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Nationwide real-world cohort of paroxysmal nocturnal hemoglobinuria in Chile

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Abstract

Paroxysmal nocturnal hemoglobinuria (PNH) is a rare hematological disorder caused by the expansion of a hematopoietic clone with a somatic *PIG-A* gene variant. In this retrospective analysis of 1,118 cases (2013–2024), PNH was confirmed in 224 patients (20%), including 103 adult and 81 pediatric cases. The annual incidence ranged from 0.7 to 2.6 cases per million, with a 2024 prevalence of 10 cases per million. The median age was 43 years (range 5–87), with a slight female predominance (55.8%). Aplastic/hypoplastic anemia (AA) was the most frequent referral diagnosis (43.6%), typically associated with clones < 10%. Hemolytic anemia (HA) followed (24.6%), predominantly featuring large clones (> 50%), while myelodysplastic neoplasms (MDN) accounted for 23.5%, mostly with small clones. Notably, thrombosis showed a weak association at 2.3%. With a median follow-up of 40.9 months, the 5- and 10-year overall survival (OS) rates were 69% and 62.8%, respectively. No significant difference in response to immunosuppressive therapy was observed between adult AA patients with or without PNH clones (5-year OS 66.8% vs. 63.7%, p = 0.27). Infection was the leading cause of death (51.5%), while thrombosis accounted for only 4.4%. Bone marrow failure syndromes were the most frequent clinical context associated with PNH clones, whereas classical hemolytic PNH presented the largest clone sizes. Thrombosis was an uncommon presentation and cause of death, while infection represented the leading cause of mortality. These findings suggest regional differences in the clinical expression of PNH and provide relevant evidence for future diagnostic and healthcare strategies.

Introduction

Paroxysmal Nocturnal Hemoglobinuria (PNH) is a rare, life-threatening hematological disorder caused by an acquired somatic mutation in the PIGA gene within hematopoietic stem cells. This mutation disrupts the biosynthesis of glycosylphosphatidylinositol (GPI) anchors, leading to a deficiency of complement regulatory proteins (notably CD55 and CD59) on the surface of blood cells. Without these protective proteins, cells—particularly erythrocytes—become susceptible to uncontrolled complement-mediated destruction (hemolysis). Besides intravascular hemolysis, thrombophilia, and bone marrow failure are common manifestation symptoms (1).

The global incidence of PNH is estimated at 1 case per million persons per year. However, reported rates vary significantly by region, with 5.7 cases per million in the USA, 3.5 in the UK (2), and 1.71 in the Nordic countries (3). Similarly, while the worldwide prevalence is estimated between 0.5 and 1.5 per million, it reaches 12–13 per million in the USA (2).

In Latin America, epidemiological data remain limited. Although recent studies in Brazil, Colombia, and Argentina have provided valuable local insights (4, 5, 6), population-based reference studies are still scarce, highlighting a significant gap in the region's epidemiological characterization.

The International PNH Registry, established in 2014, currently includes patients from Europe, Australia, USA, and Japan (7). For rare diseases like PNH, national registries are particularly valuable, as they facilitate the collection of standardized, multicenter data to better characterize epidemiology and clinical

outcomes. These platforms enable precise estimates of incidence and prevalence, the characterization of clinical manifestations, and the evaluation of real-world treatment patterns and survival. Furthermore, they provide essential evidence to support healthcare planning, optimize diagnostic strategies, and facilitate meaningful comparisons with international cohorts (8).

This study aims to describe the experience of a Chilean reference laboratory for PNH diagnosis over a 12-year period. We analyzed referral hematologic diagnoses, clinical PNH subtypes, clone size distribution, treatment modalities, and clinical outcomes.

Methodology

Study design

This study was a retrospective observational design. Patients were referred to the Flow Cytometry Laboratory of Hospital del Salvador from 22 healthcare institutions across the country (listed in the Appendix). The study included patients referred with a clinical suspicion of PNH over a 12-year period (2013–2024). Demographic, clinical, and laboratory data were prospectively recorded, ensuring patient anonymity. Cases with incomplete clinical information on the referral forms were excluded. The study protocol was approved by the *Comité de Ética Científico Metropolitano Oriente* (Santiago, Chile) on September 7, 2021, and conducted in accordance with the Declaration of Helsinki (1975). Written informed consent was obtained from all participants prior to sample processing.

Data collection, storage and analysis

Data were extracted from medical records and recorded in a password-protected spreadsheet with restricted access to the research team. The following variables were collected: anonymized identification code, age, sex, referral diagnosis, flow cytometry markers, confirmed diagnosis, treatment modalities, and date and cause of death. Only aggregated results were exported for analysis and manuscript preparation. The cause of death was obtained from the Death Certificates issued by the attending physician and registered at the Department of Statistics and Health Information (DEIS**)** of the Chilean Ministry of Health.

Flow cytometry method

Peripheral blood samples collected in EDTA were processed within 24 hours of collection, ensuring no evidence of hemolysis. A transfusion history for the seven days prior to sampling was recorded for each case. The analytical workflow comprised two components: erythrocyte (RBC) and leukocyte (WBC) evaluation.

For RBC analysis, 160 µL of whole blood was washed 2–3 times with FacsFlow solution (centrifuged at 1800 rpm for 5 minutes) and resuspended in 300 µL of FacsFlow. Samples were stained with

glycophorin A-FITC and CD59-PE, followed by a 15-minute incubation in the dark. After a final wash, a minimum of 1,000,000 events were acquired within the erythrocyte gate (defined by FSC/SSC).

For WBC analysis, sample volumes were calculated to achieve a minimum sensitivity of 250,000 events within the CD15 + granulocyte gate. RBCs were lysed using FacsLysing solution for 10 minutes, followed by centrifugation (2200 rpm for 6 minutes) and washing. The antibody panel included CD45-PerCP, FLAER-Alexa 488, CD24-PE, and CD15-APC. Following a 15-minute incubation and subsequent washing, samples were immediately acquired.

Data acquisition was performed on a BD FACSCanto II 8-color flow cytometer, and analysis was conducted using Infinicyt software. Clone size was determined by the percentage of GPI-deficient cells. In accordance with current clinical guidelines, neutrophil analysis was used to differentiate Type II and Type III PNH populations. (9, 10)

Diagnosis of PNH

A positive PNH case was defined by the presence of a GPI-deficient population exceeding 0.01% of total leukocytes across at least two peripheral blood lineages—neutrophils, monocytes, and/or erythrocytes (11). The 'PNH clone size' was determined by the percentage of GPI-deficient cells (9). Patients were stratified into three categories established by the International PNH Interest Group: (I) Classical PNH, characterized by clinical hemolysis and/or thrombotic events, typically with clone sizes > 50%; (II) PNH in the context of other bone marrow disorders, such as aplastic anemia (AA) or myelodysplastic neoplasms (MDN); and (III) Subclinical PNH, defined by the absence of clinical or laboratory evidence of hemolysis or thrombosis, often associated with bone marrow failure and clone sizes typically < 10% or < 1% (12). Additionally, clones were classified by size as large (> 50**%)**, intermediate (10–50%), small (1–10%), or very small (< 1%).

Analytical quality and assay performance were maintained through regular participation in the UK National External Quality Assessment Service (UK NEQAS) program (13). This external quality assessment involves the analysis of stabilized samples to ensure inter-laboratory consistency and the validation of high-sensitivity flow cytometry protocols.

Data regarding patient management were systematically recorded. For patients with AA, the response to immunosuppressive therapy (IST) or allogeneic hematopoietic stem cell transplantation (allo-HSCT) was compared between those with and without a detectable PNH clone. Five-year overall survival (OS) was estimated using the Kaplan–Meier method, and specific causes of death were documented for all cases.

Statistical analysis

To estimate PNH incidence and prevalence in Chile, the analysis utilizes a denominator based on 78% of the total Chilean population, reflecting coverage by the public health insurer, FONASA. Incidence and prevalence are calculated by dividing the number of new and existing cases by this estimated public-

sector population, which accounts for approximately 15.7 million people according to 2024 Census data (total general population x 0.78) (14).

Continuous variables were summarized as mean ± standard deviation or median and range, depending on their distribution. Intergroup comparisons of median age were performed using the Kruskal–Wallis test. A p-value < 0.05 was established as the threshold for statistical significance. All statistical analyses were conducted using GraphPad Prism version 9.0 (GraphPad Software, San Diego, CA, USA). OS was estimated using the Kaplan–Meier method, calculated from the date of diagnosis until the data cutoff on December 31, 2024, or death.

Results

Incidence and prevalence estimation

Based on data from the 2017 and 2024 Chilean population censuses conducted by the *Instituto Nacional de Estadística*s (INE), epidemiological parameters were estimated. Considering that 22 healthcare institutions contributed to this cohort, the incidence and prevalence of PNH were calculated using confirmed cases. Between 2013 and 2024, the annual incidence of PNH ranged from 0.7 to 2,6 cases per million population, while the prevalence in 2024 was estimated at 10 cases per million population (14).

PNH patient characteristics

PNH clones were confirmed in 224 of 1,118 referral patients´ blood samples (20%), including 1,037 adults and 81 children (< 15 years). Among patients diagnosed with PNH, the median age was 43 years (range, 5–87), and 55.8% were female. The distribution of PNH subtypes is summarized in Table 1. The majority of PNH cases (55.3%) were associated with bone marrow failure, both in adults and children. Among patients with classical PNH, mostly presented with hemolytic anemia, while only a minority had thrombosis. A smaller proportion of cases was associated with MDN, and a few were identified in the context of cytopenia.

Table 1
Distribution of patients in the different subtypes of PNH. AA: aplastic anemia, MDN: myelodysplastic neoplasm. AA: aplastic anemia, MDN: myelodisplastic neoplasm.

Subtypes of PNH	Total	Adults	Children
	n = 224 (%)	n = 212	n = 12
Classical PNH	43 (19.1)	43	0
Hemolytic anemia		36	0
Thrombosis		7	0
PNH associated to AA	124 (55.8)	113	11
PNH associated to MDN	37 (16.5)	37	0
Cytopenia	20 (8.9)	19	1

Table 2
summarizes PNH subtypes according to age, sex, and clone size. The median age of patients with PNH associated with MDN was higher 51 years (range, 19–86) compared with patients with classical PNH 44 years (range, 16–87) and those with PNH associated with aplastic anemia (AA) 37 years (range, 5–78), ($p < 0.005$). The proportion of female patients was highest in the MDN-associated PNH group (72.9%), followed by the other subgroups. It was clearly observed that patients diagnosed with classical PNH had the highest proportion of large clones (> 50%) (60.4%), followed by PNH associated with MDN (24.4%), and finally PNH associated with AA (10.4%). Conversely, PNH subtypes with clones < 1% were most common in PNH associated with cytopenia (95%) (Table 2).

Subtype of HPN	PNH subtypes (nº of patients)			
	Classical PNH	Associated to AA	Associated to MDN	Associated to cytopenia
	(n = 43)	(n = 124)	(n = 37)	(20)
Age, years, median (range)	44 (16–87)	37 (5–78)	51 (19–86)	49 (9–79)
Females, n (%)	16 (38)	70 (56)	27 (72.9)	12 (60)
Granulocytes (clone size)	12 (27.9)	57 (45.6)	17 (45.9)	19 (95)
≤ 1, n (%)	1 (2.3)	38 (30.4)	7 (18.9)	1 (5)
1–10, n (%)	4 (9.3)	17 (13.6)	4 (10.8)	0
10–49, n (%)	26 (60.4)	13 (10.4)	9 (24.4)	0
> 50, n (%)				

Table 2. Description of subtypes of PNH according to patient characteristics and clone size

Diagnosis of PNH and referral hematological diagnosis

Among the referral diagnoses, aplastic/hypoplastic anemia had the highest PNH positivity 44.1% (124/281), predominantly clones < 10%. It was followed by HA, 24.6% (37/150), mainly associated with large clones (> 50%), and MDN, with 23.5% (37/156), mostly involving small or very small clones. Lower positivity rates were observed in patients with anemia/cytopenia/pancytopenia: 9.3% (14/150), all with small clones (Table 3).

Table 3
Referral diagnosis of patients with PNH (n = 224).* Other: acute leukemia 2, multiple myeloma 1, myeloproliferative neoplasia 1, chronic liver disease 1. * Other: acute leukemia 2, multiple myeloma 1, myeloproliferative neoplasia 1, chronic liver disease 1.

Referral diagnosis	**n° PNH patients**	**Types of PNH clones (n°)**			
	(%)	**> 50%**	**50 – 10%**	**10 – 1%**	**< 1%**
Aplastic/hypoplastic anemia (n = 284)	124 (43.6)	13	17	37	57
Hemolytic anemia (n = 150)	37 (24.6)	25	4	1	7
Myelodysplastic neoplasm (n = 156)	37 (23.7)	9	4	7	17
Anemia/cytopenia/pancytopenia(150)	14 (9.3)	0	0	1	13
Thrombosis (n = 300)	7 (2.3)	1	0	1	5
Other (78)	*5 (6.4)				5

It is noteworthy that thrombosis, as referral diagnosis, showed a very weak association with PNH, with only 2.3% (7/300) of cases testing positive; among these, only one had a large clone, while the remainder had very small clones. Among other referral diagnoses—including leukemia (n = 28), MPN (n = 20), lymphoproliferative neoplasm (n = 12), chronic liver disease (n = 5), infection (n = 4), and chronic renal failure (n = 2), PNH clones were identified in only 6.4% (5/78) of cases, all of which were very small.

Treatments for PNH

Among patients with AA, treatment was available in 64.5% (80/124), mostly receiving immunosuppressive therapy (IST), 87.5% (70/80), while the rest underwent allo-HSCT. Two cases that failed to IST went into transplant. In cases with HA, treatment was available in 56.7% (21/37), IST was administered in 38% (8/21), eculizumab or ravulizumab in 23.8% (5/21), allo-HSCT in 14.3% (3/21), one patient received anticoagulation, another folic acid, other occasional red blood cell transfusion and three, did not receive treatment. In patients with MDN, treatment was known in 56.7% (21/37), one-half had steroids/azacitidine, 3 underwent allo-HSCT, one patient received eculizumab, one ravulizumab one iron and 4 none.

PNH patients who received an allo-HSCT had a significant improvement in OS compared with pharmacologic treatments such as IST (horse and mouse anti-thymocyte globulin, prednisone, azathioprine, cyclosporine), anticoagulants, platelet stimulators, and iron ($p < 0.018$). After 5 years, the allo-HSCT group had an overall survival probability of 83%, and other treatments group, 64%. (Fig. 1).

As far as we know, only five patients in the cohort received C5 inhibitors: three eculizumab: two with HA with partial response, and one with hypoplastic myelodysplasia with complete response. Two patients with ravulizumab with HA and MDN, both with complete response.

Overall Survival

With a median follow-up of 40.9 months (range, 16.9 to 81.3) OS of the entire PNH cohort was 69% and 62.8% at 5 and 10 years, respectively (Fig. 2A). Comparing patients among the clinical subtypes of PNH (Fig. 2B), there were no statistically significant differences in OS, with a trend toward better outcomes, in those with classical PNH. If we compared the outcome of classical PNH, AA/ PNH, and PNH/MDM with all size of clones, 5-year OS was 79.6%, 66.8%, and 62.0%, respectively, and at 10 years 56.6%, 63.6%, and 62.0%, respectively.

We analyzed if the presence of PHN clones impacted the response to treatment and survival in cases with AA. We found no difference (5-year OS: 66.8% with PNH vs. 63.7% without PHN, $p < 0.27$). (Fig. 3A). A similar finding was observed comparing those who received an allo-SCHT, with and without clones (5-year OS 82.0% vs. 70.5%, $p < 0.3$ (Fig. 3B). Regarding pediatric patients with AA, we did not observed significant difference in overall survival between the subgroup with PNH and without clone (8-year OS: 70.9% with PNH vs. 52.5% without PNH, $p < 0.7$).

Dynamics of clones.

PNH clones were longitudinally evaluated in 31 cases over varying follow-up periods. Among 20 patients with AA, 8/20 went to transplant, the clone increased in 6/8 and disappeared in 2/8. Among patients who received IST, the clone increased in 4/10, disappeared 3/10 y remained the same 4/10. No treatment information in two, 1 remained stable and the other disappeared. As a whole, the clone increased in 50% (10/20), disappeared in 25% (5/20) and remained stable in 25% (5/20). Among MDN cases, in 5/8 the clone increased, reduced in 2 and remained stable in 1. About HA, in 2 patients increased and one reduced.

Causes of death

During the 12-year data collection period, 68/224 (30.3%) patients died. The causes of death were infection (51.5%), hemorrhage (22.0%), anemia (7.3%), thrombosis (4.4%), MDN (2.9%), chronic liver disease (2.9%), and, less frequently, multiple myeloma, non-Hodgkin lymphoma, pulmonary fibrosis, graft vs host disease, thrombotic thrombocytopenic purpura and lupus, one each (8.9%) (Fig. 4).

Pediatric cases

Among pediatric patients, a PNH clone was identified in 14.8% (12/81) of cases. All clones were < 10%, and the referral diagnosis in all cases was bone marrow failure. Looking about the response to treatment, from 12 children with clones, there was information about treatment in 8: 7/8 TIS and 1/8

TPH. And from 38 children without clones, there was information about treatment in 14: 11/14 TIS and 3/14 TPH.

Discussion

This national cohort represents the first Latin American initiative aimed at characterizing patients with PNH across more than 20 healthcare institutions, providing a nationwide representation of the Chilean population. Furthermore, this study reflects the natural course of the disease prior to the eculizumab era.

This retrospective study enabled the characterization of a large patient cohort spanning most regions of the country, in accordance with international guidelines (15). The annual incidence rate remained stable over time at approximately 1 case per million inhabitants per year, aligning with international data (16; 17). The prevalence was also consistent with global estimates (2, 3, 18).

A study from Argentina reported a PNH prevalence of 22.7% when considering all clone sizes, which is similar to the 20% observed in our series (4). The median clone size differed significantly across the three clinical subtypes of PNH. In classical PNH, HA was associated with the largest clone sizes, accounting for 16% of the total cases identified in the registry. In contrast, among patients presenting with thrombosis, only seven cases (2.3%) had a detectable PNH clone; of these, only one, had a large clone (> 50%), while the remaining cases exhibited very small clones (< 1%).

The median age at diagnosis in our registry was 43.5 years (range: 5–87), which is similar to the findings of the International PNH Registry (7). This is noteworthy given the limited literature on PNH in Latin America. In contrast, two Brazilian studies reported younger populations: a retrospective study of 87 patients showed a median age of 29 years (19), while another study of 103 cases reported an even younger median age of 24.1 years (5). In our cohort, 10% of the patients were pediatric cases (defined as < 15 years old).

The referral diagnoses with the highest probability of detecting PNH clones were aplastic/hypoplastic anemia, HA, and MDN (Table 3). Conversely, the probability of clone detection was low in patients referred for thrombosis or other hematologic malignancies, such as myeloproliferative neoplasms. These findings highlight a significant discrepancy between clinical suspicion and diagnostic confirmation. Notably, thrombosis accounted for over one-quarter of all indications for testing (26.8%), followed by aplastic anemia (23.4%), MDN (13.3%), and HA (12.7%).

The International PNH Registry has reported that Asian populations experience significantly fewer thrombotic events (3.6%) compared to European and American populations (8.9%) (20). Previous studies (21) had already described a significantly lower incidence of thromboembolism (TE) in Asia. Interestingly, the low number of PNH cases (2.3%) identified among thrombosis diagnoses in our cohort may be partly explained by our genetic ancestry. In Chile, approximately 43% of the genetic makeup is of Amerindian origin—which is ultimately derived from ancestral Asian populations—while the remainder is 54% European and 2% African (22). These findings suggest that genetic factors may play a pivotal role in

the development of TE within this patient population. These results with an extremely low diagnosis of PHN in patients with thrombosis, may help to refine diagnostic algorithms across referring centers, encouraging prioritizing other causes of thrombosis in unusual sites, before considering PNH testing.

Regarding the clone dynamics over time, Pires da Silva and colleagues retrospectively analyzed 87 patients from a Brazilian single-referral center (19). They reported that PNH presentation was variable, and clone size remained stable or decreased in the majority of cases. Similarly, when Cannizzo and colleagues analyzed clone change after 1 year in 151 patients with PNH. They observed in therapy-naïve patients, that in 88.7% of cases, the clone size remained stable, whereas only 11.3% showed a significant change, increase or decrease (23). Our study differs, considering dynamics of clones, since in half of our patients the clone increased over time, disappeared in a quarter and remained stable in the rest. This finding suggests that treatment doesn't influence deeply, the nature of the disease.

Within the pediatric population, PNH was diagnosed less frequently than in adults, and all had marrow failure, similar to the literature, being HA and thrombosis uncommon, with a prevalence of small clones, similar to other studies (23, 24, 25).

Historically, OS in PNH was limited, with a median survival ranging from approximately 10 to 20 years post-diagnosis (26). Our cohort showed a 5-year OS of 69%, which is consistent with previously published data from the era of C5 complement inhibitors. While allo-HSCT remains the only curative treatment for PNH—with reported long-term survival rates of 60–80%—outcomes are heavily influenced by age, donor compatibility, and transplant-related complications (27). In our study, we observed superior outcomes in patients who underwent HSCT compared to those receiving immunosuppressive therapy (IST); however, this is likely attributed to the fact that the transplant group consisted of a significantly younger and fitter individuals.

In the context of AA, with or without PNH clones, several studies suggest that patients harboring PNH clones are more likely to respond to immunosuppressive therapy (IST) and achieve faster response rates than those lacking GPI-AP-deficient populations (28, 29, 30, 31). In contrast, our findings showed no significant difference between the two groups, aligning with observations by (32). While we noted a favorable trend in OS for pediatric patients with AA/PNH compared to those without clones, the small sample size precluded a robust statistical analysis.

In our cohort, infection was the primary cause of death, accounting for 51.5% of cases. This aligns closely with trends observed in Asian countries (40.2%; Yu, 2016) and is significantly higher than the 25.2% reported in Europe and America ($p = 0.011$). Conversely, thromboembolism is the leading cause of death among Western patients at 43.7%, a figure that far exceeds the 6.9% found in Asian populations ($P = 0.000$). In our study, thrombosis ranked only fourth, representing just 4.4% of deaths. These stark disparities further support the hypothesis that such clinical differences may be driven by underlying genetic factors.

This study has several limitations. First, treatment information was available for only 64% of the PNH cohort, which restricted our ability to analyze therapeutic responses across all cases. Second, the absence of a standardized follow-up protocol resulted in a low rate of systematic longitudinal monitoring (14%). International guidelines emphasize that PNH clones should be monitored regularly, even when small or subclinical, particularly in the context of AA and MDN. Current guidance suggests reassessing clone size every 6 to 12 months in stable patients, and more frequently in those experiencing clinical changes, treatment initiation, or disease progression (33, 34). This proactive approach is essential for the early detection of clinically significant clonal expansion, which impacts thrombosis risk, hemolysis, and therapeutic decision-making. Consequently, the lack of consistent longitudinal follow-up in our cohort may have led to an underestimation of clonal evolution and limits our ability to fully characterize the natural history of PNH in this population. Future research should incorporate standardized monitoring protocols to better define clonal kinetics and their association with clinical outcomes.

Conclusion

This nationwide real-world cohort provides the first comprehensive epidemiological characterization of PNH in Chile and one of the largest reports from Latin America. The prevalence and annual incidence observed in our cohort were consistent with international estimation. Bone marrow failure syndromes, particularly aplastic anemia represented the most frequent clinical context associated with PNH clones in both adult and pediatric populations, whereas classical hemolytic PNH was associated with the largest clone sizes. In contrast, thrombosis showed a remarkably low association with detectable PNH clones, resembling patterns previously described in Asian populations. Infection was the leading cause of death, exceeding thrombotic complications. This findings highlight potential regional differences in the clinical expression of PNH and provide valuable real-world evidence to support future diagnostic strategies, longitudinal monitoring programs and healthcare planning in Chile and Latin America.

Declarations

Conflict of interests

The authors declare that they have no conflicts of interest.

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Author Contribution

MEC and MRS made substantial contributions to the conception or design of the work, analysis, or interpretation of data. They drafted the work or revised it critically for important intellectual content; approved the version to be published; and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.SL, KF, RG, CS, LA, PC and RG was an important role in data acquisition.

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Data Availability declaration

The data that support the findings of this study are not openly available due to reasons of sensitivity and are available from the corresponding author upon reasonable request.

References

1. Bravo-Perez C, Guarnera L, Williams ND, Visconte V (2023) Paroxysmal Nocturnal Hemoglobinuria: Biology and Treatment. Med (Kaunas) 59(9):1612. 10.3390/medicina59091612
2. Waheed A, Shammo J, Dingli D (2024) Paroxysmal nocturnal hemoglobinuria: Review of the patient experience and treatment landscape. Blood Rev 64:101158. 10.1016/j.blre.2023.101158
3. Pedersen TH, Tjønnfjord E, Korkama ES, Vikman M, Ahmadi A, Overgaard UM, Kjellander C (2025) The incidence of paroxysmal nocturnal hemoglobinuria cell clones in the Nordic countries. Ann Hematol 104(8):4277–4283. 10.1007/s00277-025-06450-7. Epub 2025 Jul 9. PMID: 40632273; PMCID: PMC12431871
4. Halperin N, Altube A, Agriello E, Caride C, Galeano A, Galeazzi Z, Pavón J, López J, Palazzi J, Iommi P (2018) C Malusardi D Carelli C Rodríguez E Ledesma C Rodríguez J Correa E Ramis R Guevara V Novoa Estudio multicéntrico argentino de detección y seguimiento de clones de hemoglobinuria

paroxística nocturna (HPN) por citometría de flujo multiparamétrica (CFM) de alta resolución Hematología Vol. 22 Núm. 3

5. de Azambuja AP, Malvezzi M, Bitencourt MA, Oliveira MM, Medeiros LA, Pasquini R (2015 Mar-Apr) Paroxysmal nocturnal hemoglobinuria clone in 103 Brazilian patients: diagnosis and classification. Rev Bras Hematol Hemoter 37(2):90–97. 10.1016/j.bjhh.2015.01.001
6. Apodaca V, Gálvez EI, Wannesson KM, Scheinberg B (2025) Clinical characteristics and management of paroxysmal nocturnal haemoglobinuria in Latin America: a narrative review. Ann Hematol 104(2):867–879. 10.1007/s00277-024-05968-6
7. Schrezenmeier H, Muus P, Socié G, Szer J, Urbano-Ispizua A, Maciejewski JP, Brodsky RA, Bessler M, Kanakura Y, Rosse W, Khursigara G, Bedrosian C, Hillmen P (2014) Baseline characteristics and disease burden in patients in the International Paroxysmal Nocturnal Hemoglobinuria Registry. Haematologica 99(5):922–929. 10.3324/haematol.2013.093161
8. Parums DV (2025) A Review of the Importance and Relevance of Real-World Data and Real-World Evidence. Med Sci Monit 31:e951118. 10.12659/MSM.951118
9. Dezern AE, Borowitz MJ (2018) ICCS/ESCCA consensus *guidelines to detect GPI-deficient cells in paroxysmal nocturnal hemoglobinuria (PNH) and related disorders part 1–clinical utility. Cytometry Part B: Clinical Cytometry.* ; 94(1): 16–22
10. Richards SJ, Dickinson AJ, Newton DJ, Hillmen P (2022) Immunophenotypic assessment of PNH clones in major and minor cell lineages in the peripheral blood of patients with paroxysmal nocturnal hemoglobinuria. Cytometry Part B: Clin Cytometry 102(6):487–497. https://doi.org/10.1002/cyto.b.22094
11. Borowitz MJ, Craig FE, Digiuseppe JA, Illingworth AJ, Rosse W, Sutherland DR, Wittwer CT, Richards SJ, Clinical Cytometry Society (2010) Guidelines for the diagnosis and monitoring of paroxysmal nocturnal hemoglobinuria and related disorders by flow cytometry. Cytometry B (Clin Cytom) 78:211–230
12. Parker C, Omine M, Richards S et al (2005) International PNH Interest Group. Diagnosis and management of paroxysmal nocturnal hemoglobinuria. Blood 106(12):3699–3709
13. UK NEQAS Available in https://ukneqas.org.uk/programmes/result/?programme=paroxysmal-nocturnal-haemoglobinuria
14. Censo (2024) Instituto Nacional de Estadísticas https://censo2024.ine.gob.cl/resultados/
15. Sutherland DR, Illingworth A, Marinov I, Ortiz F, Andreasen J, Payne D, Keeney M (2018) ICCS/ESCCA consensus guidelines to detect GPI-deficient cells in paroxysmal nocturnal hemoglobinuria (PNH) and related disorders part 2–Reagent selection and assay optimization for high-sensitivity testing. Cytometry Part B: Clin Cytometry 94(1):23–48
16. Père NM, Lenaerts T, Pacheco JM (2018) David Dingli. Evolutionary dynamics of paroxysmal nocturnal hemoglobinuria. PLoS Comput Biol. ; 14(6): e1006133
17. López M, Roa, Macarena B, Cristián, Rojas C, Navarrete MA (2025) Current Perspectives on Paroxysmal Nocturnal Hemoglobinuria (PNH). Revista médica de Chile 153(11):815–824 Epub 15

de octubre de 2025. https://dx.doi.org/10.4067/s0034-98872025001100815

18. Röth A, Maciejewski J, Nishimura JI, Jain D, Weitz JI (2018) Screening and diagnostic clinical algorithm for paroxysmal nocturnal hemoglobinuria: Expert consensus. Eur J Haematol 101(1):3–11
19. Pires da Silva BGP, Fonseca NP, Catto LFB, Pereira GC, Calado RT (2022) The spectrum of paroxysmal nocturnal hemoglobinuria clinical presentation in a Brazilian single referral center. Ann Hematol 101(5):999–1007. 10.1007/s00277-022-04797-9
20. Sakurai M, Jang JH, Chou WC, Kim JS, Wilson A, Nishimura JI, Chiou TJ, Kanakura Y, Lee JW, Okamoto S (2019) Comparative study on baseline clinical characteristics of Asian versus non-Asian patients with paroxysmal nocturnal hemoglobinuria. Int J Hematol 110(4):411–418. 10.1007/s12185-019-02699-7. Epub 2019 Jul 5. PMID: 31278635
21. Yu F, Du Y, Han B (2016) A comparative analysis of clinical characteristics of patients with paroxysmal nocturnal hemoglobinuria between Asia and Europe/America. Int J Hematol 103(6):649–654. 10.1007/s12185-016-1995-1. Epub 2016 Apr 8. PMID: 27059871
22. Fuentes M, Iván P, Gallo C, María-Cátira B, Canizales-Quinteros, Samuel, Bedoya, Gabriel, González-José, Rolando, Ruiz-Linares, Andrés, Rothhammer F (2014) Geografía génica de Chile: Distribución regional de los aportes genéticos americanos, europeos y africanos. *Revista médica de Chile*, *142*(3), 281–289. https://dx.doi.org/10.4067/S0034-98872014000300001
23. Cannizzo E, Raia M, De Propris MS, Triolo A, Scarpati B, Marfia A, Stacchini A, Buccisano F, Lanza F, Regazzoli A, Michelutti A, Cesaro S, Conte CA, Vanelli L, Tedone E, Omedè P, Ciriello MM, Caporale R, Catinella V, Pantano G, De Rosa C, Lo Pardo C, Poletti G, Ulbar F, Pavanelli MC, Del Pup L, Ottaviano V, Santonocito AM, Bartocci C, Boscaro E, Arras M, Amodeo R, Mestice A, Oliva B, Ferrari L, Statuto T, D'Auria F, Pianezze G, Tanca D, Visconte F, Rubba F, Musto P, Geuna M, Gatti A, Brando B (2019) Del Vecchio L. Features, reason for testing, and changes with time of 583 paroxysmal nocturnal hemoglobinuria clones from 529 patients: a multicenter Italian study. Ann Hematol 98(5):1083–1093. 10.1007/s00277-019-03644-8
24. Donohue RE, Marcogliese AN, Sasa GS, Elghetany MT, Redkar AA, Bertuch AA, Curry CV (2018) Standardized high-sensitivity flow cytometry testing for paroxysmal nocturnal hemoglobinuria in children with acquired bone marrow failure disorders: A single center US study. Cytometry B Clin Cytom 94(4):699–704. 10.1002/cyto.b.21536. Epub 2017 Jun 27. PMID: 28574201
25. Urbano-Ispizua Á, Muus P, Schrezenmeier H, Almeida AM, Wilson A, Ware RE (2017) Different clinical characteristics of paroxysmal nocturnal hemoglobinuria in pediatric and adult patients. Haematologica 102(3):e76–e79. 10.3324/haematol.2016.151852
26. Brodsky RA (2014) Paroxysmal nocturnal hemoglobinuria. Blood 124(18):2804–2811. 10.1182/blood-2014-02-522128
27. Ussowicz M, Przystupski D, Mensah-Glanowska P, Piekarska A (2025) Current status and perspectives of hematopoietic cell transplantation in patients with paroxysmal nocturnal hemoglobinuria. Front Immunol 15:1521484. 10.3389/fimmu.2024.1521484

28. Sugimori C, Chuhjo T, Feng X et al (2006) Minor population of CD55-CD59-blood cells predicts response to immunosuppressive therapy and prognosis in patients with aplastic anemia. Blood 107(4):1308–1315
29. Fattizzo B, Pedone GL, Metafuni E et al (2021) Clinical and prognostic significance of small paroxysmal nocturnal hemoglobinuria clones in myelodysplastic syndrome and aplastic anemia. Leukemia 35(11):3223–3231
30. Ren X, Li X, Huo J et al (2020) Small PNH clones detected by fluorescent aerolysin predict a faster response to immunosuppressive therapy in patients with severe aplastic anaemia. Hematology 25(1):346–353
31. Tu J, Pan H, Li R, Wang Z, Lian Y, Li W, Shi J, Fang L (2021) PNH Clones for Aplastic Anemia with Immunosuppressive Therapy: A Systematic Review and Meta-Analysis. Acta Haematol 144(1):34–43 Epub 2020 Sep 2. PMID: 32877903
32. Yoshida N, Kobayashi R, Yabe H et al (2010) Paroxysmal nocturnal haemoglobinuria clones in children with acquired aplastic anaemia: prevalence and clinical features. Br J Haematol 150(5):581–586
33. Babushok DV (2021) When does a PNH clone have clinical significance? Hematology Am Soc Hematol Educ Program. ; 2021(1):143–152.-
34. Wong RSM, Ho Jang J, Wong LLL, Kim JS, Rojnuckarin P, Goh YT, Ueda Y, Chou WC, Lee JW, Kanakura Y, Chiou TJ (2024) Monitoring and Treatment of Paroxysmal Nocturnal Hemoglobinuria in Patients with Aplastic Anemia in Asia: An Expert Consensus. Int J Mol Sci 25(22):12160. 10.3390/ijms252212160

Figures

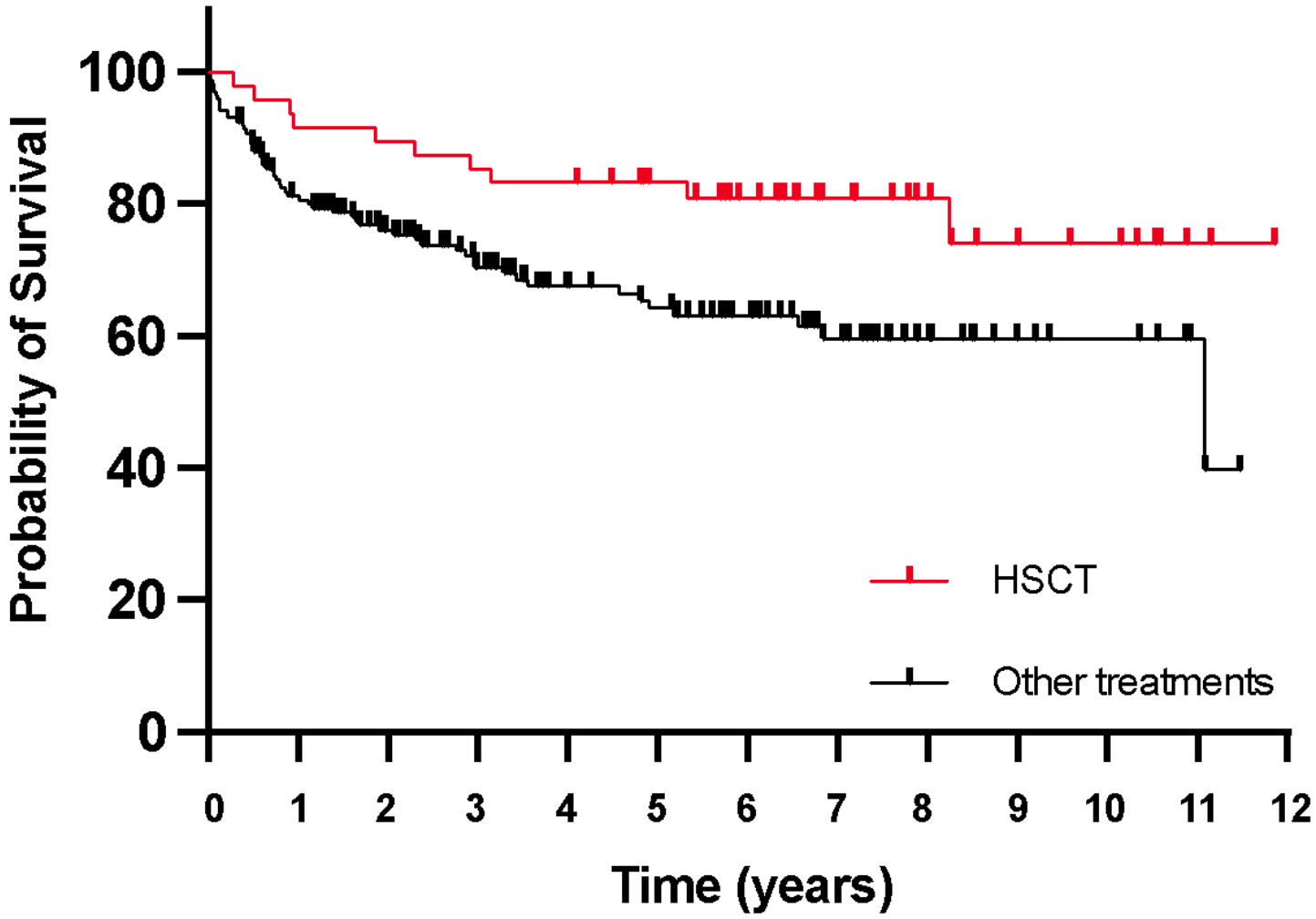


Figure 1

Kaplan-Meier survival of PNH patients that received HSCT versus IST. Immunosuppressive treatment (IST) were described in the results section.

As far as we know, only five patients in the cohort received C5 inhibitors: three eculizumab: two with HA with partial response, and one with hypoplastic myelodysplasia with complete response. Two patients with ravulizumab with HA and MDN, both with complete response.

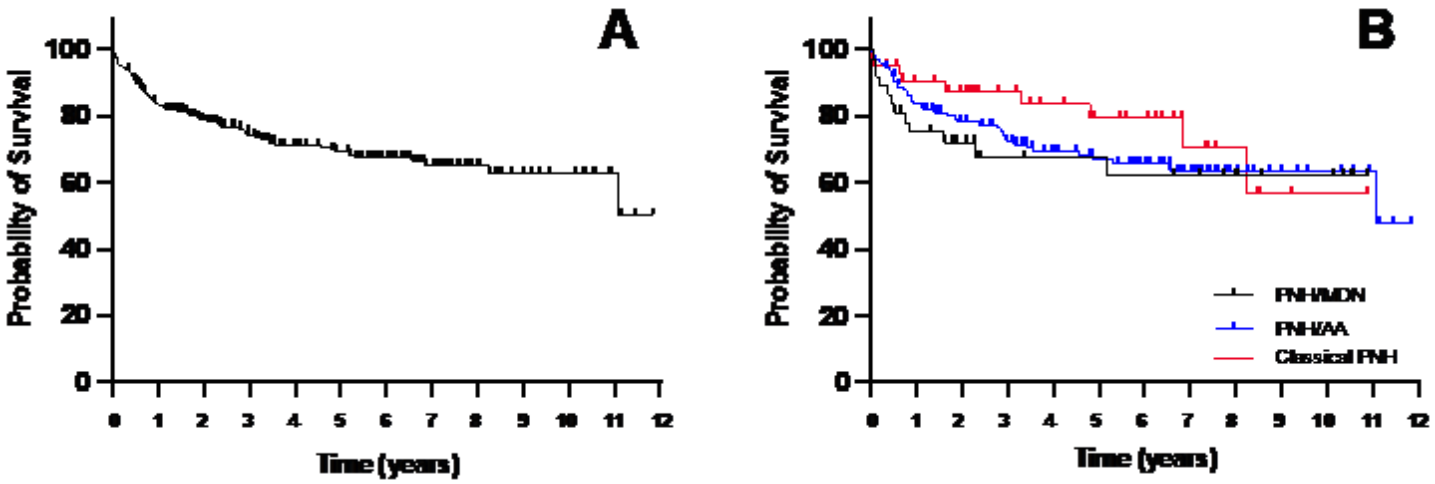


Figure 2

Kaplan-Meier survival of all PNH patients during the whole period of the registry (A) and Kaplan-Meier survival for each clinical subtype of PNH (B).

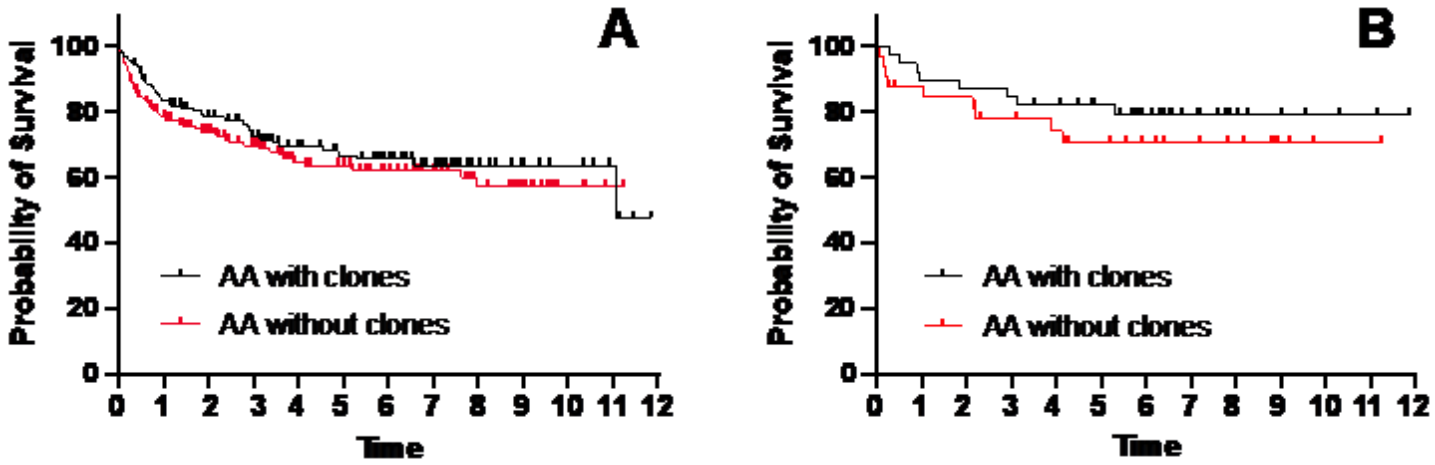


Figure 3

Kaplan-Meier survival for AA patients with or without clones. (A) Comparison of overall survival in patients considering all types of treatments and (B) comparison of overall survival in patients who received HSCT. Curve comparison was analyzed by the Log-rank test. Statistically significant was defined as $p<0.05$. PNH/AA: Patient with PNH associated to AA.

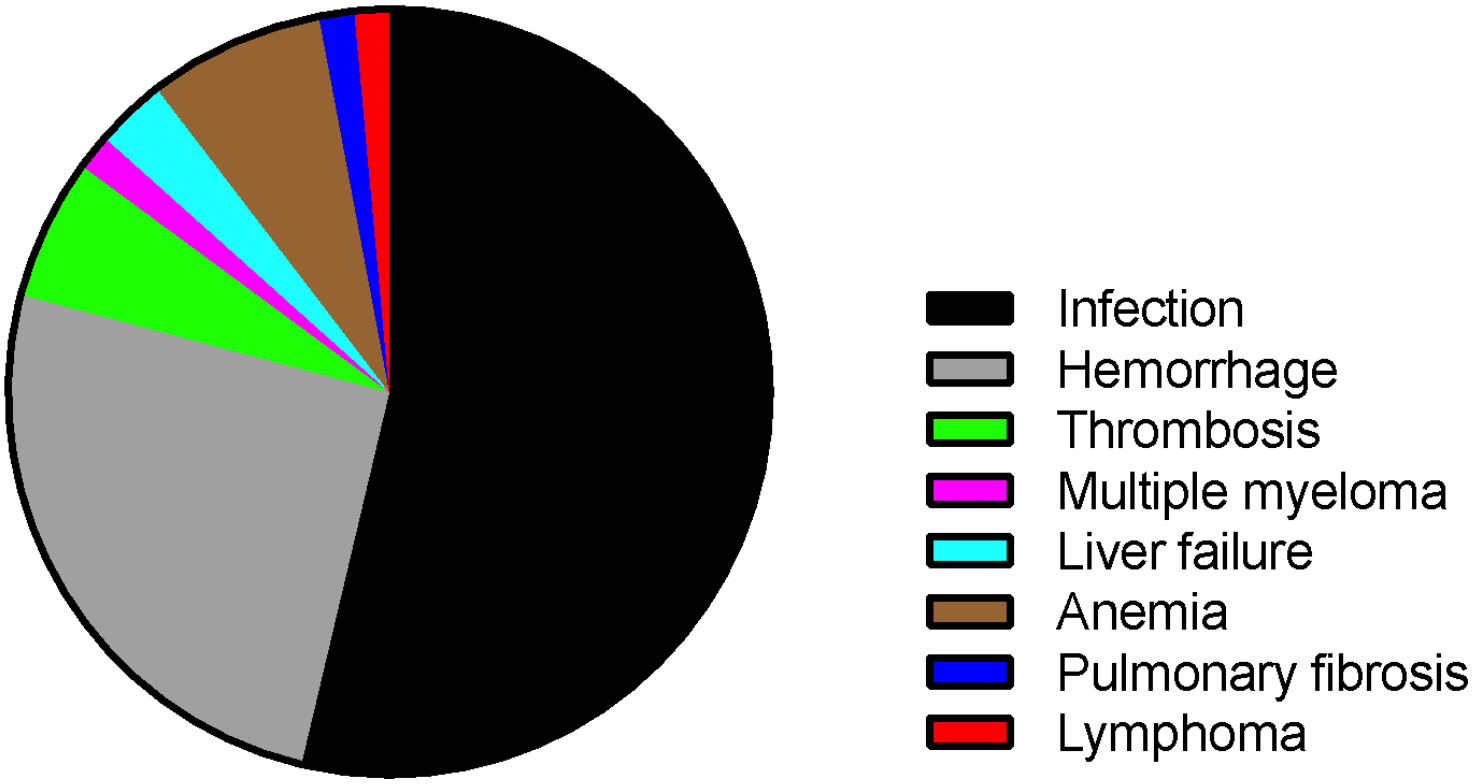


Figure 4

Distribution of causes of death among PNH patients. The pie chart illustrates the proportion of deaths attributed to infection, hemorrhage, thrombosis, and other causes within the study cohort.