Case-Study Guided Discovery of Calciphylaxis Biomarkers

Joshua Lee

Abstract

Calciphylaxis is a highly fatal disease that is seen through vascular calcification and skin necrosis, most often seen in dialysis patients. Despite the severity, the mechanisms underlying this field remain unclear. In this study, we examined and reported cases to evaluate the correlations between inflammatory and metabolic markers with patient outcomes. A strong positive correlation was observed between the C-reactive protein and calcium levels. Thus suggesting that systemic inflammation may promote disordered vascular calcification.
On the other hand, C-reactive protein showed us that there was a negative correlation with phosphate, surprisingly, likely due to the small sample size and data variability. The positive associations with the white blood cells, along with albumin and wound healing results, revealed that the preserved immune function and nutritional status were all linked to improving healing and survival. The findings in all of these highlight the interplay between inflammation and metabolic imbalance with the patient's overall health in calciphylaxis. Although the analysis is limited to the reliance on these case reports, the results emphasize that we would need larger studies integrating the inflammatory and nutritional markers. These effects can guide the development of therapies and improve the prognosis of patients with calciphylaxis.

Introduction

Calciphylaxis, otherwise known as calcific uremic arteriolopathy, is a rare yet debilitating condition associated with advanced renal disease, with an annual incidence rate of 35 in every 10,000 patients [1]. While the condition, presenting as vascular calcification leading to potentially life-threatening ulcerative skin lesions [1], is most often associated with patients with end-stage renal disease receiving dialysis, a subset of patients present with nonuremic calciphylaxis (NUC) where there is no underlying renal issues [1]. While the exact cause of calciphylaxis remains unknown, the presence of sparse but detailed case studies allows for the identification of putative biomarkers that can predict outcomes associated with calciphylaxis.

Given the rarity of this condition in human populations, along with sparse animal models of the disease [2], identification of clinical phenotypes and biomarkers is critical to the characterization of the pathophysiology of the condition as well as potential treatments/interventions. However, to date, most clinical research in this domain occurs in single case reports or meta-analyses. This makes it difficult to identify the risk factors and the patient's outcomes for broader clinical populations. However, careful analysis of these clinical reports makes it possible to uncover some patterns that point to the biological processes and the disease progression/outcomes.

The main aim of this project was to review patient data and perform an analysis on all of the key clinical biomarkers. By analyzing the correlations between inflammation, mineral metabolism, wound healing, and mortality, this study highlights the variables that may be biologically significant. These findings could provide a starting point for future research and may even help clinicians improve the diagnosis and monitoring of calciphylaxis.

Specifically, here I report on published patient data and perform statistical analysis of key clinical and laboratory markers. By examining correlations between inflammation, mineral metabolism, wound healing, and mortality, this study identified putative clinical biomarkers for monitoring the progression of calciphylaxis. These findings could provide starting points for future research and may eventually help clinicians improve the diagnosis, monitoring, and management of calciphylaxis.

Methods

Data was gathered through many different literature reviews of published cases and clinical studies regarding patients with calciphylaxis. All of these publications were reviewed to note down each person's clinical variables relevant to disease presentation and outcomes. The variables included patient age, sex, wound healing statuses, intact parathyroid hormone (pg/mL), serum calcium and phosphate (mg/dL), white blood cell count (µL), C-reactive protein (mg/dL), left and right ankle-brachial index, and serum albumin (g/dL).

Once collected, the data were entered into a spreadsheet where each row represented a single case or patient report, and each column corresponded to a specific clinical variable. Entries with missing data were left as 'Na' (not applicable) to allow for statistical processing while maintaining the integrity of the reported values.

After it was collected, the data were entered into a spreadsheet, and each row represented a single case report, while each column corresponded to each clinical variable. The entries with missing data were left as 'Na' (not applicable) to maintain the integrity of the reported values.

Statistical analysis was conducted through the Python library Pandas, which helped with data cleaning and organization. Pearson correlation coefficients were calculated for them and were there to help identify the relationships among all of them. This correlation quantified the strength and direction of relationships between two variables. +1 correlation indicates a perfect positive relationship, while -1 represents a perfect negative relationship, and 0 implies no correlation. By analyzing these correlations, the goal was to identify all of the markers that may be either biologically or clinically associated with all of wound healing and mortality.

Results

The correlation analysis revealed that there were several strong associations with the clinical and laboratory variables in patients with caliphylaxis. The C-reactive protein and calcium illustrated almost perfect positive correlation, it being around +0.99. This reveals that a systemic inflammation increase, calcium levels rise at the same time, while also potentially revealing underlying metabolic dysregulation. At the same time, while blood cell count and albumin showed a somewhat strong positive relationship, it was +0.66. This indicates that the patients

with stronger immune responses also exhibit better nutritional or hepatic status. White blood cell count was also positively associated with wound healing, demonstrating that immune activation may support tissue recovery.

On the other hand, a lot of negative correlations were seen. C-reactive protein and phosphate were inversely related to the coefficient close to -1.00. Thus illustrating that there might be inflammation that is suppressing the phosphate levels. Wound healing nd mortality were also inversely correlated with a coefficient of around -0.71, suggesting that poor healing is linked to higher death rates. On top of that, white blood cell count and phosphate showed a negative trend, further emphasizing the hypothesis that increased immune activity can be accompanied by lower phosphate levels.

Other variable pairs demonstrated weak or negligible correlations. For example, calcium and white blood cell count had a very weak positive relationship, with a correlation of +0.11, suggesting little to no association between these two variables in the context of calciphylaxis. Overall, these results suggest that inflammation and metabolic status may be tightly linked in calciphylaxis, with specific markers such as CRP, calcium, phosphate, and albumin providing insight into patient prognosis. While correlation does not imply causation, the strength and direction of these associations offer valuable starting points for hypothesis generation and future mechanistic studies.

Discussion:

The results of the project suggest that calciphylaxis is tied to both the inflammation and metabolic imbalance within the body. The nearly perfect positive correlation of C-reactive protein (CRP) and calcium levels indicates that inflammation may be the reason for abnormal mineral metabolism in affected patients [3]. This finding is a good idea that inflammation can affect calcium handling and cause vascular calcification [4]. On the other hand, the strong negative relationship between the CRP and phosphate levels was unexpected. This would be due to the limitations in the small number of reported cases.

Another important observation was that the positive associations between the white blood cell and albumin were, same as between the white blood cell and wound healing. This may reveal that patients with better immune response and better nutritional status are more likely to heal, as compared to ones with poor healing have a high risk of mortality, as illustrated in the correlations between healing and survival [5]. The albumin's role as a nutritional and liver function marker further supports the idea that overall patient health strongly influences calciphylaxis outcomes [6].

While all of the correlations provide valuable insights, it is good to note that correlation does not equal causation. The data was drawn from case reports along with incomplete values, and the sample size limits the ability to the accuracy of these findings. Still, identifying strong, consistent patterns across variables highlights the potential biomarkers that are worthy of further study. Future work could use larger patient datasets and mechanistic studies to test whether

inflammation directly drives the metabolic changes observed in calciphylaxis [7]. If confirmed, targeting both the inflammation and mineral imbalance can improve the outcomes of this deadly condition.

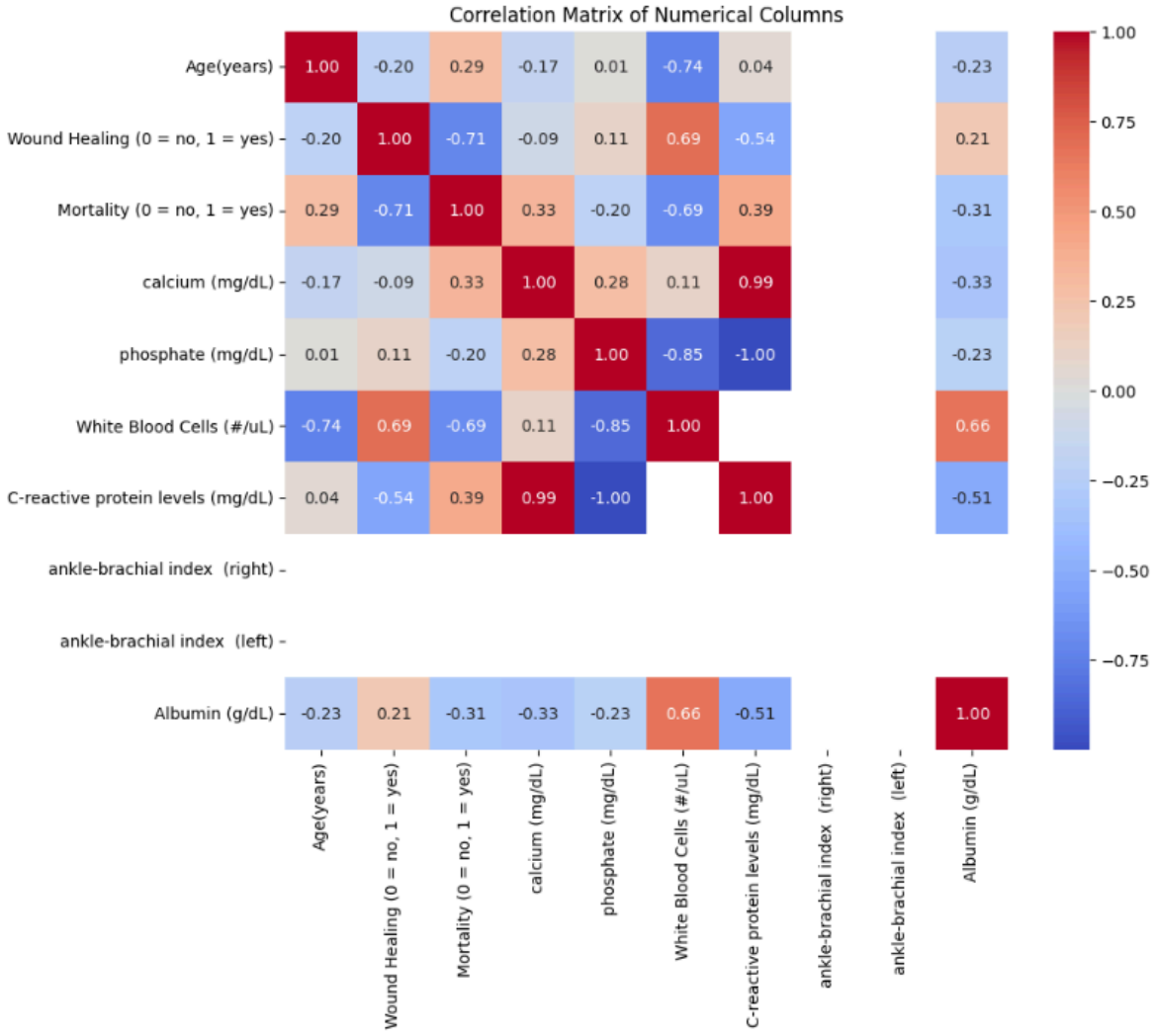

Figure 1. Correlation matrix of numerical variables.

This heatmap illustrates the pairwise Pearson correlation between clinical parameters, biochemical markers, and outcome indicators regarding calciphylaxis. All of the positive correlations would be in red while all the negatives would be in blue, with the intensity of the color reflecting the magnitude of the correlation. Strong positive correlations were seen between C-reactive proteins and serum calcium levels (r=0.99). Also seen with C-reactive protein and mortality (r = 0.39). On the other hand, phosphate showed a strong negative correlation with

C-reactive protein (r = –1.00) and white blood cell counts (r = –0.85). These associations and correlations highlight the intricate interplay between systemic inflammation and clinical outcomes, underscoring the potential relevance in the progression of calciphylaxis.

Citation	Website:	Age(years)	Gender	Wound Healing (	Mortality (0 = no	intact parathyroi	calcium (mg/dL)	phosphate (mg/c	White Blood Cell	C-reactive protei	ankle-brachial in	ankle-brachial in	Albumin (g/dL)
Miura et al.	https://pmc.ncbi.	52	F	0	1	1,250	10.4	6.4	13,200	12.77	1.13	0.94	NA
Ozdemir et al.	https://pmc.ncbi.	44	M	100	0	1431	9.2	6.8	NA	Na	Na	Na	3.9
Kurultak et al.	https://turkjineph	32	M	100	0	1487.3	9.4	7.6	17100	1.87	Na	Na	3.8
Celik et al.	https://pmc.ncbi.	32	M	100	0	Na	10.2	2.4	na	Na	Na	Na	4.2
Aihara et al.	https://www.jstag	61	F	100	0	345	5.1	7.6	14960	Na	Na	NA	1.3
Yu et al.	https://karger.co	60	M	100	0	Na	2.67	3.02	Na	Na	Na	Na	NA
Tamayo-Isla et a	https://www.tand	32	F	100	0	na	9.29	na	na	na	na	na	37
Sarkis et al.	https://pmc.ncbi.	54	M	100	100	94	na	na	na	na	na	na	na
Yeh et al.	https://link.springc	71	F	100	0	532	9.5 to 10.6	5.0 to 7.4	na	na	na	na	na
Tindni et al.	https://link.springc	60	F	0	100	2138	10.4	8	13100	na	na	na	2.2
Sprague et al	https://pmc.ncbi.	54	M	100	100	560	8.8	5.1	na	na	na	na	3.2
Gallimore et al.	https://pmc.ncbi.	74	F	100	100	na	9	4.8	na	na	na	na	na
Deng et al.	https://www.tand	46	M	100	0	330	2.21	1.4	8.5 × 10^9	na	na	na	na
Seferi et al	https://dpl6hyzq2	46	M	0	100	623	1.5 to 1.75	4,6		284.4	na	na	2.5
Kumar et al.	https://journals.s	73	M	100	0	633	8.8	11.8	na	na	na	na	na
Checherita et al.	https://www.rese	67	F	0	100	150-300	8.4 – 9.5	3.5 – 5.5	na	96	na	na	3.9
Van Rijssen et a	https://pdf.scienc	55	F	100	0	1045.95	na	na	na	na	na	na	na
Lo Monte et al.	https://biomed.p	60	F	100	0	na	9.1	5.2	na	na	na	na	na
Li et al.	https://www.tand	63	M	100	0	na	2.37	2.1–2.5	na	na	na	na	25
Shiell et al.	https://academic	74	F	0	100	na	2.11	1.87	na	na	na	na	na
Scola et al.	https://www.rese	60	F	100	0	127.2	1.5	6.58	na	na	na	na	na
Miceli et al	https://www.tand	59	F	100	100	2280	12.4	4.8	na	27.55	na	na	2.6
Cadavid et al	https://onlinelibra	44	F	100	0	1,770	7.4	7.1	na	na	na	na	3.5
Khalpey et al.	https://www.tand	66	F	100	0	216	9.0 to 10.4	2.9 to 3.7	na	na	na	na	na
Conde Rivera et	https://www.revis	55	M	100	0	537	10.1	4.5	na	na	na	na	2.2
Castro et al.	https://onlinelibra	44	F	100	0	1770	7.4	7.1	na	na	na	na	3.5
Cadavid et al.	https://www.tand	66	F	100	0	216	9-10.4	2.9 to 3.7	na	na	na	na	na
Bruccelli et al.	https://onlinelibra	78	F	100	0	620	4.83	4.83	na	na	na	na	na
Khalpey et al.	https://academic	43	M	100	0	361.5	8.5	8	na	na	na	na	3.1
Dong et al.	https://onlinelibra	56	F	100	0	737	9.7	8.4	na	na	na	na	na
Shaheen et al.	https://www.scie	44	F	100	0	75.4	8.5	5.5	17,800	na	na	na	na
Sanchez et al.	https://www.revis	66	F	0	100	653	8.6	6.6	na	na	na	na	na
Niggliazo et al.	https://www.prog	68	F	0	100	1300	na	na	na	na	na	na	na
Musso et al.	https://journals.lv	67	F	100	0	1440	9	8	na	na	na	na	na
Maroulis et al.	https://www.horr	43	M	100	0	1472.5	2.35		14220	na	na	na	na
Oikawa et al.	https://journals.s	56	M	0	100	na	9.3	6	12,300	na	na	na	3
Dean et al.	https://journals.s	66	F	100	100	913	10.9	7.4	na	na	na	na	3.3

Figure 2. Clinical and biochemical characteristics of patients with suspected or confirmed calciphylaxis reported in the literature.

The table summarizes the clinical and laboratory data from published case reports. Variables include patient age, gender, wound healing outcomes, mortality status, intact parathyroid hormone, calcium, phosphate, white blood cell counts, C-reactive protein levels, ankle–brachial indices, and serum albumin. The variability was observed across many reports regarding entries of missing ranges, reflecting the heterogeneity in data collection. Many of these patients experienced elevated parathyroid hormone levels and abnormalities in the calcium-phosphate metabolism. Inflammatory markers such as the C-reactive protein and white blood cells were often elevated. These data provided a foundation for identifying all of the potential clinical and biochemical patterns of calciphylaxis's progression.

```
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 35ms/step - accuracy: 1.0000 - loss: -1581.0757 - val_accuracy: 0.8333 - val_loss: -107.8675
Epoch 128/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -1801.5264 - val_accuracy: 0.8333 - val_loss: -109.6663
Epoch 129/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 35ms/step - accuracy: 1.0000 - loss: -1630.7883 - val_accuracy: 0.8333 - val_loss: -111.5960
Epoch 130/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -1593.7596 - val_accuracy: 0.8333 - val_loss: -112.4360
Epoch 131/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -1807.2233 - val_accuracy: 0.8333 - val_loss: -114.5451
Epoch 132/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -1813.8059 - val_accuracy: 0.8333 - val_loss: -116.7079
Epoch 133/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 35ms/step - accuracy: 1.0000 - loss: -2116.6404 - val_accuracy: 0.8333 - val_loss: -117.6117
Epoch 134/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 40ms/step - accuracy: 1.0000 - loss: -1775.5823 - val_accuracy: 0.8333 - val_loss: -121.2084
Epoch 135/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 34ms/step - accuracy: 1.0000 - loss: -1854.4688 - val_accuracy: 0.8333 - val_loss: -123.3249
Epoch 136/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 37ms/step - accuracy: 1.0000 - loss: -1905.8832 - val_accuracy: 0.8333 - val_loss: -123.0872
Epoch 137/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 38ms/step - accuracy: 1.0000 - loss: -2155.5500 - val_accuracy: 0.8333 - val_loss: -125.2173
Epoch 138/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 37ms/step - accuracy: 1.0000 - loss: -1975.8708 - val_accuracy: 0.8333 - val_loss: -127.6880
Epoch 139/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 38ms/step - accuracy: 1.0000 - loss: -1933.7924 - val_accuracy: 0.8333 - val_loss: -129.3807
Epoch 140/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 39ms/step - accuracy: 1.0000 - loss: -2026.5847 - val_accuracy: 0.8333 - val_loss: -131.2178
Epoch 141/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -1836.2827 - val_accuracy: 0.8333 - val_loss: -134.0634
Epoch 142/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 46ms/step - accuracy: 1.0000 - loss: -2242.4258 - val_accuracy: 0.8333 - val_loss: -134.7769
Epoch 143/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 35ms/step - accuracy: 1.0000 - loss: -2172.0264 - val_accuracy: 0.8333 - val_loss: -136.9354
Epoch 144/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -2116.1250 - val_accuracy: 0.8333 - val_loss: -139.2732
Epoch 145/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -2152.8025 - val_accuracy: 0.8333 - val_loss: -142.5060
Epoch 146/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 37ms/step - accuracy: 1.0000 - loss: -2243.2747 - val_accuracy: 0.8333 - val_loss: -141.1030
Epoch 147/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 36ms/step - accuracy: 1.0000 - loss: -2410.1658 - val_accuracy: 0.8333 - val_loss: -141.7852
Epoch 148/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 38ms/step - accuracy: 1.0000 - loss: -2341.2693 - val_accuracy: 0.8333 - val_loss: -145.5319
Epoch 149/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 38ms/step - accuracy: 1.0000 - loss: -2454.7949 - val_accuracy: 0.8333 - val_loss: -150.0101
Epoch 150/150
3/3 ━━━━━━━━━━━━━━━━━━━━ 0s 34ms/step - accuracy: 1.0000 - loss: -2458.7900 - val_accuracy: 0.8333 - val_loss: -150.4848
```

Figure 3. Training history of the deep learning model for calciphylaxis outcome prediction.

The figure displays training and validation metrics across epochs during model development. Model accuracy rapidly converged to 1.000 on the training set, while validation accuracy plateaued at 0.8333, suggesting potential overfitting. Training loss continued to decrease to large negative values, whereas validation loss remained relatively stable, indicating that the model fit the training data exceptionally well but generalized less effectively to unseen data. These results highlight the importance of further model optimization, such as regularization or cross-validation, to improve predictive robustness in the context of calciphylaxis risk stratification.

	Age(years)
Age(years)	1.000000
Wound Healing (0 = no, 1 = yes)	-0.199801
Mortality (0 = no, 1 = yes)	0.286396
calcium (mg/dL)	-0.171493
phosphate (mg/dL)	0.014428
White Blood Cells (#/uL)	-0.738468
C-reactive protein levels (mg/dL)	0.036326
ankle-brachial index (right)	NaN
ankle-brachial index (left)	NaN
Albumin (g/dL)	-0.228625

	Wound Healing (0 = no, 1 = yes)
Age(years)	-0.199801
Wound Healing (0 = no, 1 = yes)	1.000000
Mortality (0 = no, 1 = yes)	-0.713641
calcium (mg/dL)	-0.086984
phosphate (mg/dL)	0.106168
White Blood Cells (#/uL)	0.689749
C-reactive protein levels (mg/dL)	-0.541937
ankle-brachial index (right)	NaN
ankle-brachial index (left)	NaN
Albumin (g/dL)	0.207635

	Mortality (0 = no, 1 = yes)
Age(years)	0.286396
Wound Healing (0 = no, 1 = yes)	-0.713641
Mortality (0 = no, 1 = yes)	1.000000
calcium (mg/dL)	0.330638
phosphate (mg/dL)	-0.199126
White Blood Cells (#/uL)	-0.689749
C-reactive protein levels (mg/dL)	0.392909
ankle-brachial index (right)	NaN
ankle-brachial index (left)	NaN
Albumin (g/dL)	-0.308012

	calcium (mg/dL)	phosphate (mg/dL)
Age(years)	-0.171493	0.014428
Wound Healing (0 = no, 1 = yes)	-0.086984	0.106168
Mortality (0 = no, 1 = yes)	0.330638	-0.199126
calcium (mg/dL)	1.000000	0.276000
phosphate (mg/dL)	0.276000	1.000000
White Blood Cells (#/uL)	0.113891	-0.845128
C-reactive protein levels (mg/dL)	0.994688	-0.999989
ankle-brachial index (right)	NaN	NaN
ankle-brachial index (left)	NaN	NaN

Figure 4. Pairwise correlation coefficients between clinical and biochemical variables in patients with calciphylaxis.

The table presents the Pearson correlation values that illustrate the values among clinical trials. A strong negative correlations were observed between mortality and wound healing (r= -0.71) and between age and white blood cell counts (r= -0.73). At the same time, calcium levels were positively correlated with C-reactive protein (r= 0.99), whereas phosphate was inversely correlated with both C-reactive protein (-0.99) and white blood cells (r= -0.85). There is,

however, missing data for the ankle–brachial index values limited correlation for those variables. The findings highlight the potential mechanistic links with the mineral metabolism and clinical outcomes of calciphylaxis.

(1) Nigwekar, S. U.; Thadhani, R.; Brandenburg, V. M. Calciphylaxis. *N. Engl. J. Med.* **2018**, *378* (18), 1704–1714. https://doi.org/10.1056/NEJMra1505292.
(2) Zhao, X.; Li, L.; Ge, S.; Liu, J.; Li, S.; Wang, H.; Cai, D. A Novel Model of Dermatomyositis Induced by Membrane Antigen and Calciphylaxis in Rats. *Biomed. Rep.* **2022**, *16* (4), 31. https://doi.org/10.3892/br.2022.1514.
(3) Byers, A.; Herrera, N.; Owoyemi, I. Chronic Inflammation and Calciphylaxis. *BMJ Case Rep.* **2022**, *15* (4), e248668. https://doi.org/10.1136/bcr-2021-248668.
(4) Baby, D.; Upadhyay, M.; Joseph, Md.; Asopa, S.; Choudhury, B.; Rajguru, J.; Gupta, S. Calciphylaxis and Its Diagnosis: A Review. *J. Fam. Med. Prim. Care* **2019**, *8* (9), 2763. https://doi.org/10.4103/jfmpc.jfmpc_588_19.
(5) Souza-Tavares, H.; Miranda, C. S.; Vasques-Monteiro, I. M. L.; Sandoval, C.; Santana-Oliveira, D. A.; Silva-Veiga, F. M.; Fernandes-da-Silva, A.; Souza-Mello, V. Peroxisome Proliferator-Activated Receptors as Targets to Treat Metabolic Diseases: Focus on the Adipose Tissue, Liver, and Pancreas. *World J. Gastroenterol.* **2023**, *29* (26), 4136–4155. https://doi.org/10.3748/wjg.v29.i26.4136.
(6) Begovic, M.; Schneider, L.; Zhou, X.; Hamdani, N.; Akin, I.; El-Battrawy, I. The Role of Human-Induced Pluripotent Stem Cells in Studying Cardiac Channelopathies. *Int. J. Mol. Sci.* **2024**, *25* (22), 12034. https://doi.org/10.3390/ijms252212034.