WHITE PAPER

HSCT MANAGEMENT

Managing haematopoietic stem cell transplantation pre- and post-apheresis

Challenges during haematopoietic stem cell transplantation

Haematopoietic stem cell transplantation, or HSCT, is a very effective therapy for haematological malignancies, tumours and other diseases. It's also a challenging procedure for clinicians and associated with certain risks for the patients. During HSCT, patients' haematopoietic systems are suppressed by chemotherapy or radiotherapy and replaced with either stem cells previously harvested from this patient or with cells from another individual (donor). One of the treatment's benefits is that it lets one use more vigorous therapies in patients with resistant tumours. In addition, the transplanted cells themselves may have a curative effect on the patient's haematological malignancies [1, 2].

HSCT is defined by the following phases:

- Chemotherapy (in autologous HSCT) followed by stem cell mobilisation by cytokine regimens
- Collecting stem cells by apheresis
- Post-processing of the apheresis product
- Intensive chemotherapy of the patient
- Reinfusing stem cells
- Engrafting in the bone marrow after the stem cell infusion

This white paper looks at the pre- and post-apheresis stages of HSCT. It addresses the challenges and points out the added value an XN haematology analyser can deliver in this context. For more details about the 'XN Stem Cells' solution for stem cell apheresis please refer to the white paper '*Managing stem cell apheresis effectively*'.

Treating physicians face particular challenges at different stages of HSCT. During chemotherapy, the patient's own haematopoietic system is being either completely destroyed or severely compromised. This leads to severe leucopenia and depletion of the immune system, which raises the risk of infection and sepsis. The same applies to the engraftment stage – the patient has a high risk of infection until his/her own immune system has recovered. The clinical question at these stages is **'Does the patient have an infection?'**.

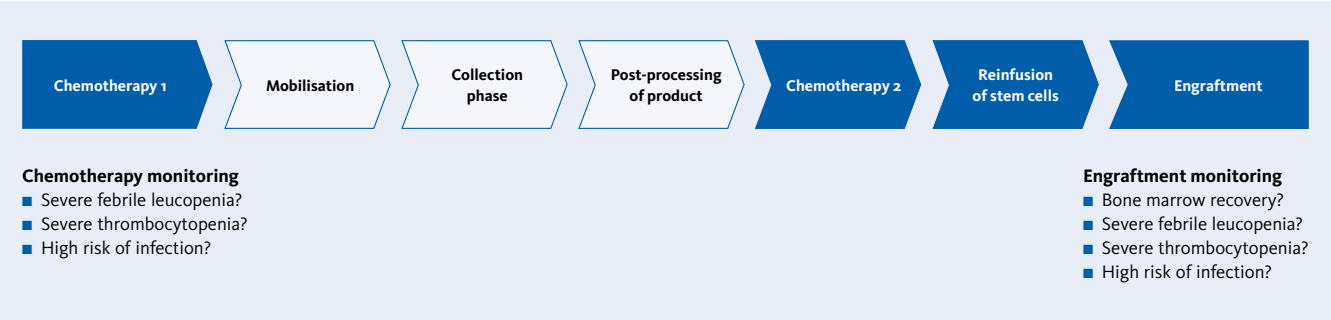

Fig. 1 *Challenges during HSCT*

Not only the leucocytes but also the megakaryocytic cell lineage is affected by chemotherapy, leading to severe thrombocytopenia. Thrombocytopenia is observed during the engraftment stage until the production of platelets has recovered. Here, the clinical question is **'Does the patient require platelet transfusion?'**.

During the engraftment stage the treating physician wants to know **'Was the stem cell transplantation successful? When will the patient be out of the risk group for bleeding and infection?'**.

Advanced applications and parameters reported by the XN haematology analyser help to address all these questions through a routine blood test, aiding clinical decision-making during HSCT.

XN solution for infection monitoring during HSCT

Patients undergoing chemotherapy are often severely leucopenic and have a high risk of infection. Data on the patients' current status can be obtained from a routine blood count by the haematology analysers of the Sysmex XN-Series by providing highly precise and reliable white blood cell count and differential results for severely leucopenic samples ('Low WBC' mode). They also offer various haematological inflammation parameters that quantify or characterise activated neutrophil and lymphocyte populations.

Several recent studies have shown that these parameters are valuable for the early detection and monitoring of infections [3 – 6]. The structural neutrophil parameters obtained from the XN-Series could predict the appearance of later-stage infection markers such as the presence of immature granulocytes [4]. Furthermore, a recent study found that both RE-LYMP (reactive lymphocytes) and AS-LYMP (antibody-synthesizing lymphocytes) counts were mainly increased in viral infections [3]. RE-LYMP counts were only increased in some bacterial infections and AS-LYMP counts were only mildly increased in bacterial infections (non-specific T-independent plasma cells).

Another study in children younger than five years found that NEUT-RI (neutrophil reactivity) was increased in patients with bacterial infections compared to controls [7]. However, only RE-LYMP and AS-LYMP counts were significantly higher in patients with viral infections than in patients with bacterial infections or in healthy individuals.

These parameters may therefore help to detect and differentiate infections so that effective countermeasures can be taken without delay. For more detailed information about the inflammation- and infection-related parameters on the XN-Series analyser please refer to the white paper '*Novel haematological parameters for rapidly monitoring the immune system response*'.

XN solution for thrombocytopenia monitoring during HSCT

During and immediately after chemotherapy, patients often have a low platelet count due to the depletion of megakaryocytes in the bone marrow. Low platelet counts are associated with a high risk of bleeding, and often require a platelet transfusion. A platelet transfusion threshold of 10,000/µL is recommended for prophylactic transfusion in stable patients, and a threshold of 20,000/µL in case of risk factors such as splenomegaly, coagulation factor deficiencies, or severe bleeding [8]. However, transfusion is associated with a higher risk of viral infection, bacterial contamination, and alloimmunisation and should therefore be avoided whenever possible [8]. It is therefore important to accurately count platelets at the transfusion trigger value (10,000 or 20,000/µL, depending on the hospital).

The XN-Series haematology analyser, which uses fluorescence flow cytometry, offers a very accurate platelet count (PLT-F), which has an excellent correlation with the 'gold standard' immune flow cytometry method [8 – 10]. It also delivers reliable platelet counts with severely thrombocytopenic patients.

Chemotherapy destroys cells in the peripheral blood and bone marrow, which significantly increases the concentration of debris in the blood. This debris does not interfere with the PLT-F platelet count, since the fluorescence flow cytometry method minimises interferences with the platelet population. Therefore, the XN's PLT-F count can provide the most accurate platelet count – also at the transfusion threshold.

XN solution for engraftment monitoring during HSCT

After transplantation, infused stem cells find their way from the blood to the bone marrow through chemotaxis and a so-called 'homing effect'. After settling in the bone marrow niche, stem cells start to divide asymmetrically to generate another stem cell and a haematopoietic progenitor. These progenitors will eventually replenish the patient's whole haematopoietic system. On average, this process takes up to two weeks, but the time of engraftment depends very much on the prior malignancy and the therapy regimen. While the haematopoietic system recovers (engraftment), the patient is very vulnerable to infections and at the risk of bleeding due to severe thrombocytopenia. Treating physicians want to know as early as possible that the engraftment has been successful and functional mature blood cells will soon be at a sufficient concentration to substantially reduce the risk of infection or bleeding. Certain parameters obtained from a blood count can be useful as markers of bone marrow activity.

Reticulocytes can be counted using fluorescence thanks to the high RNA content in immature reticulocytes. The IRF (Immature Reticulocyte Fraction) parameter indicates the fraction of the least mature reticulocytes in the blood.

The IPF (Immature Platelet Fraction) parameter indicates the ratio of immature platelets to the total number of platelets in a patient's peripheral blood. These cells were described in 1992 by Ault *et al.*, who coined the term 'reticulated platelets' to describe large, newly released platelets with elevated RNA content, whose appearance correlates with megakaryocytic activity. Reticulated platelets or IPF can be considered analogous to the reticulocytes in red cell populations.

Both IRF and IPF are markers of bone marrow activity and have been demonstrated to be predictors of bone marrow recovery after haematopoietic stem cell transplantation [11 – 13].

IRF, an indicator of erythropoiesis, showed a good correlation with neutrophil engraftment [11, 12]. This phenomenon can be explained by the fact that all cell lineages have a common progenitor. (In a similar manner, IPF correlates with engraftment not only of platelets but also of red blood cells [13].) Similar to IPF rising prior to platelet engraftment, the rise in IRF was observed five days before the neutrophil count increased (Fig. 2A). The cut-off for IRF, indicating a successful engraftment of neutrophils, ranged from 6.2 to 10 % [11, 12].

IPF is a marker of successful engraftment of the megakaryocyte lineage and a predictor of platelet recovery. The IPF count increased four to five days before the increase in the PLT count (Fig. 2B) [11, 12]. Different studies have reported various cut-off values for IPF as a predictor of successful engraftment, ranging from 3.5 to 10 % [11 – 13]. So, based on the IPF value during the engraftment stage post-HSCT, the treating physician may predict an upcoming increase in the platelet count and avoid unnecessary platelet transfusions.

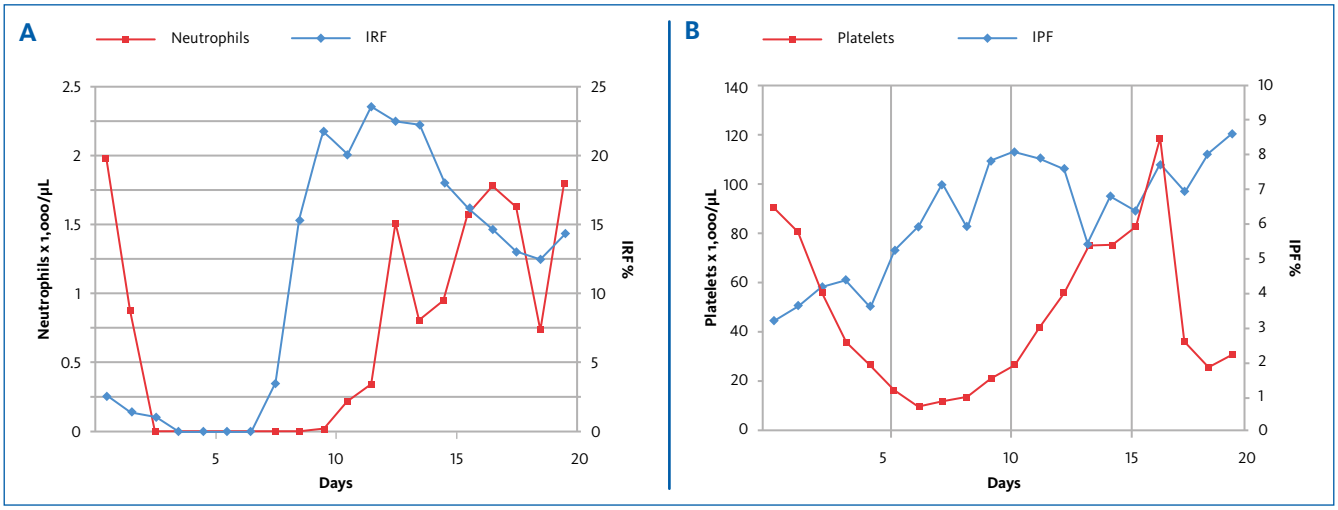

Fig. 2 *IRF and IPF as markers of bone marrow recovery after HSCT. Modified from Morkis et al. 2014 [12].*
A) IRF is a marker for neutrophil engraftment. B) IPF is a marker for platelet engraftment.

Conclusion

HSCT is an effective but challenging therapy for haematological malignancies and some other conditions. An HSCT's success depends very much on the care of the patient during the chemotherapy and recovery stage.

The XN-Series haematology analysers do not only offer a solution for optimising stem cell apheresis. They also deliver parameters to better monitor the patient throughout the therapy, including pre- and post-apheresis phases. Reliable leucocyte counts and differentials can be obtained from severely leucopenic samples thanks to a special 'Low WBC' mode. A combination of special white cell parameters allows early detection and characterisation of infections.

PLT-F measurements provide a highly accurate platelet count, also in severely thrombocytopenic samples, and so support clinical decisions about platelet transfusions. And the IPF and IRF parameters serve as early markers of engraftment of platelets and neutrophils.

All the parameters mentioned above can be obtained alongside a CBC with a routine blood test and so do not place any additional strain on the patient.

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