching the native beat-to-beat sampling density. Grand-average trajectories (mean $\pm$ Standard Error of the Mean (SEM)) were computed separately for each phenotype, providing a qualitative, time-resolved picture of how phenotypes diverge over the course of the scene.

To quantify this divergence while retaining full temporal resolution, phenotypes were compared at each time point with a Welch t-statistic, and a cluster-based permutation test [4] was used to control for multiple comparisons across time points. Contiguous time points exceeding an uncorrected two-sided threshold ($p < 0.05$) were grouped into candidate clusters, each summarized by its cluster mass (the sum of $|t|$ within the cluster). Phenotype labels were randomly shuffled 5,000 times (keeping group sizes fixed) to build a null distribution of the maximum cluster mass, and a corrected p-value was assigned to each observed cluster as the proportion of permutations producing an equal or larger maximum cluster mass. Given the very small minority-phenotype samples for Relax ($n=1$), the comparison was not performed for that emotion; for Happiness and Sadness the minority phenotype remained small ($n=4$), and results for these two emotions should be interpreted with corresponding caution.

As robustness checks, phenotypes were also compared using two whole-scene summary statistics per subject: the baseline-referenced mean of SAI and PAI over the full 150-second scene, and a robust (Theil-Sen) linear slope fitted to each subject's raw SAI/PAI series over the same window. Both summaries were compared between phenotypes with the same two-sided Mann-Whitney U test and rank-biserial effect size used for the cluster-based analysis.

3. Results

3.1. Phenotype prevalence

For all four primary emotions, the silhouette-optimal clustering solution identified two physiologically distinct subgroups of subjects (Table 1). Fear showed the largest heterogeneity, with two phenotypes at 24% and 76% of subjects; Happiness, Sadness, and Relax showed a majority phenotype (90–97%) and a small minority phenotype (3–10%).

Emotion	Phenotype A (%)	Phenotype B (%)
Fear	(24%)	(76%)
Happiness	(90%)	(10%)
Sadness	(90%)	(10%)
Relax	(97%)	(3%)

Table 1. Prevalence of the two silhouette-optimal autonomic phenotypes identified within each of the four primary emotions.

3.2. Temporal SAI and PAI signatures of the phenotypes

Grand-average SAI and PAI trajectories (Figure 1) characterized these subgroups at the cardiovascular level. In Fear, the two phenotypes are hereafter referred to as fight-or-flight (FoF) (24%) and freeze (76%), based on their opposing signatures: the FoF phenotype showed a marked, sustained SAI increase together with a PAI decrease below baseline, consistent with sympathetic activation and vagal withdrawal, whereas the freeze phenotype showed the opposite pattern, with SAI remaining at or below baseline and PAI increasing above it, consistent with sympathetic suppression and preserved parasympathetic tone. In Sadness, we similarly label the two phenotypes calm (90%) and stressed (10%): the stressed phenotype showed a sustained, markedly positive SAI deviation together with a predominantly negative PAI deviation for most of the scene, mirroring the sympatho-vagal pattern seen in Fear's FoF phenotype, whereas the calm phenotype showed a stable, mildly positive PAI deviation with minimal SAI engagement. In Happiness, the two phenotypes are labeled relaxed (90%) and rigid (10%): the relaxed phenotype showed a stable, mildly positive PAI deviation with SAI at or below baseline, whereas the rigid phenotype showed no comparably stable sign, consistent with a dysregulated rather than sustained autonomic profile. In Relax, the minority phenotype (a single subject) showed a predominantly negative SAI and a markedly elevated, variable PAI deviation, visually resembling a freeze-like profile, though this cannot be statistically evaluated with $n=1$; the dominant cluster (97%) remained close to baseline on both indices throughout.

The cluster-based permutation test (Table 2) confirmed

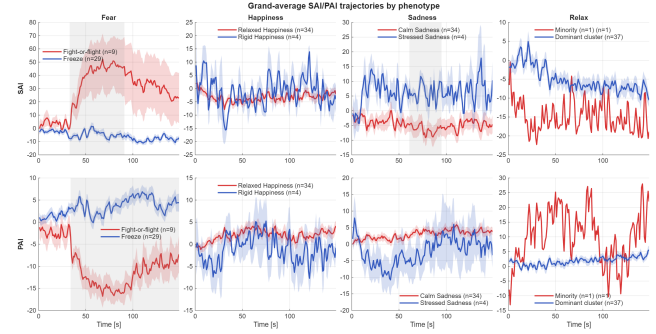


Figure 1. Grand-average instantaneous SAI (top) and PAI (bottom) time courses for the two phenotypes of each primary emotion across the 150-second scene duration.

this divergence quantitatively for Fear on both signals: SAI showed two adjacent significant clusters spanning approximately 33–91 s ($p=0.005$ and $p=0.002$), and PAI showed a single cluster spanning approximately 34–149 s (cluster mass = 574.8, $p<0.001$), together covering nearly the entire second half of the scene. Sadness SAI showed several significant clusters, the most prominent spanning approximately 61–95 s ($p=0.008$ – 0.021); given the small size of the stressed-phenotype sample ($n=4$), this result should be interpreted with caution and treated as preliminary. No supra-threshold cluster survived correction for Happiness (either signal) or Sadness PAI. Relax was not formally tested, as its minority phenotype comprised a single subject; the qualitative divergence visible in Figure 1 for that subject is reported descriptively only.

As a robustness check, whole-scene baseline-referenced means (per subject) were also compared between phenotypes. For Fear, this confirmed the cluster-based result with a markedly larger effect size on both signals (SAI: $p<0.001$, $r=-0.985$; PAI: $p<0.001$, $r=0.931$), consistent with near-complete separation between phenotype distributions. The preliminary Sadness SAI finding was similarly reinforced ($p=0.013$, $r=0.779$), though still based on $n=4$ in the stressed phenotype. In contrast, a simpler linear slope over the same window did not reach significance for any emotion or signal, indicating that phenotypes are distinguished primarily by a sustained shift in absolute autonomic level rather than by differences in the rate of change over time.

Emotion / Signal	Significant cluster (time window)	Cluster mass	p (corrected)
Fear – SAI	33–91 s (FoF > Freeze)	64.2 + 84.2	0.005 / 0.002
Fear – PAI	34–149 s (FoF < Freeze)	574.8	<0.001
Happiness – SAI/PAI	None	–	–

Sadness – SAI	61–95 s (Stressed > Calm)*	27.6–40.2	0.008–0.021
Sadness – PAI	None	–	–
Relax – SAI/PAI	Not tested (n=1 minority)	–	–

Table 2. Cluster-based permutation test results (Welch t-statistic per time point, 5,000 label permutations) comparing phenotype SAI/PAI trajectories for each primary emotion. FoF = fight-or-flight. *Sadness stressed phenotype: n=4, interpret with caution.

4. Discussion

The temporal analysis provides an independent line of evidence, based solely on instantaneous cardiac autonomic indices, that converges with the multimodal clustering solutions. For Fear, the cluster-based permutation test isolated sustained, significant divergence windows on both SAI (33–91 s) and PAI (34–149 s), together spanning nearly the entire second half of the scene. This joint sympathetic-and-parasympathetic divergence indicates that, in this protocol, the FoF and freeze phenotypes are distinguished by a coordinated, sustained shift in both autonomic branches rather than by either alone. The bifurcation into FoF and freeze phenotypes is consistent with defensive cascade theory [5], which describes an escalating repertoire of defensive responses as a function of threat proximity and perceived possibility of escape. Notably, whole-scene averages were themselves highly discriminative for Fear (Section 3.2), indicating that the phenotype distinction is not solely a matter of timing; however, only the cluster-based approach revealed that this divergence emerges gradually roughly one third into the scene rather than being present from onset — a temporal signature invisible to any single-number summary, and directly relevant to the escalating structure posited by cascade theory. These results reinforce the case, already suggested for this protocol at the level of state discrimination [3], that instantaneous SAI/PAI are more sensitive than traditional HRV to individual-level autonomic dynamics. No comparable cardiac-only signature emerged for Happiness or Relax, suggesting their phenotypic differences are carried predominantly by other channels (e.g., EEG, respiration, GSR) rather than cardiac dynamics alone. Sadness showed a preliminary SAI divergence (61–95 s) in the same sympathetic direction as Fear's FoF phenotype, but with only 4 subjects in the stressed phenotype, this should be treated as a hypothesis for future, adequately powered replication rather than a confirmed effect. Overall, Fear stands out as the emotion with the clearest, most robust cardiac-only phenotype signature in this protocol.

These findings support phenotype-aware, personalized models in affective computing and physiological emotion recognition, with applications in cardiovascular

monitoring and stress assessment. Limitations include the small minority-phenotype samples for Happiness, Sadness, and Relax and the restriction to the four primary emotions; the remaining, more complex emotions of the protocol, together with external validation on independent datasets, will be addressed in future work.

5. Conclusion

We quantified the temporal evolution of instantaneous sympatho-vagal indices for autonomic phenotypes previously identified within four primary emotions of an immersive VR protocol, providing convergent, time-resolved physiological evidence for the clustering solutions. A cluster-based permutation test isolated significant, sustained divergence on both SAI (33–91 s) and PAI (34–149 s) between the Fear fight-or-flight and freeze phenotypes, spanning nearly the entire second half of the scene, while the other three emotions showed no comparably robust cardiac-only signature. These results indicate that instantaneous cardiac autonomic dynamics alone can recover a physiologically meaningful, time-resolved phenotypic distinction within Fear, and support phenotype-aware approaches to affective computing and personalized cardiovascular monitoring.

References

- [1] R. Barbieri, E. C. Matten, A. A. Alabi, and E. N. Brown, “A point-process model of human heartbeat intervals: new definitions of heart rate and heart rate variability,” *Am. J. Physiol. Heart Circ. Physiol.*, vol. 288, no. 1, pp. H424–H435, Jan. 2005.
- [2] G. Valenza, L. Citi, J. P. Saul, and R. Barbieri, “Measures of sympathetic and parasympathetic autonomic outflow from heartbeat dynamics,” *J. Appl. Physiol.*, vol. 125, no. 1, pp. 19–39, Jul. 2018.
- [3] Mura, M., Facchini, E., Polo, E. M., Ferraris, D., Drudi, C., & Barbieri, R. (2026, July). Characterizing negative emotions through temporal autonomic coordination in virtual reality. In *2026 48th Annual International Conference of the IEEE Engineering in Medicine & Biology Society (EMBC)*. IEEE. (in press).
- [4] Maris, E., & Oostenveld, R. (2007). Nonparametric statistical testing of EEG-and MEG-data. *Journal of neuroscience methods*, 164(1), 177-190.
- [5] Roelofs, K. (2017). Freeze for action: neurobiological mechanisms in animal and human freezing. *Philosophical transactions of the royal society B: biological sciences*, 372(1718), 20160206.

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