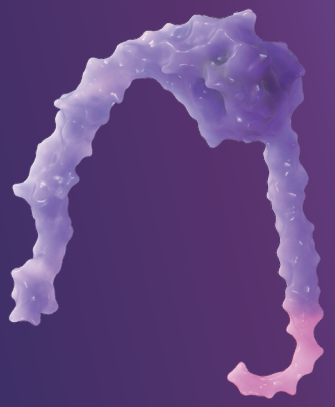
mutCALR & INCA033989 BACKGROUNDER*

**This backgrounder includes Forward-Looking Statements*



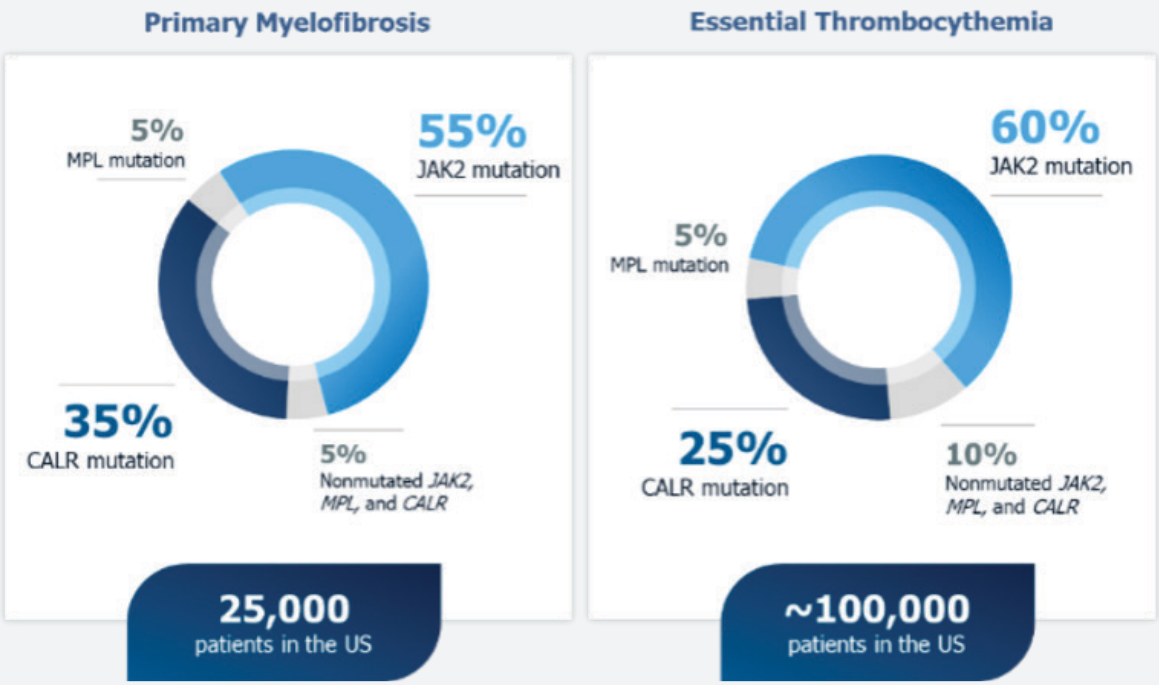
mutCALR Biology

Calreticulin (CALR) is a ubiquitously expressed intracellular protein located in the endoplasmic reticulum (ER), where it plays roles in calcium homeostasis and protein folding. In myeloproliferative neoplasms (MPNs), somatic mutations in the CALR gene arise in hematopoietic progenitor cells and act as oncogenic drivers, most notably in essential thrombocythemia (ET) and myelofibrosis (MF).[1, 2]

mutCALR Mutations

CALR mutations are oncogenic drivers in MF and ET, making them a compelling and rational target for disease-modifying therapies.

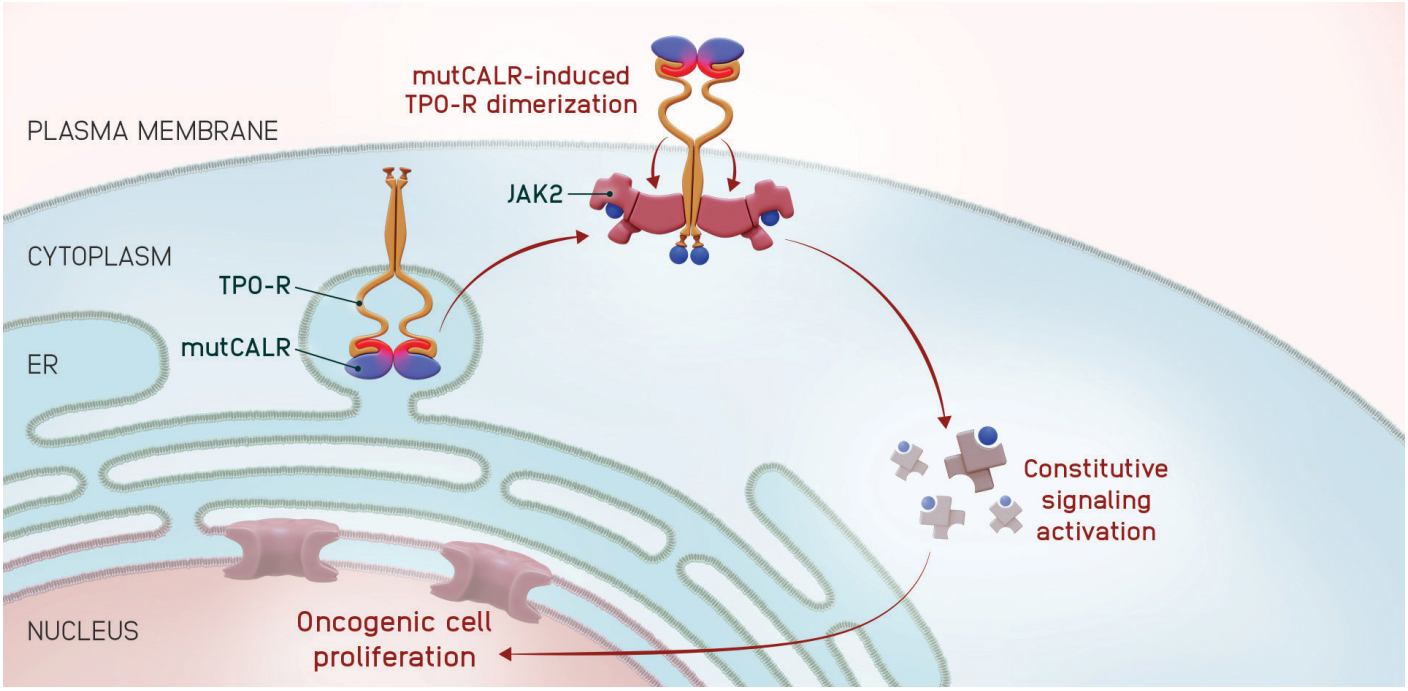
These mutations, classified as type 1 or non-type 1, result in a novel, positively charged C-terminal domain that drives their pathogenicity. Across studies, CALR mutations are found in approximately 35% of MF patients and 25% of ET patients.[1,3-5] The high prevalence of CALR mutations in these MPN subsets, combined with their distinct biology, supports CALR as a rational therapeutic target in the pursuit of disease-modifying treatments.



mutCALR as an Oncogenic Driver

mutCALR drives disease in ET and MF through aberrant activation of TPOR-JAK/STAT signaling in a TPOR-dependent process that leads to clonal expansion and abnormal hematopoiesis.

CALR mutations are established oncogenic drivers in ET and MF. Mechanistically, mutant CALR (mutCALR) gains a positively charged C-terminus that enables aberrant interaction with the thrombopoietin receptor (TPOR) within the endoplasmic reticulum. This interaction facilitates trafficking of the mutCALR-TPOR complex to the cell surface, where it triggers constitutive activation of the TPOR-JAK/STAT signaling pathway, driving uncontrolled proliferation.[6]



Critically, TPOR expression and direct binding to mutCALR are both required for this oncogenic signaling cascade.[7,8] Imbalanced mutCALR-driven JAK/STAT signaling is particularly prominent in hematopoietic stem and progenitor cells (HSPCs) and megakaryocyte precursors, where it promotes expansion of malignant clones and disrupted hematopoiesis.[9]

INCA033989 ('989) Antibody Selectivity

'989 demonstrates the potential to be disease-modifying by selectively targeting mutCALR$^+$/TPOR$^+$ disease-driving HSPCs, resulting in the suppression of clonal expansion and abnormal megakaryocyte differentiation and platelet production while sparing healthy hematopoiesis.

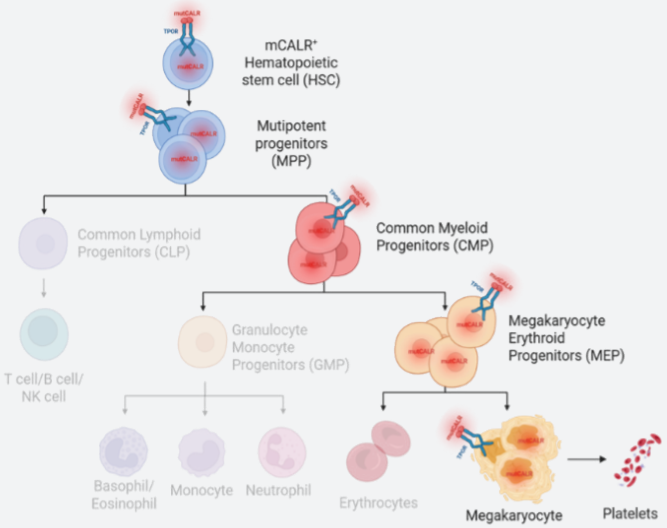
HSPCs are responsible for producing trillions of blood cells each day, and TPOR is a key regulator of this process, playing a central role in maintaining HSPC function. Additionally, TPOR plays a key role in promoting megakaryocyte differentiation and platelet production. Importantly, TPOR expression is largely restricted to the megakaryocyte lineage and is absent in all white blood cells, which contribute the majority of DNA in whole blood (WB) samples. This restricted expression profile supports the lineage specificity of TPOR signaling and provides a potential biomarker for distinguishing between disease-driving and non-pathogenic cells in CALR-mutant MPN.[8,10]

mutCALR is ubiquitously expressed throughout hematopoietic cell lineages, but it only translocates to the cell surface and functions as an oncogene when bound to TPOR. '989 specifically targets these mutCALR$^+$TPOR$^+$ cells, which include the disease-driving HSPCs in MPN. Over time, these HSPCs give rise to a large population of mutCALR$^+$ cells in the bloodstream, many of which do not express TPOR and are long-lived.

Hematopoiesis Indicating Cells With the Potential to Express mCALR

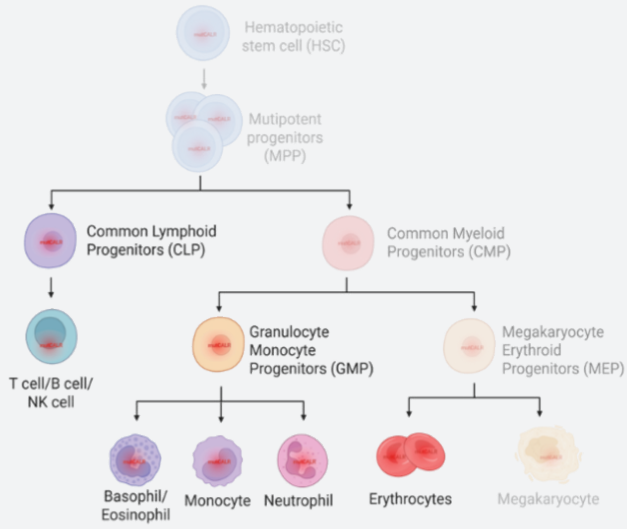
Hematopoeitic cells where mCALR and TPOR are co-expressed
(<10% of total DNA-containing blood cells)

Cells targeted by INCA033989



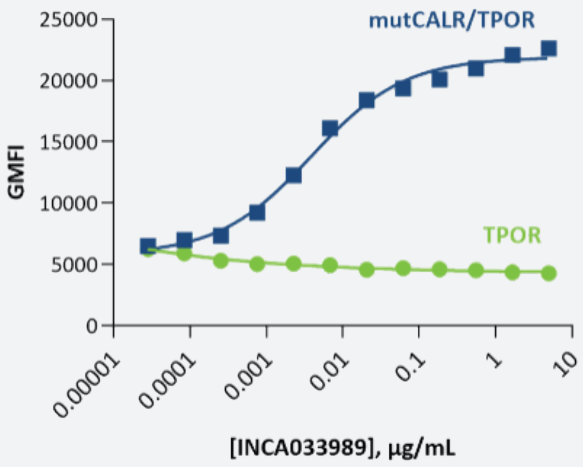
Hematopoeitic cells where mCALR may be expressed but TPOR is not
(>90% of total DNA-containing blood cells)

Cells not directly affected by INCA033989

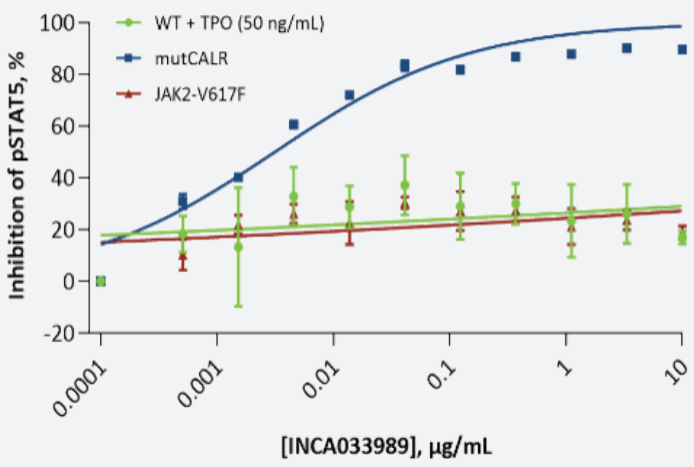


In preclinical studies, '989 has demonstrated selective activity in mutant CALR-driven disease models. In Ba/F3 cells engineered to express either the wildtype or mutant CALR, '989 selectively inhibited the proliferation of cells expressing mutant CALR, with no effect on wildtype-expressing controls. In primary CD34+ hematopoietic cells from MPN patients, treatment with '989 led to dose-dependent inhibition of phosphorylated STAT5 specifically in cells harboring CALR mutations, while sparing wildtype CD34+ cells and those from JAK2 V617F-mutated patients. This highlights the '989's mutation-selective STAT5 inhibition profile.[11]

Selective Binding of INCA033989 to mutCALR/TPOR-Expressing Cells

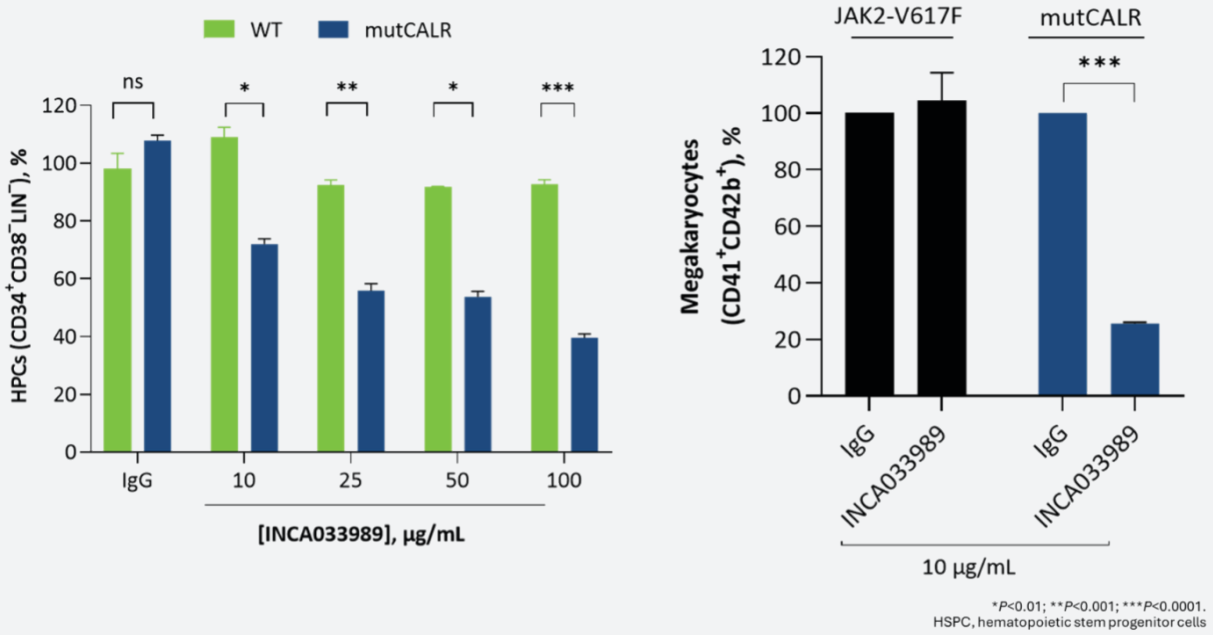


INCA033989 Selectively Inhibits pSTAT5 Signaling in CALR-Mutant CD34+ Cells



Importantly, ’989 significantly reduced the population of disease-initiating mutant CALR-positive HSPCs, while preserving wildtype progenitors. Within the megakaryocyte lineage, which is disproportionately impacted by CALR mutations, ’989 blocked differentiation of CALR-mutant CD34⁺ cells into megakaryocytes, while showing no impact on JAK2 V617F-derived megakaryocyte differentiation.[11] These findings suggest that ’989 selectively targets and suppresses the mutant CALR clone, potentially enabling disease modification in CALR-mutant MPN without disrupting normal hematopoiesis.

INCA033989 Selectively inhibits function of MPN patient-derived mutant CALR cells



*P<0.01; **P<0.001; ***P<0.0001.
HSPC, hematopoietic stem progenitor cells

Translation of Preclinical Results to Clinical Data: What to Expect

VAF reduction is an important marker of therapeutic response.

Upon treatment with efficacious doses of ’989, a reduction in mutCALR⁺/TPOR⁺ cells, including HSPCs, is expected. Over time, this targeted depletion is anticipated to result in a decrease in whole blood variant allele frequency (VAF) as the mutCALR⁺TPOR⁺ stem cells, which produce all downstream blood lineages, will be eliminated. However, VAF reduction is a lagging indicator of ’989 clinical activity due to the biological persistence of long-lived mutCALR⁺/TPOR⁻ cells, which are not directly targeted by ’989. As a result, the observed kinetics of VAF reduction reflect both the activity of ’989 against disease-driving HSPCs and the gradual clearance of these non-targeted, long-lived TPOR cells.

The kinetics of VAF reduction is affected by several biological factors:

- HSPCs comprise only ~0.01 – 0.1% of whole blood cells, while the majority of nucleated cells are WBCs, which do not express TPOR and are not direct targets of ’989
- Among WBCs, granulocytes are short-lived (days to weeks), but lymphocytes are long-lived, persisting for months to years
- Even within the myeloid lineage, mutCALR⁺ progenitors that lack TPOR expression may continue to generate mutCALR⁺ daughter cells (i.e., cells that are no longer disease-driving, but may still contribute to VAF for a period).

As a result, it may take several months or more to observe significant reductions in whole blood VAF that reflect the full therapeutic impact on mutated HSPCs. While inhibition of the mutCALR⁺ clone occurs at the early progenitor level, significant whole blood VAF decline may be delayed due to the longevity and gradual clearance of differentiated, non-targeted cells, for example, long-lived mutCALR⁺ monocytes and WBCs that do not drive disease but continue to contribute to the circulating mutCALR⁺ DNA pool.

Decreasing versus Normalizing Platelets: How ’989 is Differentiated

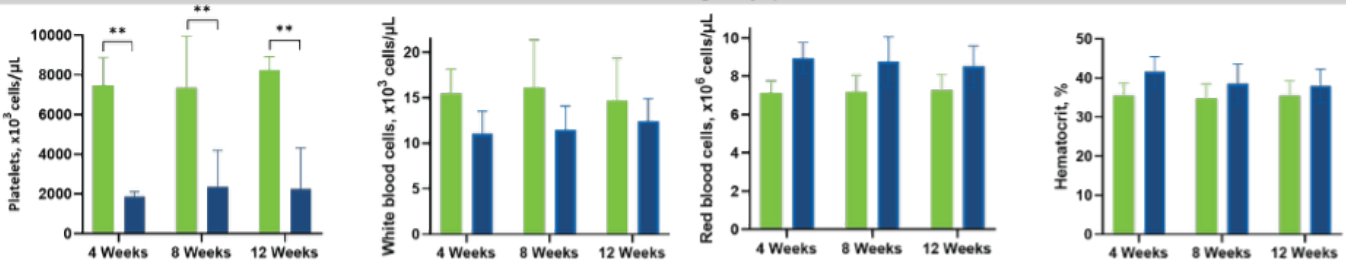
’989 has the potential to be a disease-modifying therapy by selectively correcting dysregulated megakaryopoiesis and normalizing platelet counts, unlike HU or anagrelide, which non-specifically reduce platelets, carry risks of cytopenias and require frequent dose adjustments because they do not precisely target the mutant clone.

A primary goal in the treatment of ET is the **normalization**, not just the reduction, of platelet levels.[12-14] This distinction is important:

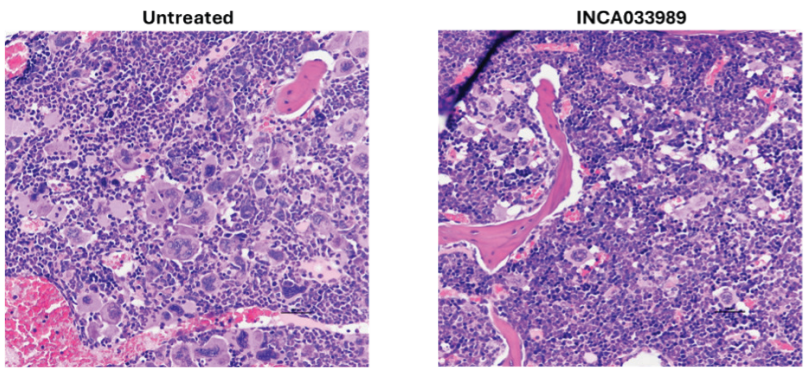
- Normalization restores platelet counts to the physiologic range by correcting the underlying dysregulation in megakaryopoiesis. Once the platelet count is normalized it should stabilize at the normal level
- Reduction is non-specific affecting both normal and mutCALR-driven megakaryopoiesis and can result in uncontrolled cytopenias and impaired hematopoiesis

’989 selectively normalizes platelet counts by targeting mutCALR^{+} -driven megakaryopoiesis and platelet production without broadly suppressing hematopoiesis or inducing cytopenias. In preclinical studies using a mutCALR knock-in (KI) genetic model, treatment with ’989 restored normal platelet counts without inducing cytopenias after just four weeks. These normalized platelet levels were sustained through week 12 and remained stable for at least four weeks after treatment cessation, suggesting a durable biological response. Additionally, ’989 treatment was shown to re-establish normal megakaryopoiesis.[11] This demonstrates that ’989 normalizes platelet production by correcting dysregulated megakaryopoiesis, as opposed to the broad suppression of hematopoiesis that is observed with conventional cytoreductive therapies.

INCA033989 re-establishes normal megakaryopoiesis in the bone marrow



INCA033989 re-establishes normal megakaryopoiesis



In contrast, hydroxyurea (HU) reduces platelet counts by inhibiting DNA synthesis across all rapidly dividing cells, including normal non-CALR mutant megakaryocyte precursors. Anagrelide, while more selective, impairs megakaryocyte maturation without addressing the mutant clone. Neither agent is disease-modifying, and both carry the risk of myelosuppression, cytopenias, and other long-term side effects with chronic use.[12-14]

*Forward-Looking Statements

Except for the historical information set forth herein, the matters set forth in this backgrounder contain predictions, estimates and other forward-looking statements, including any discussion of the potential presented by mutCALR and INCA033989. These forward-looking statements are based on Incyte's current expectations and subject to risks and uncertainties that may cause actual results to differ materially, including unanticipated developments in and risks related to: further research and development and the results of clinical trials possibly being unsuccessful or insufficient to meet applicable regulatory standards or warrant continued development; the ability to enroll sufficient numbers of subjects in clinical trials and the ability to enroll subjects in accordance with planned schedules; determinations made by the FDA, EMA and other regulatory agencies; and other risks as detailed in Incyte's reports filed with the Securities and Exchange Commission, including its annual report on form 10-K for the year ended December 31, 2024 and its report on form 10-Q for the quarter ended March 31, 2025. Incyte disclaims any intent or obligation to update these forward-looking statements.

References

1. Klampfl, T., H. Gisslinger, A. S. Harutyunyan, H. Nivarthi, E. Rumi, J. D. Milosevic, N. C. Them, T. Berg, B. Gisslinger, D. Pietra, D. Chen, G. I. Vladimer, K. Bagienski, C. Milanesi, I. C. Casetti, E. Sant'Antonio, V. Ferretti, C. Elena, F. Schischlik, C. Cleary, M. Six, M. Schalling, A. Schonegger, C. Bock, L. Malcovati, C. Pascutto, G. Superti-Furga, M. Cazzola and R. Kralovics, Somatic mutations of calreticulin in myeloproliferative neoplasms. *N Engl J Med* **2013,** 369 (25), 2379-2390.
2. Nangalia, J., C. E. Massie, E. J. Baxter, F. L. Nice, G. Gundem, D. C. Wedge, E. Avezov, J. Li, K. Kollmann, D. G. Kent, A. Aziz, A. L. Godfrey, J. Hinton, I. Martincorena, P. Van Loo, A. V. Jones, P. Guglielmelli, P. Tarpey, H. P. Harding, J. D. Fitzpatrick, C. T. Goudie, C. A. Ortmann, S. J. Loughran, K. Raine, D. R. Jones, A. P. Butler, J. W. Teague, S. O'Meara, S. McLaren, M. Bianchi, Y. Silber, D. Dimitropoulou, D. Bloxham, L. Mudie, M. Maddison, B. Robinson, C. Keohane, C. Maclean, K. Hill, K. Orchard, S. Tauro, M. Q. Du, M. Greaves, D. Bowen, B. J. P. Huntly, C. N. Harrison, N. C. P. Cross, D. Ron, A. M. Vannucchi, E. Papaemmanuil, P. J. Campbell and A. R. Green, Somatic CALR mutations in myeloproliferative neoplasms with nonmutated JAK2. *N Engl J Med* **2013,** 369 (25), 2391-2405.
3. Andrikovics, H., T. Krahling, K. Balassa, G. Halm, A. Bors, M. Koszarska, A. Batai, J. Dolgos, J. Csomor, M. Egyed, A. Sipos, P. Remenyi, A. Tordai and T. Masszi, Distinct clinical characteristics of myeloproliferative neoplasms with calreticulin mutations. *Haematologica* **2014,** 99 (7), 1184-1190.
4. Pietra, D., E. Rumi, V. V. Ferretti, C. A. Di Buduo, C. Milanesi, C. Cavalloni, E. Sant'Antonio, V. Abbonante, F. Moccia, I. C. Casetti, M. Bellini, M. C. Renna, E. Roncoroni, E. Fugazza, C. Astori, E. Boveri, V. Rosti, G. Barosi, A. Balduini and M. Cazzola, Differential clinical effects of different mutation subtypes in CALR-mutant myeloproliferative neoplasms. *Leukemia* **2016,** 30 (2), 431-438.
5. Vallapureddy, R. R., M. Mudireddy, D. Penna, T. L. Lasho, C. M. Finke, C. A. Hanson, R. P. Ketterling, K. H. Begna, N. Gangat, A. Pardanani and A. Tefferi, Leukemic transformation among 1306 patients with primary myelofibrosis: risk factors and development of a predictive model. *Blood Cancer J* **2019,** 9 (2), 12.
6. Elf, S., N. S. Abdelfattah, E. Chen, J. Perales-Paton, E. A. Rosen, A. Ko, F. Peisker, N. Florescu, S. Giannini, O. Wolach, E. A. Morgan, Z. Tothova, J. A. Losman, R. K. Schneider, F. Al-Shahrour and A. Mullally, Mutant calreticulin requires both its mutant C-terminus and the thrombopoietin receptor for oncogenic transformation. *Cancer Discov* **2016,** 6 (4), 368-381.
7. Chachoua, I., C. Pecquet, M. El-Khoury, H. Nivarthi, R. I. Albu, C. Marty, V. Gryshkova, J. P. Defour, G. Vertenoeil, A. Ngo, A. Koay, H. Raslova, P. J. Courtoy, M. L. Choong, I. Plo, W. Vainchenker, R. Kralovics and S. N. Constantinescu, Thrombopoietin receptor activation by myeloproliferative neoplasm associated calreticulin mutants. *Blood Cancer J* **2016,** 127 (10), 1325-1335.
8. Masubuchi, N., M. Araki, Y. Yang, E. Hayashi, M. Imai, Y. Edahiro, Y. Hironaka, Y. Mizukami, Y. Kihara, H. Takei, M. Nudejima, M. Koike, A. Ohsaka and N. Komatsu, Mutant calreticulin interacts with MPL in the secretion pathway for activation on the cell surface. *Leukemia* **2020,** 34 (2), 499-509.
9. Nam, A. S., K. T. Kim, R. Chaligne, F. Izzo, C. Ang, J. Taylor, R. M. Myers, G. Abu-Zeinah, R. Brand, N. D. Omans, A. Alonso, C. Sheridan, M. Mariani, X. Dai, E. Harrington, A. Pastore, J. R. Cubillos-Ruiz, W. Tam, R. Hoffman, R. Rabadan, J. M. Scandura, O. Abdel-Wahab, P. Smibert and D. A. Landau Somatic mutations and cell identity linked by genotyping of transcriptomes. *Nature* **2019,** 571 (7765), 355-360.
10. Edahiro, Y., Araki, M., Komatsu, N., Mechanism underlying the development of myeloproliferative neoplasms through mutant calreticulin. *Cancer Sci* **2020,** 111 (8), 2682–2688.
11. Reis, E. S., R. Buonpane, H. Celik, C. Marty, A. Lei, F. Jobe, M. Rupar, Y. Zhang, D. DiMatteo, R. Awdew, B. L. Ferreira, L. Leffet, L. Lu, E. Rosa, M. Evrard, G. Trivedi, B. Wass, A. Horsey, X. He, M. Covington, A. Volgina, F. Pasquier, L. Legros, G. Fouquet, W. Vainchenker, Y. O. Yang, B. Barker, J. Zhou, S. Stewart, I. S. Hitchcock, D. Dhanak, R. Macarron, I. Plo, H. Nastri and P. A. Mayes, Selective targeting of mutated calreticulin by the monoclonal antibody INCA033989 inhibits oncogenic function of MPN. *Blood* **2024,** 144 (22), 2336-2348.
12. Carturan, A., S. More, A. Poloni, S. Rupoli and E. Morsia, Shaping the Future of Myeloproliferative Neoplasm Therapy: Immune-Based Strategies and Targeted Innovations. *Cancers (Basel)* **2024,** 16 (23).
13. Tefferi, A., Vannucchi, A. M., Barbui, T., Essential thrombocythemia: 2024 update on diagnosis, risk stratification, and management. *Am J Hematol* **2024,** 99, 697-718.
14. Rumi, E., Cazzola, M. , How I treat essential thrombocythemia. *Blood* **2016,** 128 (20), 2403-2414.