FACTORS INFLUENCING ENGRAFTMENT IN PERIPHERAL AUTOLOGOUS HAEMATOPOIETIC STEM CELL TRANSPLANTATION FOR LYMPHOPROLIFERATIVE DISEASE PATIENTS IN HOSPITAL UNIVERSITI SAINS MALAYSIA

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LIST OF ABBREVIATIONS

ANC	Absolute Neutrophil Count
APBSCT	Autologous Peripheral Blood Stem Cell Transplantation
CR	Complete Remission
CD	Cluster of Differentiation
DLBCL	Diffuse Large B-Cell Lymphoma
EBV	Epstein Barr Virus
EBMT	European Bone Marrow Transplant
ECOG	Eastern Cooperative Oncology Group
G-CSF	Granulocyte-Colony Stimulating Factor
GM-CSF	Granulocyte Macrophage Colony Stimulating Factor
GVHD	Graft Versus Host Disease
HCT-CI	Haematopoietic Cell Transplantation Co-morbidity Index
HLA	Human Leukocyte Antigen
HSCT	Haematopoietic Stem Cell Transplantation
HL	Hodgkin Lymphoma
HPIS	Haematology Patient Information System
HUSM	Hospital Universiti Sains Malaysia

MRD	Minimal Residual Disease
NHL	Non Hodgkin Lymphoma
OS	Overall Survival
PFS	Progression-free survival
RBC	Red Blood Cell
VGPR	Very Good Partial Response
WBC	White Blood Cell
WHO	World Health Organisation

ABSTRAK

FAKTOR-FAKTOR YANG MEMPENGARUH PEMULIHAN SEL-SEL DARAH SELEPAS PEMINDAHAN AUTOLOGOUS PERIFERAL DARAH SEL STEM UNTUK PESAKIT LIMFOPROLIFERATIF DI HOSPITAL UNIVERSITI SAINS MALAYSIA.

Pengenalan: Pemindahan Autologous Periferal Darah Sel Stem (APBSCT) adalah antara pilihan terapi yang boleh digunakan dalam pelbagai penyakit neoplastik hematologi . Di Hospital Universiti Sains Malaysia (HUSM), APBSCT digunakan untuk *multiple myeloma* dan limfoma kerana ianya dikaitkan dengan pelbagai kebaikan dan komplikasi yang kurang berbanding alogenik pemindahan darah sel stem.

Objektif: Kajian ini adalah bertujuan untuk mengenal pasti faktor-faktor yang mempengaruhi kepantasan pemulihan sel sel darah pada pesakit penyakit limfoproliferatif yang menjalani APBSCT.

Metodologi: Analisis data retrospektif dilakukan kepada semua pesakit limfoma dan *multiple myeloma* yang dirawat dengan APBSCT di HUSM dari tahun 2010 hingga 2013. Kesemua data dikumpulkan melalui pencarian di dalam fail rekod dan juga Informasi Hematologi System Pesakit (HPIS) intranet. Dua belas faktor dianalisa untuk menentukan faktor apa yang mempengaruhi kepantasan pemulihan untuk platelet dan neutrofil. Definisi pemulihan neutrofil ialah hari pertama dari tiga hari berturut-turut dimana jumlah neutrofil melebihi $\geq 0.5 \times 10^9$/L tanpa sokongan transfusi dan definisi untuk platelet ialah hari

pertama dari tujuh hari berturut-turut dimana jumlah platelet melebihi $\geq 20 \times 10^9$/L dimana tiada transfusi platelet diberi. Kepantasan pemulihan untuk platelet dan neutrofil didefinasikan sebagai sama atau kurang dari 14 hari dari hari transfusi darah sel stem. Data dianalisa dengan menggunakan Pearson Chi Square/Fisher's Exact test dan kemudian, faktor-faktor yang signifikan dianalisa dengan menggunakan Multiple Logistic Regression test. Ujian statistik dengan nilai $p < 0.05$ adalah menunjukkan statistik yang signifikan.

Hasil Kajian: Daripada 40 pesakit, keputusan kajian menunjukkan bahawa 95% pesakit telah mencapai pemulihan neutrofil yang berjaya dan 82.5% pesakit mempunyai pemulihan platlet yang berjaya. Dari dua kumpulan yang berjaya mencapai pemulihan ini, faktor utama yang mempengaruhi kecepatan pemulihan untuk neutrofil adalah jumlah bilangan stem sel ($CD34^+$) dan faktor-faktor yang mempengaruhi kecepatan pemulihan platlet adalah jumlah bilangan stem sel ($CD34^+$) dan tahap penyakit tersebut pada masa diagnosis.

Kesimpulan: Mengenal pasti faktor-faktor yang boleh memberi manfaat kepada pesakit yang menjalani pemindahan sel stem adalah penting dan hasil kajian yang diperoleh dapat digunakan sebagai panduan kepada pakar perubatan klinikal untuk mengoptimumkan pengurusan rawatan pesakit mereka.

ABSTRACT

FACTORS INFLUENCING ENGRAFTMENT IN AUTOLOGOUS PERIPHERAL BLOOD STEM CELL TRANSPLANTATION FOR LYMPHOPROLIFERATIVE DISEASE PATIENTS IN HOSPITAL UNVERSITI SAINS MALAYSIA.

Introduction: Autologous peripheral blood stem cells transplantation (APBSCT) is a therapeutic option which can be used in various haematological neoplastic disorders. In Hospital Universiti Sains Malaysia (HUSM), APBSCT is practiced in multiple myeloma and lymphoma cases as it is associated with substantial advantages and fewer complications compared to allogenic stem cell transplantation.

Objectives: The aim of this study was to identify factors associated with rapid haematopoietic engraftment in lymphoproliferative disease patients undergoing APBSCT.

Methodology: A retrospective data analysis was done on lymphoma and multiple myeloma patients treated with APBSCT at HUSM from 2010 until 2013.Data was collected from the record file Haematology Patient Information System (HPIS) in the institution intranet. Twelve factors were analyzed to determine the engraftment time for platelet and neutrophil. Engraftment of neutrophils is defined as count at least 0.5×10^9/L for three consecutive days and platelet count at least 20×10^9/L for seven consecutive days without transfusion support. Early engraftment for both neutrophils and platelet were defined as less than 14 days from the day of stem cell infusion. Data were analyzed using Pearson Chi Square/ Fisher's Exact and significant factors were analysed with Multiple Logistic Regression test with $p < 0.05$ indicates statistically significance.

Results: Forty patients had been studied, the results showed that 95% patients had successful neutrophil engraftment and 82.5% patients had successful platelet engraftment. From these success groups, the main predictive factor for rapid engraftment for neutrophil is stem cell (CD34+) count while predictive factors that influenced rapid platelet engraftment were (CD34+) count and the stage of the disease at diagnosis.

Conclusion: In conclusion, identifying factors that may benefit patients undergoing stem cell transplantation are important and the results obtained can be useful as a guidance to clinical haematologists to optimize their management.

CHAPTER 1

INTRODUCTION

1.0 GENERAL INTRODUCTION

A stem cell transplantation is a specialized procedure where it is done with the aim of eliminating the cancer cells with high dose chemotherapy. After completing the treatment, stem cells that were collected earlier were re-infused into the patients and homing of the stem cells will then occur naturally. The stem cells infused can overcome the toxicities that are caused by the myeloablative dose of chemotherapy or radiotherapy given. The main purpose of bone marrow stem cell transplantation is to provide cure of the disease.

The source of stem cells can be acquired through bone marrow aspirate, peripheral blood or from the cord blood. However nowadays, stem cells taken from the peripheral blood is the first choice since it is not that invasive compared to bone marrow aspirate and the enough amount of stem cells can be collected compared with stem cells collected from the cord blood where usually it has low number of enough stem cells.

There are three types of stem cell transplantation namely allogeneic transplant, autologous transplant and syngeneic transplant. In allogeneic transplant, it involves harvesting stem cells from a genetically similar donor or another name of it is Human Leucocytes Antigen (HLA) matched donor. HLA matched can be from related donor (found within family members) or from unrelated donor. The autologous transplant involves harvesting stem

cells from the patient itself and this stem cells will be infused back into the donor's blood for homing into the bone marrow. The syngeneic transplant involves using graft or stem cells from an identical twin.

Many diseases now can be cured by doing bone marrow stem cell transplantation. Example of diseases that are proven to be effective with autologous stem cell transplantation can be divided into malignant and non-malignant diseases. For malignant diseases, examples are non-Hodgkin lymphoma, Hodgkin lymphoma, multiple myeloma, acute myeloid leukaemia, neuroblastoma and etc. and for non malignant diseases, examples are amyloidosis and autoimmune diseases (Collagen disease). In Hospital Universiti Sains Malaysia (HUSM), there are two main diseases treated with autologous peripheral blood stem cell transplantation (APBSCT) namely the lymphoma (includes Hodgkin and non-Hodgkin) and multiple myeloma. Both are members of a big group of diseases called lymphoproliferative disease.

Previous, when planning for autologous Haematopoietic Stem Cell Transplantation (HSCT), sources of the stem cells were taken from the bone marrow but currently, most institutions have moved to autologous peripheral HSCT as it gives less stress to the patient where patient do not require general anaesthesia which is used in bone marrow transplantation procedure. Besides, in autologous peripheral HSCT it is associated with

advantages such as faster haematological engraftment and the treatment related cost is low compared to bone marrow transplantation (Hartmann *et al.*, 1997).

So far in HUSM, no study has been conducted to see the successful of engraftment in our lymphoproliferative patients (multiple myeloma and lymphoma). By conducting this study, it is hope that results that are obtained may be useful as a guidance to the clinical haematologist to predict and choose which patient that may benefit from undergoing stem cell transplantation and optimize their individual management. Besides, this study may indirectly assess the quality of our stem cell mobilizing procedure and processing in the Stem Cell Transplantation Laboratory Unit, HUSM.

CHAPTER 2

LITERATURE REVIEW

2.0 LITERATURE REVIEW

2.1 OVERVIEW OF STEM CELL TRANSPLANTATION IN MALAYSIA

The first bone marrow transplant conducted in Malaysia was in 1987. Since then, the number has been increased steadily over the years. The latest data that are available from the National Transplant Registry showed that from 1987 until 2011, the total number up to the date were 2260 patients. These procedures were done in ten transplant centres which were available in Malaysia; three of which were university based, three were government based and the remaining four were private hospitals. From these ten centres, three are specialized in paediatric transplantation and others are for adult transplantation which most of them conducting allogeniec transplantation. Hospital Ampang has the highest number of procedures among all. This is probably because it is the main tertiary referral centre in the country. Table 2.1 shows the list of transplant centres in Malaysia.

Table 2.1: Transplant centres in Malaysia (Adapted from National Transplant Registry, 2011)

Transplant Centre Category	Name of Transplant Centre
University based	1. Hospital Universiti Kebangsaan Malaysia
	2. Hospital Universiti Sains Malaysia
	3. University of Malaya Medical Centre (Adult)
	4. University of Malaya Medical Centre (Pediatric)
Government based	1. Hospital Kuala Lumpur, (Adult)
	2. Hospital Kuala Lumpur, Institute Pediatrics (Pediatric)
	3. Hospital Ampang
	4. Hospital Pulau Pinang
Private based	1. Sime Darby Medical Centre, Subang Jaya (Adult)
	2. Sime Darby Medical Centre, Subang Jaya (Pediatric)
	3. Gleneagles Medical Centre, Penang
	4. Lam Wah Ee Hospital, Penang
	5. Ampang Puteri Specialist Hospital

Many diseases can be treated with stem cell transplantation. Diseases that are being treated with stem cell transplantation in Malaysia can be categorized into malignant and non-malignant. Table 2.2 shows the list of the diseases.

Table 2.2: Diseases treated with stem cell transplantation in Malaysia (Source from national Transplantation Registry 2011).

Malignant	Non- malignant
Acute leukaemia • Acute leukaemia, unclassified • Acute undifferentiated leukaemia • Acute Lymphocytic Leukaemia (ALL) • Acute Myelogenous Leukaemia (AML) denovo • AML post-chemotherapy • AML post-MDS	Hypoplasticanaemia • Aplastic anaemia • Fanconi anaemia
Chronic leukaemia • Chronic lymphocytic leukaemia • Chronic myeloid leukaemia	Erythrocytic Disorders • Diamond-Blackfan anaemia • Congenital Dyserythropoeitic Anaemia (CDA)
Lymphoma • Hodgkin lymphoma • Non-Hodgkin lymphoma, Aggressive • Non-Hodgkin lymphoma, Indolent	Haemoglobinopathy • Thalassaemia major • Sickle Cell Anaemia
Solid tumors • Carcinoma, breast • Carcinoma, ovary • Germ Cell Tumour (GCT)-testicular • GCT-primary non-testis • Ewing sarcoma • Glioma • Hepatoblastoma • Neuroblastoma • Rhabdomyosarcoma • Soft tissue sarcoma (non-RMS) • Wilms tumour • Primitive NeuroectodermalTumour (NET)	Others • Haemophagocytic Lymphohistiocytosis Syndrome • Congenital Immunodeficiencies Others • Osteopetrosis
Myelodysplasia • Juvenile Myelomonocytic Leukaemia • Myelodyplastic syndrome (MDS) • Myelofibrosis	
Multiple Myeloma	

Overall, from 1987 until 2011, there was a slight male preponderance with 56% male patients and 44% female patients. Chinese is the largest ethnic group that were involved in transplantation (43%) followed by the Malays (40%) and the Indians (8%). The youngest age undergone for transplantation was at one month and the oldest age was at 72 years old in this country. Looking at the age category, 47% patient's age below 19 years old were transplanted. Fifty-one percent patients were transplanted in the age group of 20-59 years and patients older than 60 years old constitute the minority group with only 2%.

The two most popular type of transplantation performed were allogeneic and syngeneic transplantation with 61 % and the rest was autologous. Most of allogeneic transplantation was done on malignant diseases and majority falls under haematological malignancy. For example, acute leukaemia remains the top disease for transplantation with 37% followed with lymphomas (23%) and chronic myeloid leukaemia (9%). For non-malignant diseases, haemoglobinopathies constitute about 8% of cases. The percentage of chronic myeloid leukaemia planned for allogeneic transplantation were not that high compared to other malignant haematological disease perhaps due to the advances in the used of novel drugs such as the first generation tyrosine kinase inhibitor (Imatinib) or second generation (Nalotinib).

The sources of stem cell transplant were harvested mostly from peripheral (70%). Bone marrow source is used in 26% of cases and cord blood is the less popular with 4% of cases.

The top three causes of death in transplanted patients were the underlying illness itself (59%), sepsis (17%) and graft versus host disease (8%). Other less common causes include haemorrhage, veno-occlusive disease, organ failure, interstitial pneumonitis and the remaining 5% of cases were unknown.

The current problem is not every patient can afford for stem cell transplantation. The cost for allogeniec transplantation is quite high compared to autologous transplantation. Only three government hospitals provide transplant services on subsidized basis and the procedure depends on limited financial budget. Unlike in the university hospital, there are social welfare and non government organizations that will help in finding financial resources in order to help patients in need.

2.2 HAEMATOPOIETIC STEM CELL TRANSPLANTATION

2.2.1 Haematopoietic Stem Cell

Haematopoietic stem cells are cells that are capable to differentiate into red cells, white cells (neutrophil, monocyte, lymphocyte, basophil and eosinophil) and platelet. They have the ability of self renewal and they are located in the bone marrow in a small number. They can be identified by immunoflowcytometry where they are positive for $CD34^{+}$. Figure 2.1 shows schematic cartoon on how stem cells are able to differentiate into various cell lineages.

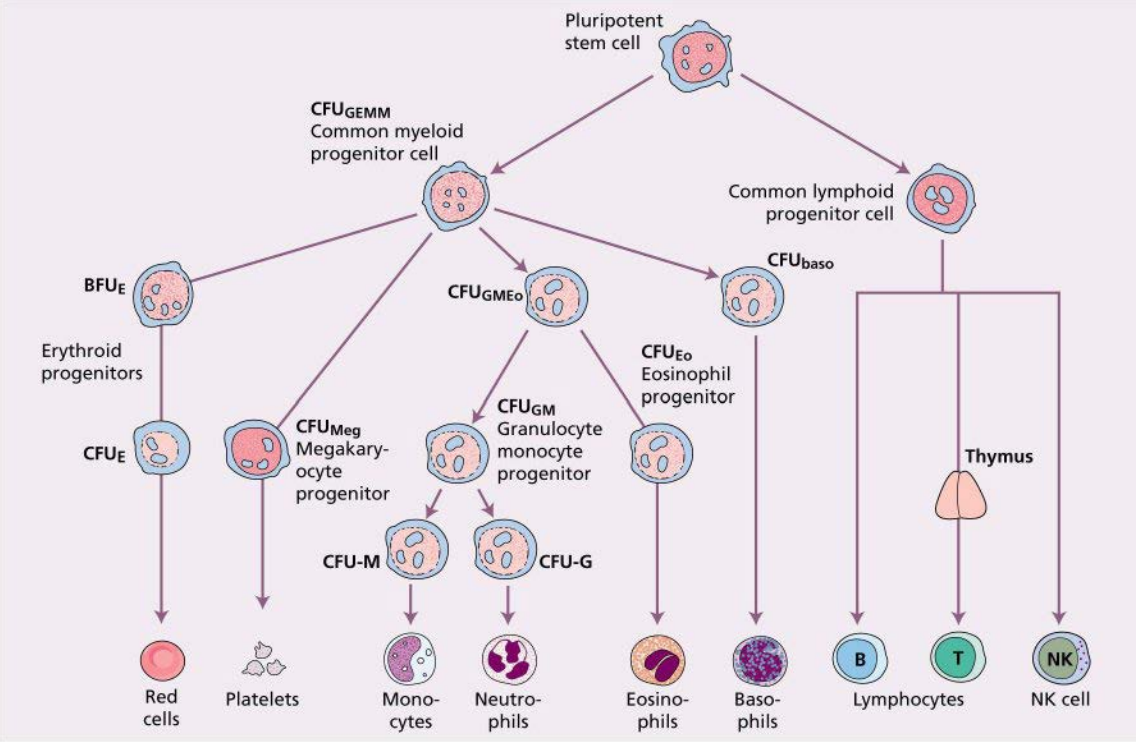

Figure 2.1: Diagrammatic presentation of the capability of the bone marrow pluripotent stem cell to differentiate into different cell lineages (Source: Essential Haematology, 6th edition, 2011)

2.2.2 Types of Haematopoietic Stem Cell Transplantation

Before transplantation, patient is given chemotherapy , radiotherapy or both in combination to destroy remaining malignant cells in the body including in the bone marrow. During this process (myoablative), all the healthy cells in the marrow are also destroyed and this will allow a new place for the transfused stem cells to grow or engraft later. In general, there are three types of transplantation namely autologous, allogeniec and cord haematopoietic blood stem cell transplantation (Hoffbrand *et al.*, 2010).

2.2.3 Autologous Haematopoietic Stem Cell Transplantation

The meaning of the term auto is self. It is a type of transplant that utilizes stem cells that is collected earlier from the patient himself or herself before they are given high dose chemotherapy or radiotherapy treatment. The stem cells are stored in a nitrogen liquid container and this is known as cryopreservation. After completing the myoablative regime, the stem cells collected earlier is transfused back into the patient's blood. Homing of stem cells will take place in the bone marrow to generate new population of haematopoietic cells. Figure 2.2 shows the overview process of autologous HSCT. Diseases for which autologous HSCT may be indicated are listed in Table 2.3.

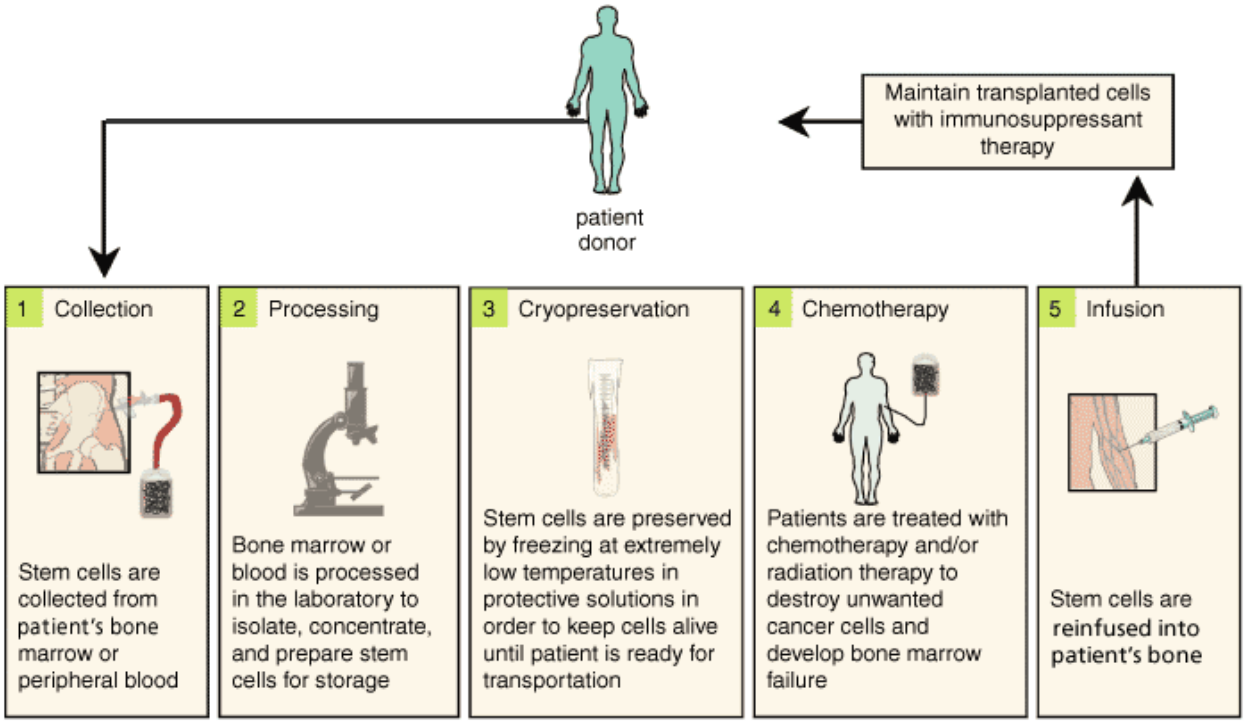

Figure 2.2: Autologous HSCT process (Source from Georgia Regents University Transplant Centre 2014).

Table 2.3: Diseases for which autologous HSCT may be indicated (Source: Approach to Haematopoietic Stem Cell Transplantation, Akiyoshi Takami, Sysmex Corporation 2010).

Malignant diseases	Nonmalignant diseases
Multiple myeloma	Autoimmune disease (Collagen disease)
Non-Hodgkin lymphoma	Amyloidosis
Hodgkin lymphoma	
Acute myeloid leukemia	
Neuroblastoma	
Rhabdomyosarcoma	
Brain tumors (medulloblastoma, ependymoma, peripheral neuroectodermal tumor, etc)	
Hepatoblastoma	
Wilms tumor	
Ewing sarcoma	
Osteosarcoma	
Germ cell tumor	
Breast cancer	
Ovarian cancer	
Lung cancer	

2.2.4 Allogeneic Haematopoietic Stem Cell Transplantation

The meaning of the term allo is other. Here, the stem cells are harvested from another person called a donor. To achieve a successful allogeneic HSCT, it is very important to use blood from a donor who has the best matching of HLA antigen and if preferable best match of HLA genotype with the recipient. This can be attained by screening close family members. The most likely good match can be found from patient's brother or sister. In some circumstances, a good match also can be found in parents, children and other relatives. However, sometimes it is difficult to get it from the family members but there is another alternative option where transplantation can be done from a donor who is not related with the patient. Such transplant is known as match unrelated donor (MUD) where there is some mismatch in HLA between the donor and the recipient. The search of MUD can be applied from national bone marrow registries. But in this case, certain risk or complication is higher such as patient is prone to get severe GVHD, graft failure and others. Diseases for which allogeneic HSCT may be indicated are listed in Table 2.4.

Table 2.4: Diseases for which allogeneic HSCT may be indicated (Source: Approach to Haematopoietic Stem Cell Transplantation, Akiyoshi Takami, Sysmex Corporation 2010).

Malignant diseases	Nonmalignant diseases
Acute myeloid leukaemia	Aplastic anaemia
Acute lymphocytic leukaemia	Paroxysmal nocturnal haemoglobinuria
non-Hodgkin lymphoma	Severe combined immunodeficiency
Myelodysplastic syndrome	Fanconi anaemia
Multiple myeloma	Diamond-Blackfan anaemia
Chronic myeloid leukaemia	Sickle cell disease
Chronic lymphocytic leukaemia	Wiskott-Aldrich syndrome
Renal cell carcinoma	Osteopetrosis
Pancreatic carcinoma	Congenital metabolic disease
Large intestine carcinoma	Autoimmune disease
Breast cancer	

2.2.5 Umbilical Cord Blood Transplantation

This is also one of allogeneic type of transplant. Cord blood is a type of blood taken from the umbilical cord and placenta which connect the mother and the baby. Haematopoietic stem cells is harvested from it and stored or cryopreserved until they are needed for transplant. There are many advantages of cord blood transplantation. First, stem cells in the cord blood are very immature, therefore there is a less need for perfect matching or in other words HLA mismatched to some degree can be done for transplantation. Secondly, since the stem cells are taken from baby's cord blood, there is no stress and no risk to the donors.

Thirdly, as the cord blood is readily available, the transplantation can be performed relatively fast and finally, cases of GVHD are rare compared with other stem cell sources. However, it also comes with drawback such as it has slow engraftment rate for WBC, RBC and platelet after transplantation and making patients at disadvantage where infection can occur easily and cause prolonged hospitalization. Besides, since the stem cells are in small quantities, cases of graft failure are frequent and also additional cord blood from the same donor cannot be obtained (Schoemans *et al.*, 2006).

2.3 AUTOLOGOUS PERIPHERAL HAEMATOPOIETIC STEM CELL TRANSPLANTATION

As we already know, the sources of stem cells can be harvested from bone marrow, peripheral blood and cord blood. Among the three, stem cells collected from the peripheral blood is the most commonly used in stem cell transplant procedure for a number of diseases including non malignant and malignant (Passweg *et al.*, 2013). In Malaysia, the scenario follows the same trend as in the European country practices and this is reflected in the data from National Transplant Registry 2011.

There are many benefits of autologous PSCT, one of them is it takes fewer days for the engraftment of hematopoietic cells to occur. For example, in PBSCT for allogeneic transplantation the median day for engraftment is 12 days compared to bone marrow which took about 17 days (Bensinger *et al.*, 2001).

Besides, unlike in allogeneic HSCT, patients undergone autologous PBST are not prone to get GVHD. The treatment related mortality rate is also low compared to allogeneic HSCT. For example in lymphoma particularly Hodgkin lymphoma who undergone allogeneic HSCT, the treatment related mortality is 51.7% at 4 years (Peniket *et al.*, 2002).

However, there are still back draws or disadvantages of autologous PBST such as it is associated with high relapse rate (Peniket *et al.*, 2002). The postulation cause of this can be related to the procedure itself. Since in autologous transplantation, patient's own blood is used, immunosuppressive therapy after the transplant is not required. Therefore, there is a chance or probability that some malignant cells may intermixed with the transplant material during the process of stem cell harvest.

2.4 AUTOLOGOUS HAEMATOPOIETIC STEM CELL TRANSPLANTATION IN MULTIPLE MYELOMA

2.4.1 An Introduction to Multiple Myeloma

Multiple myeloma is a cancer of plasma cells. Majority of cases are more than 40 years old and peak incidence is in the seventh decade of life. The aetiology of multiple myeloma remains unknown. But what have been known so far, it is more common in black individual (Hoffbrand and Moss, 2011). Besides total accumulation of complex genetic changes,

dysregulation of cyclin D is said to be the early unifying event towards its pathogenesis (Chesi and Bergsagel, 2013).

Patients can present with symptoms of bone marrow failure that will result in anaemia (lethargy, pallor, weakness and tachycardia), leucopenia (frequent infections) and thrombocytopenia (easy bleeding). Besides symptoms of kidney failures (swollen legs, poor urine output) and bone pain as a result of increased osteoclast activity can be seen.

According to the WHO 2008 criteria, diagnosis of symptomatic multiple myeloma can be made when it fulfill three criteria namely the demonstration of monoclonal protein in serum or urine, increased clonal plasma cells in the bone marrow and presence of related organ or tissue impairment (hypercalcemia, renal insufficiency, anaemia and bone lesions). The International Staging System (Table 2.4) is the current standard used in staging multiple myeloma (Greipp *et al.*, 2005). Stage III is considered and advanced stage with high tumor burden.

Table 2.4: International Staging System (Source: Oxford Handbook of Clinical Haematology, 3rd edition, 2009)

Stage of multiple myeloma	Level of biochemical markers
Stage I	Serum ß$_2$-Microglobulin <3.5mg/L and serum albumin ≥35g/L
Stage II	Serum ß$_2$-Microglobulin <3.5mg/L and serum albumin <35g/L ***Or*** Serum ß$_2$-Microglobulin 3.5 - <5.5mg/L
Stage III	Serum ß$_2$-Microglobulin ≥5.5mg/L

For assessment of response treatment in multiple myeloma, there are 2 systems can be used namely the European Group for Blood and Marrow Transplant (BladÉ *et al.*, 1998) or the International Myeloma Working Group (Durie *et al.*, 2006).

According to the International Myeloma Working Group, there are overall eight response subcategory namely stringent complete response (sCR), complete response (CR), very good partial response (VGPR), partial response (PR), stable disease (SD), progressive disease, clinical relapse and relapse from CR. In HUSM, generally the aim of treatment is at least patients achieve VGPR before proceed with transplantation. Criteria for VGPR is when patients have serum and urine M-protein detectable by immunofixation but not on electrophoresis or ≥90% reduction in serum M-protein/ urine M-protein level <200mg/24 hour.

2.4.2 Treatment and Role of Autologous HSCT in Multiple Myeloma

The latest data from National Registry Transplantation showed that from 1987 until 2011, multiple myeloma sit at the second place among malignant haematological disorders for indication of autologous stem cell transplantation with a total of 6% cases. Multiple myeloma is considered incurable disease because most of the patients had disease progression and relapse after treatment. In addition, even patients who are considered minimal residual disease (MRD) negative, evidence of residual disease can still be detected. Currently, younger patients who are newly diagnosed with multiple myeloma, autologous stem cell transplantation is considered the gold standard as part of the initial treatment (Hübel *et al.*, 2014).

The long term and overall survival of multiple myeloma has dramatically improved nowadays with the invention of novel drugs such as thalidomide, bortezomib and lenalidomide together with autologous stem cell transplantation (Palumbo *et al.*, 2011). Once patient is diagnosed to have multiple myeloma, the suitability of treatment plan is dependent on the age of the patient, co morbidity (including renal, cardiac, hepatic, neurologic and pulmonary function), performance status and disease progression status at diagnosis. This then can be divided into transplant eligible and non-transplant eligible.

In Malaysia, generally patient who age below 60 years old with a reasonable health status are selected for autologous stem cell transplantation. However, some European countries had experienced of successful treatment in the older patients. In their study, they have

concluded that selected patient who is age above 70 years old should not be excluded from autologous stem cell transplantation as the toxicity and the outcome seems to be comparable with the younger age that is below 65 years old (Gertz and Dingli, 2014). There was another study done that reported the same findings (Kumar *et al.*, 2008). The option of choosing patient age less 60 years old in Malaysia is perhaps that Asian people are more fragile compared to the European where biological and genetic factors may play a role.

In the past, the selected eligible patients are initially given induction chemotherapy which could be VAD (which consist of Vincristine, Adriamycin and dexamethasone) or ABCM (which consist of Adriamycin, BCNU, Cyclophosphemide, Melphalan). However with the availability of new novel agents (Bortezomib, Thalidomide or Lenalinomide), VAD regime is no longer being used as these new drugs has shown to be superior. In HUSM, VTD regime (which consists of bortezomib, thalidomide and dexamethasone) is selected. The aim is to clear the marrow from malignant myeloma cells. After four to six cycles of chemotherapy, stem cells collection from the peripheral blood is then collected. There is a convincing study which showed that patients who undergone autologous transplantation had longer survival time or survival advantages. Their five year survival rate improves to 52% compared to only 12% for those who treated with just conventional chemotherapy (Attal *et al.*, 1996).

Achievement of a very good partial response (VGPR) and complete remission (CR) post autologous stem cell transplantation can be used as one of the predictors for long term

clinical outcome (Harousseau *et al.*, 2009; Chanan-Khan and Giralt, 2010). Usually after autologous stem cell transplantation procedure completed, patients are given treatment with a drug called interferon as a maintenance therapy in the past. However latest data in a recent study in 2012 suggest that the combination used of new drugs such as bortezomib , dexamethasone and thalidomide (VDT) has shown encouraging result where it can improved remission status of the patient from a very good partial response (VGPR) to complete remission (CR) (Cavo *et al.*, 2012).

For the role of autologous stem cell transplantation in relapse multiple myeloma, some studies shows that it is an effective treatment that is well tolerated by patients. Besides it also demonstrated an overall good response rate. For an instance in one study done in Italy, they have shown that 69% of their patients who undergone a second autologous stem cell transplantation after relapse or disease progression following first-line autologous stem cell transplantation showed major response ($\geq$ partial response) (Elice *et al.*, 2006). Besides other factor that influenced overall survival and progression-free survival includes patients who received fewer regimes of chemotherapy before transplantation and also patients who had late disease relapse (Cook *et al.*, 2011; Jimenez-Zepeda *et al.*, 2012).

2.5 AUTOLOGOUS HAEMATOPOIETIC STEM CELL TRANSPLANTATION IN LYMPHOMA

2.5.1 An Introduction to Lymphoma

Lymphoma is a group of lymphoid cancers that start in the lymphatic system where malignant lymphocytes accumulate. It is the most treatable form of malignancy. It can be divided into two major divisions namely Hodgkin lymphoma (HL) and non-Hodgkin lymphoma (NHL) and this is based on the presence or absence of Reed-Sternberg (RS) cells.

The causes are unknown exactly. However in HL, Epstein-Barr virus (EBV) has been postulated to be linked with it as the EBV genome is detected in Hodgkin tissue biopsy (Hoffbrand and Moss, 2011). Besides EBV is also linked to a rare type of NHL that is the Burkitt lymphoma. There are several known risk factors for NHL which includes microbiological infection, previous exposure to radiation, immunodeficiency state due to infection (such as Human Immunodeficiency Virus) or due to prolonged drug treatment.

Patients may present with enlarged lymph nodes at the neck, armpits or groin which are painless. The nodes can grow in size and not responsive to antibiotic. Some patients have constitutional symptoms such as weight loss and fever.