ROLE OF TC99m-BESILESOMAB SCAN WITH THE ADDED BENEFIT OF SINGLE PHOTON EMISSION COMPUTED TOMOGRAPHY/ COMPUTED TOMOGRAPHY (SPECT/CT) IN LOCALISING INFECTION

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ABBREVIATIONS

SPECT- SINGLE PHOTON EMISSION COMPUTED TOMOGRAPHY

SPECT/CT -SINGLE PHOTON EMISSION COMPUTED TOMOGRAPHY/

COMPUTED TOMOGRAPHY

CT- COMPUTED-TOMOGRAPHY

MRI- MAGNETIC RESONANCE IMAGING

PUO – PYREXIA OF UNKNOWN ORIGIN

OM – OSTEOMYELITIS

Tc99m – TECHNETIUM 99 METASTABLE

WBC- WHITE BLOOD CELLS

ESR- ERYTHROCYTE SEDIMENTATION RATE

CRP- C-REACTIVE PROTEIN

Tc99m-HMPAO – (Tc99m-HEXAMETHYLPROPYLENEAMINE OXIME)

Tc99m-MDP/HDP- (Tc99m-METHYLENE DIPHOSPHONATE/ HYDROXY

METHELENE DIPHOSPHONATE

CEA- CARCINOEMBRYOGENIC ANTIGEN

ABSTRAK

Imbasan Tc99m-besilesomab dengan manfaat tambahan *Single Photon Emission Computed Tomography/ Computed Tomography* (SPECT/CT) untuk mengenal pasti fokus lokasi infeksi dalam tubuh pesakit.

Tujuan: Kajian ini adalah untuk mengenalpasti fokus lokasi infeksi menggunakan Tc99m-besilesomab serta menilai sumbangan tambahan SPECT/CT berbanding imej planar.

Metodologi: Imbasan prospektif planar dan SPECT/CT Tc99m-besilesomab telah dilakukan ke atas 23 pesakit (8 lelaki dan 15 perempuan) yang disyaki mempunyai infeksi dan analisa data dilakukan. Keputusan imejan telah dikaji dan dibandingkan dengan keputusan ujian darah, kultur tisu dan juga diagnosa akhir sebagai piawai (*standard*) rujukan. Dua puluh tiga pesakit menjalani pengimejan planar dan pengimejan SPECT/CT dimana 13 pesakit dengan diagnosa demam tanpa mengetahui lokasinya dan 10 pesakit dengan diagnosa infeksi kuman pada tulang. Sumbangan yang diberikan oleh imejan SPECT/CT dalam kajian ini telah direkodkan dan faktor yang mempengaruhi imbasan dianalisis. Kelebihan pengunaan pengimejan SPECT/CT berbanding planar pada pengurusan pesakit juga dinilai.

Keputusan: Data menunjukkan Tc99m-besilesomab dapat mengesan infeksi dengan sensitiviti yang tinggi iaitu 88% dan spesifisiti sebanyak 83%. Melalui kajian ini, perjanjian yang diperolehi diantara kehadiran infeksi dan keupayaan pengimejan Tc99m-

besilesomab untuk mengesan infeksi adalah agak tinggi dengan perjanjian kappa sebanyak 0.7 ($p < 0.05$). SPECT/CT memberikan maklumat tambahan untuk mengesan infeksi berbanding pengimejan planar dengan peningkatan sensitiviti sebanyak 6% dan spesifisiti sebanyak 17%. Pengimejan SPECT/CT menunjukkan pengimejan benar *"True positive*" terhadap 16 daripada 23 pesakit, "*true negative"* kepada 6 daripada 23 pesakit dan "*false negative"* kepada 1 daripada 23 pesakit. Penambahan SPECT/CT telah memberi perubahan diagnosa akhir terhadap seorang pesakit di mana lokasi infeksinya tidak dapat dikesan oleh pengimejan planar. SPECT/CT juga memberikan lokasi anatomi yang tepat dalam pengimejan yang positif.

Kesimpulan: Skan Tc99m-besilesomab mempunyai sensitiviti dan spesifisiti yang tinggi untuk mengesan infeksi di dalam tubuh. Penambahan SPECT/CT telah meningkatkan sensitiviti mengenalpasti fokus infeksi dari 88% kepada 94% dan spesifisiti dari 83% kepada 100%. SPECT/CT juga dapat memberi maklumat tambahan berkenaan lokasi anatomi yang tepat dan juga sejauh mana infeksi telah merebak di dalam tubuh.

Kata Kunci: SPECT/CT; infeksi tulang; PUO; pengimejan besilesomab; pengimejan scintimun; infeksi

ABSTRACT

Role of Tc99m-besilesomab scan with the added benefit of Single Photon Emission Computed Tomography/ Computed Tomography (SPECT/CT) in localising infection.

Aim: The aim of the study is to localise site of infection using Tc99m-besilesomab scan and to assess the additional contributions of SPECT/CT over planar scan in patients.

Material and Methods: Planar and SPECT/CT Tc99m-besilesomab scan were prospectively performed in 23 patients (8 males and 15 females) with suspected infection and the data were analysed. In all patients Tc99m-besilesomab imaging findings were correlated with the results of blood cultures, tissue cultures and final diagnosis as the reference standards. Twenty three patients underwent planar and SPECT/CT imaging (13 with unknown site of infection and 10 with suspected osteomyelitis). Both cold and hot spots were considered diagnostic. SPECT/CT contribution was recorded and the factors affecting the scan were analysed. The impact of SPECT/CT on patient management was assessed.

Results: Tc99m-besilesomab scan managed to detect infection with high sensitivity of 88% and specificity of 83%. The agreement that was obtained between the presence of infection and the ability of Tc99m-besilesomab imaging to detect infection was significant with a kappa agreement of 0.7 ($p<0.05$). The addition of SPECT/CT has increased the sensitivity by 6% and specificity by 17%. Sixteen out of 23 patients had true positive

Tc99m-besilesomab SPECT/CT study. Six patients had true negative and only one patient had false negative study. SPECT/CT changed the management in one patient which was missed by planar imaging and also provided additional information for detection of infection as compared to planar imaging by providing accurate anatomical localization in all true positive foci.

Conclusion: Tc99m-besilesomab scan is useful in cases of unknown infection and has a high sensitivity and specificity. The addition of SPECT/CT increased the sensitivity of detecting focus of infection from 88% to 94% and specificity from 83% to 100%. SPECT/CT also provided extra information to the exact anatomical location and the extent of involvement of the disease as compared to planar imaging alone.

Key Words: SPECT/CT; Osteomyelitis; besilesomab scan; Scintimun scan; PUO

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CHAPTER 1: INTRODUCTION

Diagnostic imaging with modern molecular nuclear medicine is based on the availability of sensitive and relatively specific radiopharmaceuticals tailored for the different targets that can be expressed in this complex scenario. The radiopharmaceuticals that was used and currently being used for imaging infections are gallium-67 citrate (Ga67-citrate), labeled leukocytes (WBC) (indium-111 oxine and Tc99m-HMPAO), antigranulocyte antibody (Tc99m-besilesomab), bone scan (Tc99m-MDP/HDP), fluorine-18 fluorodeoxyglucose (18F-FDG), and gallium-68 citrate (Ga68-citrate). Many other newer agents are being investigated for infection and inflammation studies.

Previously gallium-67 citrate was used for infection imaging. Gallium imaging shows acute and chronic infection as well as non-infectious inflammation foci and malignancies. About 90% of circulating Gallium ions is bound to transferrin with some bound to lactoferrin in the plasma. Direct bacterial uptake also contributes to gallium-67 uptake. The normal distribution of gallium-67 is seen in bone, marrow, liver, gastrointestinal, urinary tracts and soft tissues. Due to the high radiation dose, long image acquisition time (18-72 hours), poor image quality and its low specificity for infection it is not commonly used nowadays (Palestro et al 2013).

In vitro leucocyte (WBC) labeling with indium-111 oxine or 99mTc-HMPAO is sensitive for identifying neutrophil mediated infectious process. Circulating WBC of at least 2000/1L is needed for satisfactory image. Normal distribution of indium-111 is seen in the liver, spleen, and bone marrow. Tc99m-HMPAO show uptake in reticuloendothelial

system, urinary tract, large bowel, and occasionally gall bladder (Palestro et al 2013). This leucocyte labeling is limited by low counts, low resolution images and also high radiation risk if indium-111 is used (Ziessman, 2014). Both this radiopharmaceuticals also require in vitro labeling which resulted in a long preparation time, risk of contamination and also handling of potentially infected blood products.

Mourad et al (2003) found that from ten studies of fair methodological quality using various radionuclide to test characteristics of nuclear imaging studies in PUO, antigranulocyte scan (Tc99m-besilesomab) based studies report the highest specificity ranging from 93%-94% but are insensitive (40%-75%) as compared to indium 111–oxine labeled WBC scan with a sensitivity of 45%-82% and a specificity that ranges from 69% to 86%. From their study it was noted that the best quality study of gallium-67 citrate scan reported a sensitivity of 67% and a specificity of 78% but it only included 20 patients.

Richter et al (2011) found that Tc99m besilesomab imaging was comparable with Tc99m-WBC imaging with slightly higher sensitivity at 74.8% in Tc99m-besilesomab and 59.0% in Tc99m-WBC; however besilesomab has slightly lower specificity at 71.8% as compared to 79.5% in Tc99m-WBC to detect infection. In their analysis also they found that besilesomab had higher sensitivity than WBCs in patients with chronic osteomyelitis with an agreement of 0.79 in the chronic group and 0.80 in the acute infection group (Richter et al 2011). The use of Tc99m-besilesomab is also easier compared to the use of radiolabelled WBC as it does not need the handling of blood.

Imaging with F-18 FDG is related to its nonspecific accumulation in inflammation as well as malignancies. Its uptake in inflammatory and malignancy is related to its increased GLUT expression and upregulation of hexokinase enzymes (Lazzeri et al 2013).

F-18 FDG imaging show a high negative predictive value, therefore it is the imaging of choice in patients with low to intermediate probability of infection. In cases with high probability of infection then the suggested infection imaging is with labeled leucocyte or antigranulocyte antibody (Lazzeri et al 2013). F-18 FDG scans hold promise, but more studies are needed to fully understand its usefullness (Mourad et al 2003). Another limitation of this scan is the cost involved as it requires a PET or PET/CT machine and a cyclotron to produce the tracer F-18 FDG.

In localizing infection in bone/ joints and prosthesis, bone scan is also commonly used. Its does not have much role in PUO imaging as PUO is caused by many other factors and not related to bone all the time. Bone scan is generally cheaper, widely available, and not complex to be performed as compared to other radionuclide imaging. Bone scan is sensitive but has poor specificity. It depends on the osteoblastic activity (new bone formation) and also on the blood flow. Due to the non specific nature of this scan it will show hot spots / uptake in bone remodeling, infection, heterotropic ossification, fracture and aseptic loosening. On performing triple phase bone scan, a negative scan can exclude infection confidently, however in cases with positive triple phase bone scan it is difficult to conclude as infection. In these cases, confirmation with leucocyte or antigranulocyte antibody imaging is necessary (Gemmel et al, 2012). Table 1 summarizes the different radionuclide imaging techniques available for prosthesis infection.

Table 1: Interpretation criteria, advantages and disadvantages of radionuclide imaging technique for prosthesis joint infection.

Imaging technique	Interpretation criteria	Advantages	Disadvantages
(Three-phase) bone scintigraphy	1. Quantitative versus semi-qualitative analysis 2. Technique used (three-phase scan or only delayed bone scan)	Sensitive, high NPV, screening tool, easily performed, widely available, cheap	Low specificity, low accuracy, not useful within 1 year postoperatively
Sequential bone/gallium scintigraphy	Uptake congruency of the spatial distribution and intensity	Improved specificity versus bone scan alone, use in neutropenic patients	High additional radiation dose of 18 mSv, many equivocal cases, not widely performed
In vitro leucocyte and/or bone/marrow scintigraphy	1. Quantitative versus semi-qualitative analysis 2. Uptake congruency of the spatial distribution and intensity 3. Serial or delayed imaging	Highly sensitive and specific when combined with sulphur colloid imaging	Additional radiation dose, discomfort and inconvenience for the patient, laborious—risk of blood handling— not useful in neutropenic patients, expensive especially when combined
Antigranulocyte scintigraphy	1. Quantitative versus semi-qualitative analysis 2. Serial or delayed imaging	In vivo imaging method (use of kits)	AGS overall lower accuracy compared to in vitro LS, expensive, possibility for allergic reaction, not widely practised, role in prosthetic joint imaging not yet fully established
[^{18}F]FDG PET and/or PET/CT	1. Quantitative versus semi-qualitative analysis 2. Several FDG uptake patterns available in the literature	Highly sensitive for the inflamed prosthetic joint, improved spatial resolution especially when combined with CT, fast imaging technique	Limited specificity for infection, not useful within 1 year postoperatively, attenuation artefacts (metallic implants), lower accuracy for knee prostheses, expensive technique, not widely available, role in prosthetic joint imaging not yet fully established
SPECT/CT	Coregistered analysis of the images	Improved specificity versus SPECT alone, can differentiate between bone versus soft tissue infection	Attenuation artefacts (metallic implants), often low counts of in vitro leucocyte/gallium studies, role in prosthetic joint imaging not yet fully established

NPV negative predictive value

Table taken from European journal of nuclear medicine and molecular imaging (2012) 39:892–909, Prosthetic joint infections: radionuclide state-of-the-art imaging. (Gemmel et al, 2012)

1.1 Malaysian Scenario

In clinical practice, the diagnosis of infection is based on history, clinical findings, imaging and other investigations depending on the probable site of infection. Blood investigations such as WBC and ESR are commonly performed to look for sign of infection. Anatomical imagings are then performed to look for changes that occur to support the diagnosis of infection. However, anatomical imaging sometimes misses the site of infection as it is able to detect the changes when substantial anatomical changes have occured. Functional imaging has a role to overcome the weakness of conventional anatomical imaging (CT or MRI) due to its higher specificity and ability to detect infection before any anatomical changes has occurred (Lazzeri et al 2012).

In Malaysia, no documented incidence or prevalence data of cases of infection is available. In Hospital Kuala Lumpur, besilesomab is currently being used for diagnostic purpose with an average of 2-4 scans performed per month at Hospital Kuala Lumpur. It is used to locate areas of infection in adults. Besilesomab scan was started since 2010 to diagnose infection of bones in suspected prosthesis infection, infection of long bones and spine OM. It was also used previously to diagnose PUO but due to non-availability of sources, information and knowledge, its use was not fully utilised. This lack of information makes primary clinician more dependent on anatomical imaging such as CT for detecting site of infection.

The recently acquired Besilesomab kit in Hospital Kuala Lumpur has made this scan available again. Since then, there were few cases to rule out infection in patient

suspected of OM and PUO. This is the first infection study using nuclear medicine imaging and SPECT/CT in Malaysia for localising infection.

1.2 Besilesomab

Besilesomab is an antigranulocyte antibody kit that utilises besilesomab (murine immunoglobulin (MoAbs) of IgG1) to perform the scan. It is a functional imaging studies (radionuclide imaging), where small quantities of radioactive material is used. This immunoglobulin specifically binds to around 99.6% of mature human granulocytes in whole blood sample. The use of besilesomab scan is to identify infection site early so that treatment can be started with the appropriate antibiotics and reduce hospital stay. The correct use of antibiotics will prevent from the emerging problem of antibiotics resistance. The most common indication for this scan is to detect infection in cases of suspected osteomyelitis, prosthesis infection and PUO.

Besilesomab has an affinity for NCA-95 antigen (also referred to as CD66b and CEACAM8). These non specific antigens are present in the cytoplasm and on the cell membranes of granulocytes, mature bone marrow cells of the granulocytic lineage and are also expressed on the macrophages, epithelium of the lung, colon and hematopoietic bone marrow. Besilesomab binds to antigenic structure shared by NCA-95 glycoprotein of granulocytes and tumour marker CEA and cross reacts with tumours expressing CEA. All this can lead to significant binding of besilesomab to colon carcinomas (Bosslet et al, 1988). This binding can produce false positive images in CEA expressing tumors. False positive image can also be seen in haematological malignancies such as myeloma (Satoh, 2002). This is due to expression of CD66 on bone marrow cells of patients with multiple myeloma. Besilesomab was also found bounded to pancreatic, some lung and some breast

carcinomas but does not bind to blood vessels and connective tissue (Scintimun Monograph, 2013).

The typical uptake patterns of the radiopharmaceutical include focal uptake at sites of infection as well as staining of the hematopoietic bone marrow (Richter et al 2011). Tracer accumulation is also seen in the liver, thyroid and kidney (up to 2%), spleen (6%) and in the bowel (4%). The exact mechanism of besilesomab accumulation at the site of infection is not fully understood. It is suggested that the accumulation occurs passively which is due to the increased vascular permeability and partly through active migration of besilesomab attached human granulocytes to the infection/ inflammation site because only about 10% to 20% of the injected radiolabelled antibody bind in vivo to human circulating granulocytes (Scintimun Monograph, 2013). Specific binding of besilesomab to already migrated and activated granulocytes may also be the major part of the detection signal (Scintimun Monograph, 2013).

Besilesomab has a two phase whole blood concentration and the early phase has a half life of 0.5h and delayed phase with a half life of 16h. Besilesomab major route of elimination is via reticuloendothelial system with the most occurring in the spleen. This was shown in the clinical trials whereby within 6 hours of administration, 3.0% of the whole body radioactivity is found in the spleen and 1.5% is found in the liver. In the clinical trials the kidneys also showed low renal activity clearance of 0.2 l/h for a glomerular filtration rate around 7 l/h. This was showed by measurement of 24 hours urine radioactivity in which up to 14% of the activities administered are excreted to urine (Scintimun Monograph, 2013).

Besilesomab has few disadvantages such as having high molecular weight which leads to slow diffusion of besilesomab to target site, low target concentration in the involved site, long plasma half-life and lastly high physiological uptake in the liver and bone marrow. In order to get good target to background ratio, longer interval duration is needed from administration of Tc99m-besilesoamb to imaging. Planar images at 1h, 3h, and 5h and if necessary at 20-24 hours should be acquired. To increase the sensitivity, delayed images at 20-24h have been recommended. However due to time consumption factor and logistics reasons it is not routinely performed (Becker et al, 1994). SPECT and SPECT/CT is also suggested for better localisation of infections.

The purpose of conducting this study is to assess the usefulness of besilesomab scan and the contribution of SPECT/CT as compared to planar in specific group of patients (suspected infection) and how it affects the overall patient management. This will serve as a guide in establishing the appropriate clinical indications for besilesomab SPECT/CT imaging in this centre.

CHAPTER 2: LITERATURE REVIEW

Nuclear imaging is very useful in localising site of infection and inflammation due to its overall good test characteristics, low toxicity and its ability to image the whole body as compared to other current imaging procedures such as CT and MRI (Becker & Meller, 2001). The common indication for infection imaging in nuclear medicine is when the physician is unable to detect or localise site of unknown inflammatory focus (Das et al., 2002). In developing countries, bacterial infection is high on the list of commonly encountered disease and is a major cause of morbidity and mortality (El-Ghany et al, 2005). The diagnosis of infections such as endocarditis, OM, intra-abdominal abscesses and in patients with PUO is still a challenging problem in many hospitals and countries.

2.1 Infection

Infection is divided into acute and chronic based on its duration of illnesses. In acute infection the duration of illness typically last less than 6 months and are mediated by immune cells and cytokines. In the initial period, mast cells are stimulated to produce histamine, prostaglandin and serotonin, which cause an increase in vessels permeability. This in turn enables the neutrophils which are the main cells in acute infection to move through the capillary walls into the affected tissue. Chronic infection is when the illness last more than 6 months. The primary immune cells involved in chronic infection/ inflammation are the macrophages and T lymphocytes, which produce cytokines and enzymes that cause more damage to cells (Kumar and Robbins, 2013).

Infection can also be classified based on the type of organisms into A) opportunistic infections which is caused by organism which is a normal host, and is due to a change in environment, trauma, injuries, anti-microbial treatment, weakened immunity and also when the normal flora migrate to a new compartment in the body (Baldoni, 2009). B) Exogenous infections is caused by organism that are transmitted to healthy hosts from an infected agent such as animals/ humans or from a contaminated environment through the air, aerosol, sexual intercourse, blood transfusions or animal bites. Exogenous infections can be acquired from the community or in healthcare settings (Baldoni, 2009). When the transmission of infection occurs within the community it's called as community acquired infections whereas nosocomial infections are infections that have been contracted in a healthcare setting and are potentially caused by organisms that are resistant to antibiotics. This is commonly related to hospital associated factors such as hospital procedures/ insertion of intravascular devices like drip, recent surgery/ intervention, drug fever, clostridium difficile colitis and bed sores (Baldoni, 2009).

2.2 Pyrexia of unknown origin (PUO)

Pyrexia of unknown origin (PUO) was first defined in 1961 by Petersdorf and Beeson as the following: (1) a temperature more than 38.3°C (101°F) on several occasions, (2) more than 3 weeks' duration of illness, and (3) failure to reach a diagnosis despite 1 week of inpatient investigation.

PUO may be a cause of more than hundreds of diseases and is one of the major diagnostic challenges in medicine (Knockaert et al 1992). Over the time, new diseases are added to the causative list. Durack and Street (1991) suggested making changes to the definition proposed by Petersdorf R.G earlier by categorizing PUO into 4 subtypes which are classic, nosocomial, immunodeficient and HIV associated PUO. The second change suggested by Durack and Street, (1991) is the investigation duration before it can be called PUO which is three outpatient visits or 3 days of inpatient investigation. However, De Kleijn et al (2000) proposed to change these criteria to qualitative criteria which include a period after which no diagnosis or reasonable diagnostic hypothesis has been established following an appropriate intelligent standard inpatient or outpatient investigation.

Coming to the 4 subtypes mentioned earlier, classic PUO is caused mainly by infections, malignancy and collagen vascular disease. Nosocomial infection refers to fever in patients that have been admitted to hospital for at least 24 hours. Immunodeficiency can be seen in patients receiving chemotherapy or in hematologic malignancies. In this condition the fever is concomitant with neutropenia (neutrophil <500/uL) or impaired cell-mediated immunity. This is dangerous as it can masks a dangerous on going infection. In

this group of patient infection is the most likely cause. The last but quite important is human immunodeficiency virus (HIV)-associated which is also a subgroup of immunodeficient PUO (Durack and Street, 1991). These patients are HIV-positive and present with fever. The primary phase shows fever since it has a mononucleosis-like illness. In later stages, occurance of fever mostly is due to superimposed infections (Durack and Street, 1991). This definition is important as the treatment suggested is different whereby in classic PUO, it is not suggested to start antibiotics early until the cause has been found, however in nosocomial, HIV and immunedeficient/neutropenic patients, it is important to start treatment early to prevent morbidity and mortality (De Kleijn et al, 2000).

Lazzeri et al, (2012) mentioned that of all the causes of PUO, infection accounts for about 21-54%, followed second by neoplasm 6-31% and non-infectious inflammatory diseases (NIIDs) 13-24%. Rheumatic diseases, collagen diseases, vasculitis syndromes and granulomatous disorders are just some of the examples of NIID.

In any cases of suspected PUO, before commencing treatment the treating physician should first obtain a comprehensive history; perform physical examination, and investigations in the search for the possible cause. History of occupation, environment, contact with person with similar symptoms, recent travelling history, contact with pets and wild animals should be obtained to narrow down the cause of PUO. Blood investigations such as complete blood count, CRP, ESR, liver and renal function test and rheumatoid factor should also be performed. There is no diagnostic gold standard in PUO where the other diagnostic test can be measured (Mourad et al 2003). To obtain final diagnosis, multiple methods are used such as clinical history, biopsy, surgery, other imaging and

sometimes post mortem examination (Mourad et al, 2003). Anatomic imaging modalities (including CT, MRI, and USG) are often the first-line investigations performed to look for infection.

2.3 Osteomyelitis (OM)

OM is defined as infection in bone caused by infective microorganism. OM can be localized to a part of bone or it can involve multiple parts of the bone such as the cortex, periosteum, marrow and also soft tissue. Many classification systems has been described previously, however the Waldvogel classification that was described in 1970 is commonly used in clinical studies for its greater clinical applicability, whereas the Cierny and Mader classification is used for its clearly defined treatment strategies (Lima et al 2014).

The Waldvogel system divides OM based on its pathophysiology and duration of infection (Lima et al 2014). Pathophysiologically, infections are classified into; hematogenous OM which is usually caused by single pathogen; OM secondary to a contiguous focus of infection; and OM associated with peripheral vascular insufficiency in which soft tissue infection in the foot spreads to bone. This is described in diabetic patients and has multiple pathogens involved. Hematogenous OM is seen mostly in children, with 85% of patients aged below 17 years. In adult, around 47–50% of all OM occur after trauma. Only 2-7% of patients end up with OM of the vertebra (Trueta J 1959).

OM is also classified as acute and chronic based on the duration of the infection. Acute OM is able to be detected by histological methods 2 weeks after the onset of the disease. It presents as a suppurative infection accompanied by edema, vascular congestion and small vessel thrombosis. In early acute phase, there is reduced vascular supply to the bone and the surrounding soft tissue which leads to compromised medullary and periosteal

blood supplies. This in turn leads to large areas of sequestra (dead bone) (Emslie K R, 1983). The production of dead bone can be stopped if the infection is treated aggressively with antibiotics and surgery. In an established infection, there is formation of fibrous tissue and chronic inflammatory cells on the granulation tissue and dead bone. This will lead to a compromised vascular supply which will prevent an effective inflammatory response (Ciampolini and Harding, 2000). In acute OM the patient clinically present with systemic symptoms such as fever, bony pain and lethargy. On physical examination erythema, soft tissue swelling or joint effusion, decreased joint range of motion, and bony tenderness are commonly seen.

Chronic OM is a severe, persistent, and sometimes incapacitating infection of bone and bone marrow which can develop over months and years. Due to difficulty in treating the infection it often presents as recurrence (http://emedicine.medscape.com/article/393345-overview,2017). Local factors such as foreign body, dead bone fragments and necrosis; and general factors such as diabetes, smoking and vascular disease contribute to the development of chronicity (Ciampolini and Harding, 2000). In chronic OM there is presence of necrotic bone, formation of new bone, and the exudation of polymorphonuclear leukocytes together with lymphocytes, histiocytes, and occasionally plasma cells. There is formation of sheath of new bone (involucrum) from the surviving fragments of periosteum and endosteum in the region of the infection. This involucrum encased the dead bone under the periosteum and leads to formation of chronic sinus as the pus tracks from soft tissues through the openings and irregularity of involucrum to the skin surfaces. The involucrum will then thickens and increase in density to form a new diaphysis. After host defense response or operative

removal of the sequestrum, in children new bone formation may fill the remaining cavity, however, in adults the cavity may remain or it may be filled with fibrous tissue, and form a sinus tract (Ciampolini and Harding, 2000). In chronic OM the patient may present with clinical findings of pain, sinus tract, and raised ESR. Fever can be present or most of the time absent. WBC is mostly normal. The other clinical findings are exposed bone, necrosis of the skin and tissue and persistence of wound overlying the fracture or surgical implants (Hatzenbuehler & Pulling, 2011).

2.4 Role of imaging studies in the diagnosis of PUO and OM

Imaging also plays a role in diagnosis of OM. Plain radiography of the bone involved is one of the easier methods but it has limitation whereby bone matrix destruction of around 30-50% and extension of infection to at least 1 cm is required to produce noticeable changes in plain radiographs. In earlier phase the changes may be mild and it may not be visible up to 5 to 7 days in a child and 10 to 14 days in adults. Common early bone changes include soft tissue swelling, periosteal thickening, focal osteopenia, lytic lesions, endosteal scalloping, loss of trabecular architecture and new bone apposition (Kothari et al 2001).

CT has a role in diagnosis of OM whereby it is able to show subtle bony changes such as increase in marrow density, areas of necrotic bone and also able to show soft tissue involvement in early OM whereas in chronic OM, CT is able to show thickening and sclerosis of cortical bone, encroachment of the medullary cavity, sinus tract formation and sequestra (Pineda et al, 2009). All these changes are seen earlier on CT as compared to plain radiography. CT is superior to MRI for the detection of sequestra, cloacas, involucra,