PREPARATION AND CHARACTERISATION OF 3D PRINTED POLYAMIDE 6 COMPOSITES FOR CRANIOFACIAL RECONSTRUCTION

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by

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ABBREVIATIONS

3D	Three dimensional
ABS	Acrylonitrile butadiene styrene
Al_2O_3	Alumina
AM	Additive manufacturing
ANOVA	Analysis of variance
ASTM	American Society of the International Association for Testing Materials
ATR	Attenuated total reflectance
BAG	Bioactive glass
BCP	Biphasic calcium phosphate
BHI	Brain heart infusion
β-TCP	Beta tricalcium phosphate
CAD	Computer aided design
CAM	Computer aided manufacturing
CF	Carbon fibre
CMP	Calcium metaphosphate
CO_2	Carbon dioxide
CS	Chitosan
CT	Computed tomography
DMLS	Direct metal laser sintering
DMSO	Dimethyl sulfoxide
DSC	Differential scanning calorimetry
DTG	Derivative thermogram

FDA	Food and drug administration
FDM	Fused deposition modelling
FESEM	Field emission scanning electron microscope
FTIR	Fourier transform infrared spectroscopy
HA	Hydroxyapatite
ISO	International Standard Organization
MFR	Melt flow rate
MMA	Methyl methacrylate
MMT	Montmorillonite
MRI	Magnetic resonance imaging
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazoliumbromid)
NPs	Nanoparticles
OD	Optical density
PA	Polyamide
P. aeruginosa	*Pseudomonas aeruginosa*
PBS	Phosphate buffer saline
PCD	Programme cell death
PCL	Polycaprolactone
PE	Polyethylene
PEEK	Polyether ether ketone
PEG	Polyethylene glycol
PET	Positron emission tomography
PG	Peptidoglycan
PGA	Polyglycolide acid

PLA	Polylactic acid
PLGA	Polylactic glycolic acid
PLLA	Poly (L-lactide)
PMMA	Polymethyl methacrylate
PSI	Patient specific implant
ROS	Reactive oxygen species
RP	Rapid prototyping
RPM	Rotation per minute
S. aureus	*Staphylococcus aureus*
SBF	Simulated body fluid
SLA	Stereolitography
SLM	Selective laser melting
SLS	Selective laser sintering
SSE	Single screw extruder
STL	Standard tessellation language
TA	Teichoic acid
TE	Tissue engineering
TCP	Tricalcium phosphate
TGA	Thermogravimetric analysis
TSE	Twin screw extruder
UHMWPE	Ultra-high molecular weight polyethylene
WTA	Wall teichoic acid
ZnO	Zinc oxide

PENYEDIAAN DAN PENCIRIAN CETAKAN 3D KOMPOSIT POLIAMIDA 6 UNTUK REKONSTRUKSI KRANIOFASIAL

ABSTRAK

Implan spesifik pesakit diperlukan kerana kecacatan kraniofasial kebiasaannya unik dan bergantung kepada kondisi anatomi pesakit. Pencetak 3D berasaskan FDM boleh digunakan untuk memenuhi keperluan tersebut. Bagaimanapun bahan suapan yang terdapat secara komersial adalah tidak serasi dan kurang integriti mekanikal yang menghalang penggunaannya. Kajian ini berhasrat untuk membangunkan bahan suapan baru berasaskan poliamida 6 untuk rekonstruksi kraniofasial. Poliamida 6 telah disebatikan dengan gentian karbon dan zink oksida sebelum proses fabrikasi filamen dan pencetakan 3D. Kesan penambahan gentian karbon serta gentian karbon/zink oksida hibrid ke atas sifat fizikokimia sebatian serta sifat mekanik dan biologi cetakan 3D telah dinilai. Suhu peleburan tidak dipengaruhi oleh penambahan bahan pengisi, bagaimanapun kadar aliran leburan, tegangan, mampatan dan kekasaran permukaan komposit poliamida 6 meningkat dengan lebih baik. Komposit berkenaan juga mempamerkan sifat keliatan yang lebih baik daripada poliamida 6 tanpa pengisi selepas 60 hari rendaman di dalam larutan cecair badan tersimulasi walaupun menyerap lembapan yang tinggi. Kebolehhidupan sel *osteoblast* adalah lebih daripada 70% selepas pendedahan kepada ekstrak komposit pada kepekatan 50, 25, 12.5 dan 6.25 mg/ml. Komposit tersebut juga menunjukkan kesan antibakteria yang ketara terhadap bakteria Gram-positif *Staphylococcus aureus* dan Gram-negatif *Pseudomonas aeruginosa*. Walau bagaimanapun, kesannya adalah terpilih dan lebih ketara pada *S. aureus*. Bahan suapan filamen berasaskan poliamida 6 yang baru dibangunkan serasi untuk digunakan dengan pencetak 3D berasaskan FDM. Dengan

ciri-ciri mekanik dan biologi yang dipertingkatkan, komposit yang dibangunkan berpotensi digunakan untuk rekonstruksi kraniofasial.

PREPARATION AND CHARACTERIZATION OF 3D PRINTED POLYAMIDE 6 COMPOSITES FOR CRANIOFACIAL RECONSTRUCTION

ABSTRACT

Craniofacial defect is typically unique and depend on the anatomical condition of the patient for which patient specific implant (PSI) is desirable. The FDM based 3D printer could be utilised to cater the needs. However, the commercially available feedstock is bio-incompatible and lack of mechanical integrity which hinder the application. This study aimed to develop a new polyamide 6 based filament feedstock aiming for craniofacial reconstruction. Polyamide 6 was compounded with carbon fibre and zinc oxide prior to filament feedstock fabrication and 3D printing processes. The effect of carbon fibre as well as hybrid carbon fibre/zinc oxide incorporation on the physicochemical properties of the compounds as well mechanical and biological properties of the 3D printed parts were assessed. The melting temperature of the composites were not affected by the filler incorporation, however, the melt flow rate, tensile, compressive and surface roughness properties of the PA 6 composites increased appreciably. The composites also exhibited better toughness properties than unfilled PA 6 after 60 days of immersion in simulated body fluid despite of high moisture absorption. The viability of osteoblast cells were more than 70% following treatment with extracted composites at concentrations of 50, 25, 12.5 and 6.25 mg/ml. The composites also demonstrated appreciable antibacterial effect against Gram-positive and negative bacteria of *Staphylococcus aureus* and *Pseudomonas aeruginosa*. However, the effect was selective and more pronounced in *S. aureus*. The newly developed polyamide 6 based filament feedstock is compatible to be used with FDM based 3D printer. With enhanced mechanical and biological properties, the developed composites are potential to be used for craniofacial reconstruction.

CHAPTER 1

INTRODUCTION

1.1 Background of the study

Craniofacial reconstruction refers to a surgical procedure to restore impairment of skull and facial shape resulted from trauma, infection or congenital malformation. Typically, patient's own bone will be used to reconstruct the defect part in order to reduce the operation cost and waiting time (Lemee *et al.*, 2013). However, it is restricted to a smaller defect which could be covered by autologous bone. Patients with large craniofacial defect (Figure 1.1) require to undergo surgical procedure using biomaterial implants to correct the deformities (Cabraja *et al.*, 2009). While general implant could be used for reconstruction of other bony parts, patient specific implant (PSI) is desirable for craniofacial part as the defect varies, unique and highly depends on the anatomical condition of the patient. PSI is introduced by means of obtaining a fit customised implant with high accuracy which could shorter the rehabilitation process and minimise the cost (Maniar and Singhi, 2014).

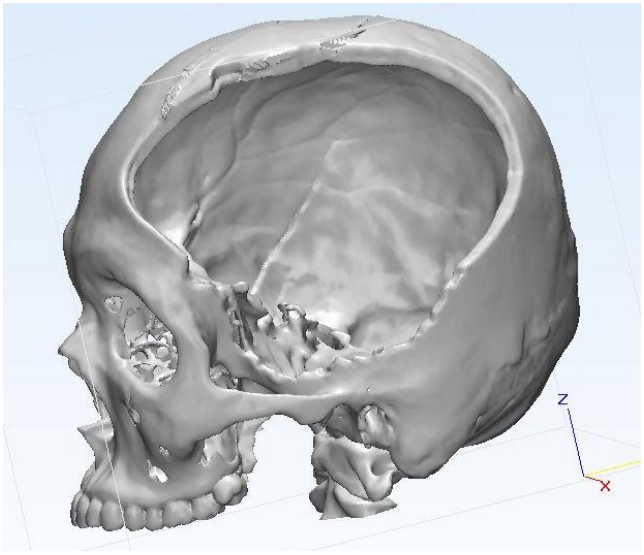

Figure 1.1: Example of large craniofacial defect

The advancement of computer aided design (CAD) and computer aided manufacturing (CAM) offers a great potential which could be utilised to achieve an accurate, fast and reliable implant. Datasets generated from medical imaging modalities such as computed tomography (CT) and magnetic resonance imaging (MRI) could be transferred to CAD environment where the virtual reconstruction could be done and analyse before sending to CAM device for implant fabrication. However, a biocompatible feedstock is required for the implant fabrication purposes. An almost similar flow was readily established for bio-model preparation to be used for preoperative planning purposes (Figure 1.2).

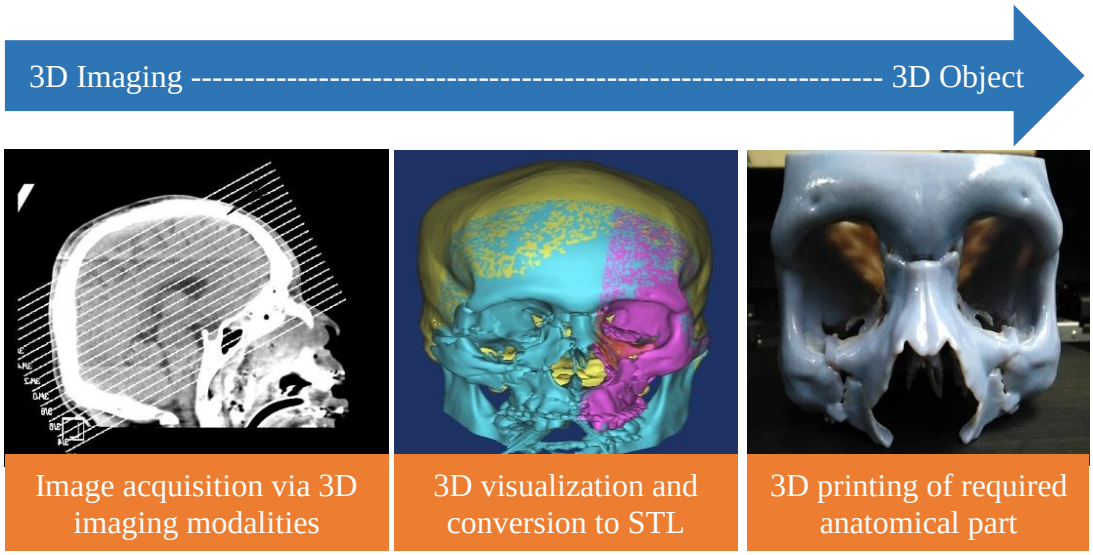

Figure 1.2: Flow of bio-model preparation

Current available PSI is generally made from titanium, polyether ether ketone (PEEK) and nylon. It is processed via high-end additive manufacturing (AM) techniques such as direct metal laser sintering (DMLS), selective laser melting (SLM) or selective laser sintering (SLS). 3D printing is a method of choice over conventional manufacturing process such as injection moulding as it permits the fabrication of complex anatomical structure at an economically effective cost. Besides, its manufacturing flexibility that requires zero tooling setup could shortened the

production cycle and quickly response to the customer demands. The breakthrough in AM towards an affordable, user friendly yet accurate techniques resulted in an emerging of fused deposition modelling (FDM) based 3D printer. The technology was inspired by FDM techniques, a technique which originally created and patented by Stratasys Corporation. FDM based 3D printer (Figure 1.3) is built to fit with typical filament diameter of 1.65 to 1.85 mm. The installed thermoplastic filament is guided through the liquefier section by a rotated drive gear and heated to a required temperature. FDM based 3D printer creates a 3D object by depositing molten thermoplastic onto a build plate, layer by layer (Figure 1.4). The nozzle movement is controlled by G codes generated by a slicing software. The build plate will be gradually lowered with every single layer deposition and the movement continues until the desired build finish.

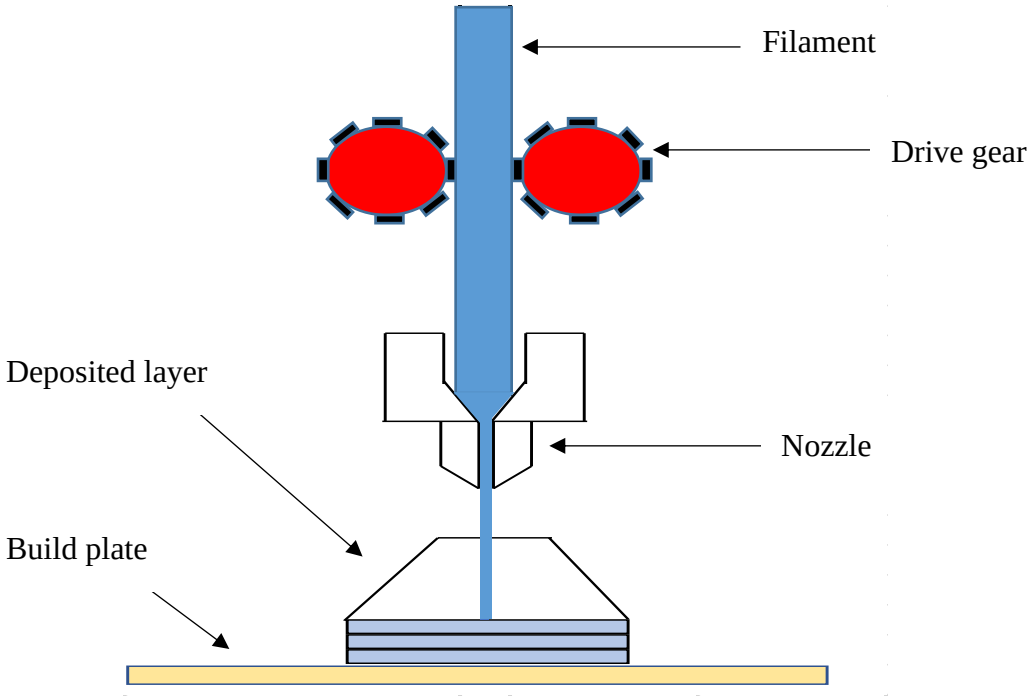

Figure 1.3: Schematic diagram of FDM based 3D printer

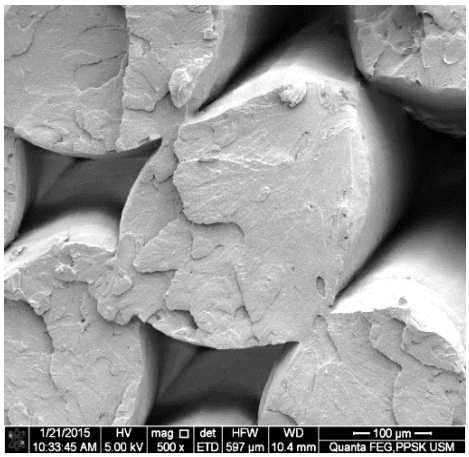

Figure 1.4: Deposited layers visualised via scanning electron microscope

This technique could be manipulated to fabricate an affordable implant by integrating with an appropriate FDM feedstock to be suited for craniofacial reconstruction application. Meanwhile, acrylonitrile butadiene styrene (ABS) and polylactic acid (PLA) are the earliest and most common FDM feedstock which later applied for optimisation of FDM based 3D printer. Up to now, ABS remains prominent feedstock in this techniques referring to several trials in alteration of properties to suit certain application (Nikzad *et al.*, 2009; Torrado *et al.*, 2015). Although ABS has been used as disposable medical device (Toray, 2019), its biocompatibility for biomedical implant is still debatable.

On the other hand, polyamide is a promising biocompatible engineering polymer. Polyamide was initially used as a suture material before being successfully applied as nasal splint, orbital floor as well as condylar implant (Ulrich, 1957; Breitbart and Ablaza, 2007; Li *et al.*, 2011), indicating its biocompatibility. Although the manufacturing process was not highlighted in the previous success attempts, it should be noted that the fabrication of complex condylar implant was realised using SLS technique, which indicated that 3D printing technique could be used to produce a

reliable, robust and tough polyamide implant. In addition, 3D printed polyamide exhibits outstanding tensile and elongation at break as compared to ABS indicating its superior bonding adhesion between the successive layers (Lederle *et al.*, 2016) that the stated advantages could be a strong basis to further explore the potential of FDM 3D printed polyamide for biomedical application.

Multifunctional material is current leading topic in biomaterial development. While mechanical integrity is one of the requirements for implantation material, an additional value such as antibacterial properties are desirable to bombard microbial colonisation. Incorporation of antibacterial agent is desirable for material to be used in health and medical application. It is even more crucial for application in sterile condition. Antibacterial agent works by inhibiting the bacterial growth and at the same time creates no harm to the host.

1.2 Problem statements

The fabrication of low volume or small batches of patient specific implant could not be effectively realised via conventional manufacturing process as it will result in high cost. In addition, conventional implant fabrication method such as injection moulding restricts the ability to fabricate complex patient specific implant as it requires the preparation of specific mould and tooling which may delay the responsiveness to the patient's need.

On the other hand, the utilisation of high-end AM technology for metallic or polymeric implant fabrication is undoubtedly able to produce an accurate and high-quality implant. However, the end cost is too high that it could not be borne by all patients, thus hinder the application. Moreover, the usage of metallic implant is often

associated to implant loosening and displacement due to inhomogeneous stress distribution resulted from stress shielding effect (Niinomi and Nakai, 2011) which make it not preferable, that a new biomaterial in combination with an affordable manufacturing process could be suggested to counter the needs.

The emergence of FDM based 3D printer could be manipulated to fabricate an affordable patient specific implant. However, the currently available FDM feedstock mainly relies on ABS and PLA. The usage of ABS for biomaterials application is controversial as it is reported to be associated to chronic white blood cancer in human (Sathiakumar *et al.*, 2015). PLA however is biodegradable which is not suitable for permanent implant application. Lack of variation in currently available FDM feedstock impede the application for craniofacial reconstruction. Despite of many advantages such as user friendly and relatively accurate technique, the bio-incompatibility of the FDM feedstock and insufficient mechanical properties of the 3D printed parts are among the prominent drawbacks of FDM which hinder the application.

While the mechanical properties enhancement in FDM model could be strategised via modification of the machine, optimisation of process parameter and additional structural support in 3D printed model, development of new materials which are suited with the system are often desirable. It is a fact that several trials have been performed to introduce biocompatible FDM feedstock such as hydroxyapatite (HA) filled polycaprolactone (PCL) and hybrid ZrO_2/HA filled polyamide 12 (PA 12) (Haq, 2015; Rahim *et al.*, 2015a). However, the obtained mechanical properties are still lower than projected for compact bone replacement. Meanwhile, an attempt was initiated by introducing HA/ZrO_2 filled polyamide 6 feedstock for similar purposes. Although 3D printed HA/ZrO_2 filled polyamide 6 shows better mechanical performance than HA filled polyamide 12 (Rahim, 2018), the combination of

hygroscopic polyamide 6 with calcium phosphate-based filler of HA could lead to high moisture absorption as HA contains hydroxyl group, that an appropriate filler combination could be suggested to maintain its mechanical integrity in an actual clinical setting.

Calcium phosphate-based fillers are often incorporated into the selected matrix to be developed as a filament feedstock for bone replacement, in order to improve the biological interaction between implant and surrounding bone. However, the fillers are incapable to bombard microbial colonization. Moreover, it should be noted that the infection rate following craniofacial reconstruction procedure is alarming that it could go as higher as 10.98% (Kwarcinski *et al.*, 2017).

Up to date, the development of new materials for FDM are mainly performed via incorporation of either particulate (Nikzad *et al.*, 2011; Singh and Singh, 2014) or fibrous (Tekinalp *et al.*, 2014) filler into a polymer matrix in order to enhance the mechanical properties. However, both highly particulate and fibrous filled polymer to be used as FDM feedstock seem to disrupt the printing mechanism which led to an adverse effect on mechanical properties (Tekinalp *et al.*, 2014; Olsson *et al.*, 2017). Besides, although the effect of particulate or fibrous filler incorporation into a polymer matrix are well discussed in the literature, the reinforcement effect via hybridisation of particulate and fibrous filler in FDM printed part is yet to be explored. Moreover, the layering techniques applied in FDM is totally different from conventional polymer processing, which a systematic investigation on the morphological aspect is essential to create a mechanically relevant FDM 3D printed part.

1.3 Justification of the study

This research is essential to introduce a new polymer composite biomaterials-based filament feedstock which could be 3D printed via FDM technique. The 3D printed polymer composite is projected to have adequate mechanical properties with antibacterial properties as an added value for craniofacial reconstruction application. The success to print the respected polymer composite via FDM technique will open a possibility for patients with craniofacial deformities to regain their regular cosmesis at an affordable cost.

1.4 Objectives of the study

This study was carried out to prepare potential polymeric biomaterials to be compatible with FDM based 3D printer. Besides, mechanical, physical, chemical and biological characterisations were also performed to evaluate the suitability of the polymer composites for craniofacial reconstruction.

1.4.1 General objective

To develop a new polyamide 6 based filament feedstock and evaluate the physicochemical, mechanical and biological properties of unfilled, carbon fibre (CF) filled and hybrid CF/ZnO filled polyamide 6 (PA 6).

1.4.2 Specific objectives

1. To compound the CF and CF/ZnO with PA 6 and to investigate the thermal properties, melt flow rate characteristic and functional group of respected PA 6 composites.

2. To fabricate the compounded composites into a filament form to be used as feedstock for FDM based 3D printer and to assess the effect of CF and hybrid CF/ZnO incorporation on the mechanical and morphological properties of 3D printed PA 6 composites.
3. To evaluate the effect of simulated body fluid immersion on moisture absorption and mechanical degradation of 3D printed PA 6 composites.
4. To assess the effect of CF and hybrid CF/ZnO incorporation on the cytotoxicity properties of 3D printed PA 6 composites.
5. To investigate the antibacterial properties of the developed composites against *Staphylococcus aureus* and *Pseudomonas aeruginosa.*

1.5 Hyphotheses

1. The filler incorporation will have no significant difference on melt flow rate of polyamide 6 composites.
2. There are no significant differences in mechanical properties, mechanical degradation, cell viability and antibacterial properties of unfilled, CF filled and hybrid CF/ZnO filled PA 6 composites.

CHAPTER 2

LITERATURE REVIEW

2.1 Craniofacial bone reconstruction

Craniofacial bone reconstruction is a surgical intervention to repair craniofacial bone defect in order to solve function and cosmetic issues. The success of bone reconstruction helps patient to repair certain functional impairment, regain regular cosmesis, relief psychological damage as well as boost self-confidence in order to mix and function in a community. Reconstruction of craniofacial bone could be strategised via several methods such as autograft, xenograft, allograft and alloplast augmentation techniques, as summarised in Figure 2.1.

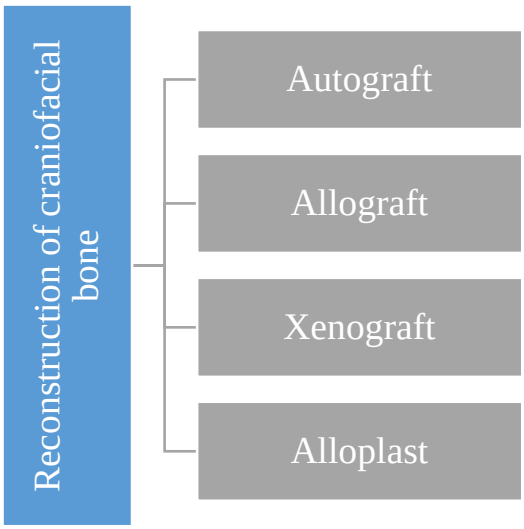

Figure 2.1: Augmentation method for craniofacial bone reconstruction

While there are various techniques available, an autograft is often a preferable method considering the availability of the patient's own bone. Autograft is a transplantation of identified bony part from a patient's own bone to cover the defect. However, details consideration needs to be made by looking at the defect size and

contour. Besides, the patient needs to be fit for multiple or prolong surgeries. Recently, a hybrid method is introduced for a patient who underwent decompressive craniectomy procedure in order to reduce cost and provide a more aesthetical feature. While the resected bone is commonly abandoned, the technique utilises the patient's own bone where alloplastic material of PMMA is topped up to the partial bone flap to produce hybrid cranioplasty plate (Hueh *et al.*, 2016). Nevertheless, tissue bank facility is required to initially preserve the resected bone prior to the surgical procedure.

On the other hand, the allograft is another option, which the bone from the donor site is being transferred to the patient. This procedure can reduce operating time as well as less pain. However, the patient may need to wait for the donor availability and possibly exposed to the donor site morbidity after implantation. Both autograft and allograft use an autologous bone either from patient or donor site. Next option is xenograft, which involves bone transfer from different species to human. Though this grafting method is debatable, xenograft is demonstrated to be a reliable grafting material following 100% success in sinus augmentation (Rahman *et al.*, 2014).

The final option is alloplast, which considers synthetic biomaterials to be implanted in a patient with a moderate and large craniofacial defect. While all listed methods are reputable techniques, the usage of autologous bone in both autograft and allograft may require a secondary operation, which involves multiple specialties resulted in a higher cost and extra pain to the patient. Moreover, a complication such as a flap displacement due to bone resorption (Bobinski *et al.*, 2013) and high complication in a paediatric patient (Martin *et al.*, 2014) are among the main concern raised by clinicians. In xenograft augmentation method, the ethical issue, as well as the fear of the microorganism transmission (Collignona and Purdy, 2001), remains controversial, which hinder this technique from being applied in a clinical setting.

Therefore, least controversial alloplastic biomaterials could be an option to be expanded for craniofacial bone reconstruction, which attention needs to be given to the materials selection and processing techniques to be established as an alternative framework.

2.2 Synthetic biomaterials for craniofacial bone reconstruction

Synthetic biomaterials for craniofacial bone reconstruction could be classified into four groups of metallic, polymeric, ceramics and composite materials. The details classification is summarised in Figure 2.2. Polymer group is divided into resorbable and non-resorbable, while ceramic is further classified into bioinert, bioactive and bioresorbable.

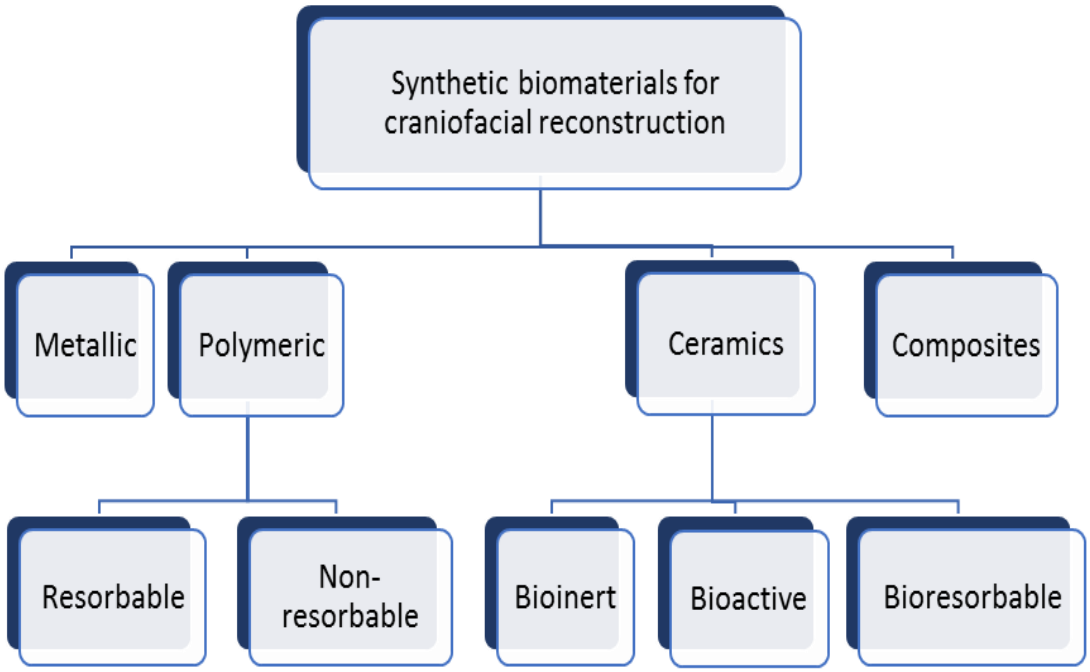

Figure 2.2: Classification of synthetic biomaterials for craniofacial reconstruction

Despite this classification, biomaterials in general should meet following requirements (Figure 2.3);

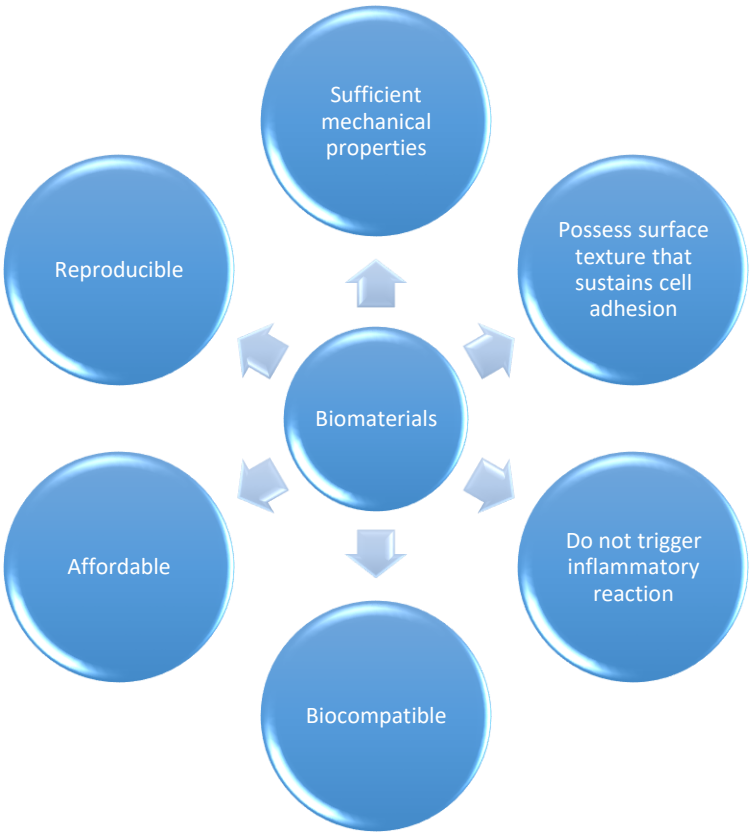

Figure 2.3: Requirement of biomaterials as defined by Ramakrishna *et al.* (2001); Hench and Thompson (2010); Bruno Zanotti *et al.* (2016)

The mechanical properties of biomaterials technically differ and merely depends on the type of materials, structure and processing technique. Metallic materials typically exhibit high mechanical properties as compared to other material such as polymer. Meanwhile, porous structure naturally correlates to a low mechanical properties (Yu *et al.*, 2008). Besides those two factors, certain processing techniques such as injection moulding and 3D printing may greatly influence the mechanical properties of the biomaterials (Rahim *et al.*, 2016). Regardless of the aforementioned factors, biomaterials should exhibit sufficient mechanical properties in order to regularly function in either load bearing or non-load bearing condition.

Adherence of cells to the surface of implant is critical as it determines the longevity and survival of implant. Metal implant for example possesses smooth surface which hinder the adherence of cells that surface modification usually needs to be carried out. However, introduction of roughness to the surface of metallic material results in adverse effect to the mechanical properties (Riemer *et al.*, 2014). While the roles of surface texture in determining the cell adherence is widely reported and established fact, a contrast finding reveals that surface energy is more influential than surface texture on cell adhesion and proliferation (Hallab *et al.*, 2001). Besides cell adhesion, bacterial adhesion also proportionally relates to the roughness of the materials (Dantas *et al.*, 2016) that additional measures need to be considered to prolong the lifespan of implant.

Inflammation is an auto response of the immune system which typically triggers due to various factors including the presence of toxic compounds. Though the response might differ and depend on the location and stimulus, common inflammatory response starts with recognition of harmful stimuli by certain receptors, follows by activation of pathways which then induce the release of inflammatory marker prior to recruitment of inflammatory cells (Chen *et al.*, 2018). Though inflammation is often mistakenly thought as healing crisis, the auto response might results in delayed wound healing (Eming *et al.*, 2007). The healing disturbance condition is more susceptible to infection which could end up to implant failure, that the ideal biomaterial should not trigger inflammatory reaction.

Biocompatibility is another crucial factor which determines the success of the implant. Biocompatibility refers to the ability of biomaterials to regularly function in the host tissue without eliciting any undesirable local and systemic effect (Schmalz and Bindslev, 2009; Perrotti *et al.*, 2017). In order to be used as implant, rigorous

biocompatibility testing such as cytotoxicity, genotoxicity and carcinogenicity need to be performed to make sure that the material is non-hazardous to the biological system. Besides, the biomaterials should also be reproducible and processable by any processing techniques and most importantly, affordable to all. Meanwhile, the key timeline of the significant event in the use of biomaterials for craniofacial reconstruction is summarised in Figure 2.4.

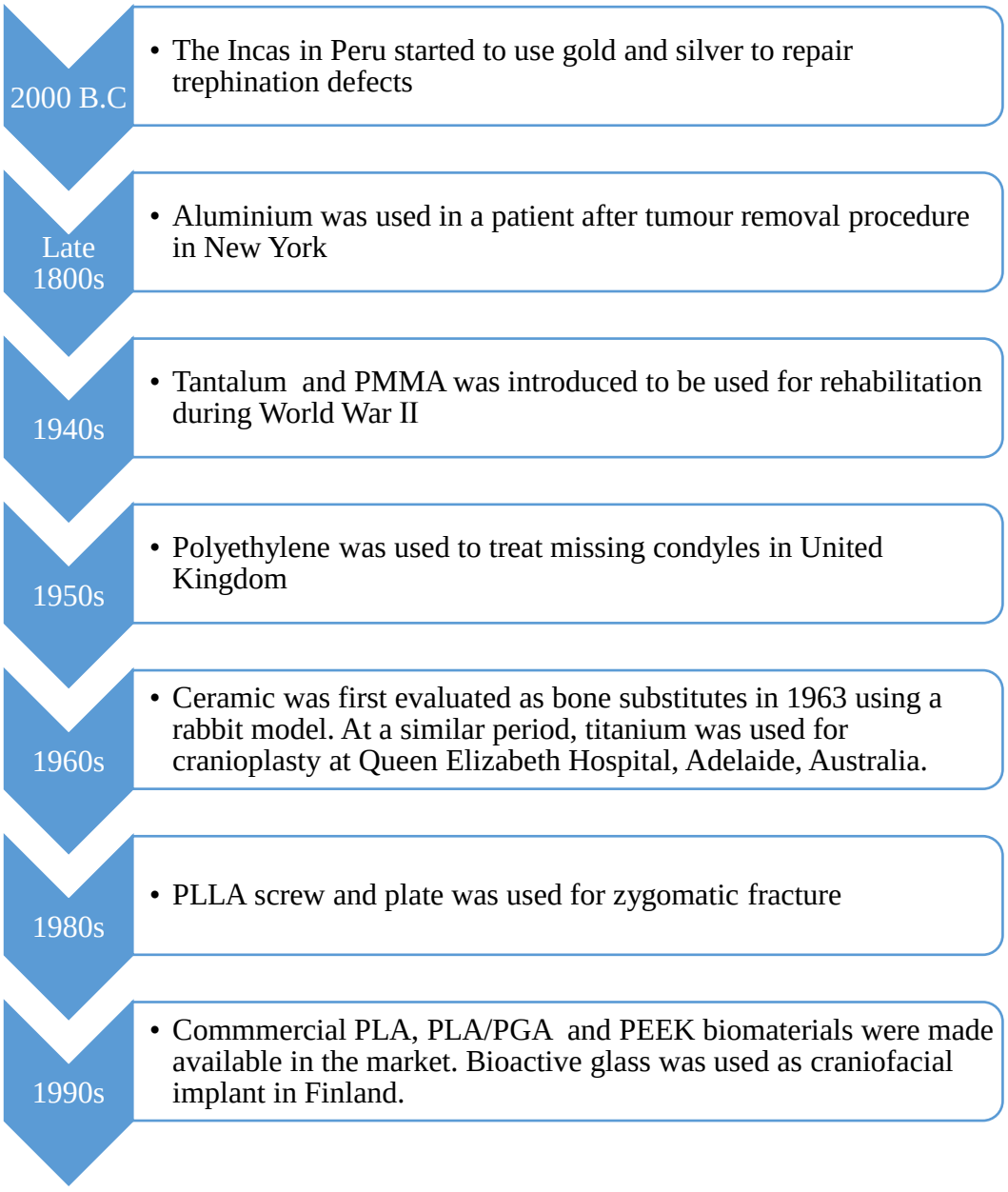

Figure 2.4: Timeline of significant event in application of biomaterials for craniofacial reconstruction

2.2.1 Metallic materials

Metallic material such as gold and silver has started to be used as early as 2000 B.C by the Incas in Peru to cover trephination defects (Abhay and Haines, 1997). Trephination or drilling of a skull is an ancient procedure to relief headache. However, it may occasionally relate to the mystical ritual to release an evil spirit (Rawlings and Rossitch, 2014). Dates back, the choice of material to repair the trephination defect is suggested to be associated with social status, where a precious metal is used for nobility.

Metal is then reappeared in the late 1800s due to its high strength, malleable and sterilizable, in which aluminium is recorded to be used in one patient after frontal lobe tumour removal in the late 1800s (Booth and Curtis, 1893). However, the patient died ten days later due to multiple complications. Further study on aluminium cranioplasty was recorded with 61 patients recruited in Missouri, Columbia. Though aluminium was reported to cause epilepsy in animal, no similar symptom was indicated in the implanted patients. Moreover, the results were cosmetically and functionally satisfactory. However, the researchers did admit that a similar complication could still arise (Black *et al.*, 1968).

Other than the cranial part, aluminium was also used to repair the supraorbital ridge and its adjacent frontal bone (SP, 1978). The aluminium was malleable enough to be bent and reconstruct the complex part. Follow up was done for a minimum of 3 years and a maximum of 19 years. No complication was reported in all 6 patients. Despite several documented success trials using aluminium, there is no updated progress in recent literature on the usage of aluminium for craniofacial bone repair. Indirectly, with several controversial reports on the association of aluminium with

alzheimer, autism and its effect on human (Stephens and Jolliff, 2015; Mold *et al.*, 2018; Klein, 2019), a clear policy should be ruled to suspend the usage of aluminium in a physiological environment.

Cranial fracture represents a significant problem during World War II due to the frequency of occurrence. The urgency to keep as many military personnel on duty during the crucial times while simplifying the cranioplasty procedure end up with the usage of tantalum. Tantalum is a dense yet malleable metal. Unlike controversial aluminium, tantalum is proved to be inert with no sign of bone or tissue reaction after implantation in both dogs and rabbits (Burke, 1940). However, the breakthrough of the tantalum usage in cranioplasty was based on the success of implantation in 11 cats with the formation of scar tissue surrounding it, which theoretically hinder the implant displacement (Pudenz, 1943). Although autograft is considered a routine, the usage of tantalum simplified the process as it could be cut and shaped and implanted in a single step procedure. The patient was able to return to duty as early as two months after the operation (Fulcher, 1943). As time goes by, tantalum is no longer retrievable as a craniofacial bone substitute. However, its bioactivity and low modulus open a new possibility towards a creation of bone graft substitute to be used in an orthopaedic application (Levine *et al.*, 2006).

On the other hand, titanium was initially established as an orthopaedic implant before being expanded to the craniofacial region. It was first reported to be successfully applied to 7 patients with craniofacial defect at Queen Elizabeth Hospital, Adelaide, Australia (Simpson, 1965). Several significant advantages of titanium over tantalum are its radiotranslucency, adequate strength with the thickness and less expensive. Titanium is less malleable; however, it’s still could be shaped into plates according to the contour of the skull. Up to date, titanium could be considered as a

prominent metallic biomaterial for craniofacial reconstruction based on the enormous studies which indicate the success of titanium usage in craniofacial reconstruction related surgical procedure.

Despite its high success rate, one significant disadvantage of using titanium other than its relatively expensive in cost is the stress shielding effect. Although the strength of titanium is comparable to the human cortical bone, its elastic modulus is extremely higher than bone, that the stress could not be homogenously distributed and shared. This phenomenon could result in an upsurge in bone porosity which could further lead to an implant displacement and fracture of surrounding bone (Niinomi and Nakai, 2011). Therefore, several strategies such as porous plate design as well as modification of crystalline structure of titanium need to be implemented in order to develop titanium with bone-like elastic modulus (Wang *et al.*, 2019).

2.2.2 Polymeric materials

Polymeric biomaterials are vastly utilised in bio-medical application. Nevertheless, polymeric biomaterials for craniofacial reconstruction are still limited and focused on a certain type of polymer. It is due to the lack of mechanical integrity which hinders the polymer from being applied as bone replacement material. Polymeric biomaterials for craniofacial reconstruction could be classified into a resorbable and non-resorbable polymer. The resorbable polymer is prevalent for application in paediatric patients, while the non-resorbable polymer is used in elderly

patients as permanent implants. Classification of polymeric biomaterials and its example are summarised in Figure 2.5.

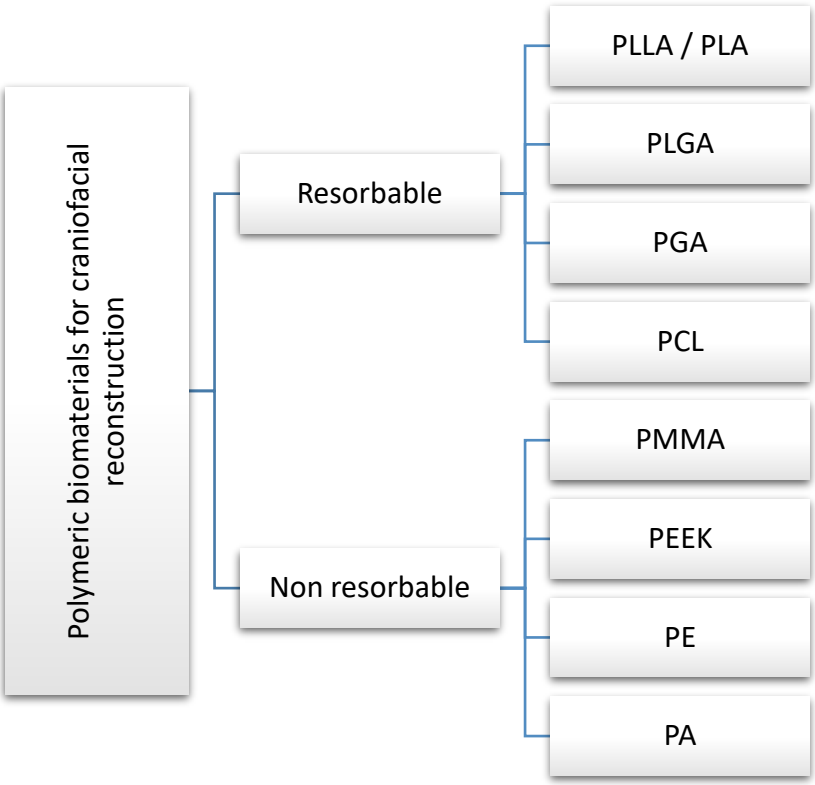

Figure 2.5: Classification of polymeric biomaterial and its examples

2.2.2 (a) Resorbable polymer

The term resorbable polymer refers to a polymer which degrades in a physiological environment with the elimination of by-product or complete resorption by host tissue (Liu *et al.*, 2017). Poly(L-lactide) (PLLA), polylactic acid (PLA), polylactic glycolic acid (PLGA), polyglycolide acid (PGA) and polycaprolactone (PCL) based materials are the most common temporary implants for paediatric patients. It should be noted that PLLA is the optical isomer of PLA. The growing interest in the biomedical application resulted in the emerging of PGA and PLA as the

earliest candidates to be evaluated as resorbable sutures (Herrmann *et al.*, 1970; E.Cutright *et al.*, 1971).

The attempt of expanding the resorbable polymer to a craniofacial region was performed by applying PLLA based screws and plate to stabilise the zygomatic fractures in ten patients, which resulted in undisturbed fracture healing (Bos *et al.*, 1987). However, three years after the operation, four patients returned due to an intermittent swelling surrounding the implantation site, which forced the team to recall another six patients. The swelling was resulted by non-specific foreign body reaction towards the degraded PLLA implant. The finding was supported by the detection of crystal-like PLLA in the cell's cytoplasm (Bergsma *et al.*, 1993).

Commercial PLA and PLA/PGA copolymer implant system were made available in 1996 to fulfil the growing demand (Moe and Weisman, 2001). The implant in a sheet form is malleable that it could be recontoured following the defect site by placing in a 56ºC saline bath. The effectiveness of the resorbable implant to provide a temporary fixation could be seen in paediatric patients with craniosynostosis (Eppley, 2002). The resorbable polymer is no doubt an ideal solution for the paediatric patient as the resorption of the materials in the human body environment would allow bone growth and hinder secondary operation for removal purposes. Timely degradation and resorption of the polymeric material enable efficient load transfer, which induces the formation of new bone and consolidates the bony defect. The polymer resorbs via two stages which includes splitting of polymer chains into monomers by hydrolysis, which then broken down into CO_2 and H_2O before eventually excreted. However, a rare case involving inflammation due to hypersensitivity reaction with the degradation of by-product as well as remaining of unresorbed material is an isolated issue which requires further clarification and attention (He and Shi, 2017).

2.2.2 (b) Non-resorbable

Polymethyl methacrylate (PMMA), polyethether ketone (PEEK), polyethylene (PE) and polyamide (PA) are the prevalent non-resorbable polymeric material for craniofacial reconstruction. The non-resorbable polymer is a repetition of long hydrocarbon chain which yields in strong molecular bonding. Among the listed non-resorbable polymer, PMMA is the oldest materials for craniofacial reconstruction. It has been used since 1943. In the early years during the evaluation stage, while repairing the head injuries, PMMA was also used to investigate intracranial phenomena in macacus monkeys due to its transparent nature (Shelden *et al.*, 1944).

PMMA is an affordable material, possesses adequate mechanical properties, exhibits excellent functional and cosmetic results at long-term follow-up and a material of choice when an autologous bone is not available (Marchac and Greensmith, 2008). Despite the stated advantages, PMMA suffers from its apparent brittleness and shrinkage (Hamad *et al.*, 2016). In addition, PMMA is well known to release heat due to an exothermic reaction during polymerisation, which may harm surrounding bone tissue. The temperature inside the PMMA implant with adjacent tissue being exposed is more than 50ºC (Golz *et al.*, 2010), that pre-operative implant preparation is recommended. However, sterility is another area of concern when dealing with a medical device outside the operation theatre. While it should not be compromised, the effect of available sterilisation methods such as autoclave, hydrogen oxide gas plasma, ethylene oxide and γ-irradiation on the mechanical properties of PMMA implant need to be elucidated to guarantee the survival of implant for long term usage (Münker *et al.*, 2018).

Another polymeric material which is immensely used is polyethylene (Ridwan-Pramana *et al.*, 2015). Although the approval by the Food and Drug Administration (FDA) for commercial usage was only received in 1984 (Ellis and Messo, 2004), the initial effort of using polyethylene for craniofacial reconstruction was documented as early as 1954 to augment the congenitally missing condyles of 12 year old white girl in England (R.Prowler and Glassman, 1954). On the other hand, the prevalent commercial high-density polyethylene (HDPE) based and ultra-high molecular weight polyethylene (UHMWPE) based implant are being sold under a trade name of Medpor and SynPOR, respectively. The polyethylene for craniofacial reconstruction is normally engineered to be porous in structure with pore size of 100 to 200 µm to allow tissue in-growth. Although produce by several companies, polyethylene-based implant is well-known for its flexibility yet strong enough to be used for reconstruction of craniofacial region. While PMMA and polyethylene are established materials for the purposes, the usage of other variation of polymeric materials for craniofacial reconstruction is still limited.

On the other hand, polyether ether ketone (PEEK) is a phenomenal high-performance polymeric material. It has started to be commercialised in April 1998 as a biomaterial by Victrex, a company based in the United Kingdom (Green and Schelegel, 2001). The mechanical properties of PEEK resemble the properties of cortical bone (Petrovic *et al.*, 2006) and are preferable than titanium due to its lightweight. Moreover, it is rarely associated with artefacts in magnetic resonance tomography (MRT) images as typically showed by titanium (Maier, 2009). PEEK did not induce any new bone formation when implanted in rat (Li *et al.*, 2005), that it can be considered as inert polymer. The use of PEEK for craniofacial reconstruction was first documented in 2007 in an attempt to reconstruct large and complex orbito-fronto-

temporal defect. The attempt was performed as a counter treatment for failed and infected reconstruction site resulted in a purulent discharge and wound dehiscence. The initial reconstruction was conducted using titanium with PMMA (Scolozzi *et al.*, 2007). Short follow up of one year revealed that the patient seemed to regain regular facial cosmesis.

Polyamide or typically known as nylon is one of the widely used engineering thermoplastic. While there are various types of polyamide available, polyamide 6,6 for example, was first invented by Carothers in 1935 during his early career at DuPont, U.S.A. The evolution of Polyamide was followed by the discovery of polyamide 6 by Paul Schlack at IG Farben, Germany, in his attempt to unviolate the patented route. Around a similar time, Toray Japan also announced success in synthesising Polyamide 6. In the early years, the production of Polyamide 6,6 was dominated by U.S, while polyamide 6 was mainly produced by Europe and Japan (Sastri, 2014). Early literature on polyamide for biomedical application were written in German language using a polyamide-based material called supramid. Although polyamide was first established as a suture material, it was then expanded to a craniofacial region. Among the initial attempt was a flat saddle nose correction using a supramid splint (Ulrich, 1957). Besides, polyamide has also been successfully utilised as an orbital floor implant (Breitbart and Ablaza, 2007). Polyamide was also implanted as a condylar implant in one patient with a condylar defect after an aesthetic mandibular angle reduction procedure (Li *et al.*, 2011). The motivation of using polyamide is due to the presence of polar molecular structure (CO-NH) which imitates the structure of collagen, a crucial factor that induces the osteoblast. While the presence of CO-NH seems to give an advantage in term of biocompatibility, it is also contributing to the hygroscopic nature of the material that a storage condition needs to be well defined.

2.2.3 Ceramics

Ceramics has long been a subject of interest for bone reconstruction due to its excellent mechanical properties, thermodynamically stable, etc. Ceramics, in general, could be classified into bioinert, bioresorbable as well as bioactive based on its response to the physiological environment. Despite having superior properties, the first trial of utilising ceramics for bone substitute was only reported in 1963. The evaluation used sintered porous alumina, silica, calcium carbonate and magnesium carbonate mixture which then impregnated in an epoxy resin and implanted in a rabbit model (Smith and Elgin, 1963). The success of this trial has embarked the usage of ceramics specifically for bone replacement as it could achieve the strength of bone if it is prepared at certain porosity. The classification of ceramics and its representative are summarised in Figure 2.6.

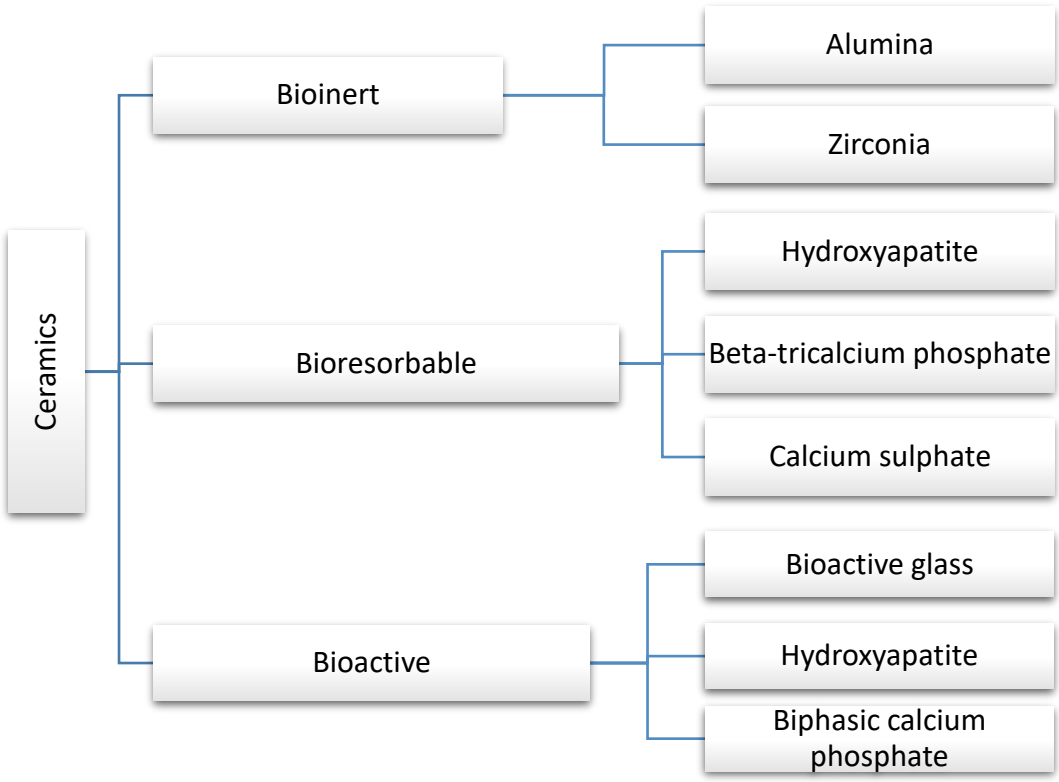

Figure 2.6: Classification of ceramics and its examples