

# Characterization tools

## 1. Biomaterials analysis:

Technique		Description	Applications	Scale	Features	Advantages	Disadvantages
Optical Microscopy (光學顯微鏡)	Fluorescence Microscopy (螢光)	Uses fluorescence (螢光) to image specific structures in cells or tissues by using dyes (染料) that emit light at certain wavelengths	-Cell imaging -Histology (組織學) e.g. proteins, bacteria, gene expression	0.2μm - 1μm	Can observe live cells and dynamic(動態) processes	-Cheap -Can observe live cells	-Limited range of focus
	Confocal-3D (limitation: depth)						
Scanning Electron Microscopy (SEM) (掃描電子顯微鏡)		Uses a focused electron beam to scan a sample surface, providing high-resolution images and information about surface topology.	-Surface structure -Morphology analysis (形態分析)	0.1nm-10nm	Requires vacuum environment, samples must be dry and dead	-High spatial resolution -Good depth of field -3D-like images	Requires a vacuum – not suitable for biological tissue, unless fixed/dried
Transmission Electron Microscopy (TEM) (透射電子顯微鏡)		Uses transmitted(透射) electrons to produce high-resolution images, suitable for internal structure and crystallography (晶體)	-Internal structures	<0.1nm	requires very thin sections (薄切片)	-Very high resolution -2D images	-requires very thin sections -time consuming -costly
Energy Dispersive Spectroscopy (EDS) (能量色散光譜分析)		Analyzes X-rays during electron bombardment→elemental composition	Elemental composition 與 SEM/TEM 結合使用		Combination with SEM (or TEM)	Provide both structural and composition	- no bonding, crystal structure - not sensitive to light elements

## Compared SEM with TEM

Technique	Description	Applications	Features	Advantages	Disadvantages
SEM	-Focus electron beam -Scattered electrons	Surface	-Vacuum, dry -3D	Easier sample preparation	-only surface details
TEM	-High-energy electron beam passes through -Transmitted electrons	Internal	-Thin samples -2D	Extremely high resolution	-complex sample preparation -expensive

Technique	Description	Applications	Advantages	Disadvantages
Atomic Force Microscopy (AFM) (原子力顯微鏡)	Uses a sharp probe (尖銳探針) to scan surfaces at the atomic level, providing topographical maps and measuring surface forces.	-Surface roughness -Mechanical properties	-Surface analysis -Live cell -Can measure insulator and conductor	-Slow scanning speed, small area (掃描速度慢，適用範圍小) -Contact technique, can wear sample
X-ray diffraction(衍射) (XRD)		-Crystal structure -Symmetry (對稱性)	- crystal structures - Operate in ambient conditions (no vacuum) - Relatively cheap, quick	- Overlapping - Not suitable for organic or biological materials - Requires a minimum sample size
X-ray Photoelectron Spectroscopy (XPS) (X射線光電子能譜)	Measures the binding energy (結合能) of electrons ejected by X-rays, giving information about surface chemical composition and bonding states	-Chemical composition -Bonding states	-Non-destructive, quantitative method for elemental composition -Chemical shifts: oxidation states and chemical	Complex Expensive instrumentation
Vibrational Spectroscopy (IR, Raman) (振動光譜學)	Uses infrared (紅外線)→molecular vibrations , identifying functional groups and molecular structure	-Molecular identification -chemical bonding	Non-destructive	
Fourier-Transform Infrared Spectroscopy (FTIR)		-Biomaterial structure -Tissue growth -Protein adsorption		-not very high resolution -not very common –
Energy Dispersive Spectroscopy (EDS) (能量色散光譜分析)	Analyzes X-rays during electron bombardment→elemental composition	Elemental composition 與 SEM/TEM 結合使用	Provide both structural and composition	- no bonding, crystal structure - not sensitive to light elements
Quartz Crystal Microbalance (QCM) (微量天平)	Measures mass changes on a quartz crystal surface by detecting changes in its resonant frequency (共振頻率).	-Measure a mass variation per unit area	High sensitivity for detecting small mass changes (對小質量變化高靈敏度)	
Raman spectroscopy and microRaman		-molecular structure -functional group	Can distinguish material based on chemical signatures	

## Summary (總結):

- EDS 和 XRD 適合分析金屬成分和晶相結構。
- SEM 非常適合觀察斷裂表面形貌、腐蝕特徵、以及細菌形態。
- XPS 和 FTIR 可以提供詳細的化學成分信息，適合分析表面有機物和腐蝕產物。
- QCM 和 AFM 可用於測量吸附層的厚度和組織結構。
- Fluorescence Microscopy 螢光顯微鏡有助於分析生物材料、基因表現與細菌或其他生物分子的交互作用。

## \* Questions:

### **1. A medical insurance company wants to know why a 316L stainless steel femur repair plate fractured in service:**

- **confirm that it is 316L (other phases/impurities?)**

XRD (Crystal structure) or EDC (element composition)

- **investigate the topography of the fracture surface (flaws, fatigue?)**

SEM (Surface structure and Morphology analysis)

- **look for signs of corrosion**

XPS (Chemical composition) SEM (Surface structure and Morphology analysis) + EDS (Elemental composition)

### **2. You want to confirm the presence of adsorbed protein and biological tissue on the surface of an implanted heart valve:**

- **confirm composition of adsorbed material (organic?)**

QCM (Measure a mass variation)

- **measure the thickness/amount of tissue material**

AFM (thickness)

- **confirm what protein it might consist of**

FTIR (Protein adsorption)

### **3. Doctors suspect a bacterial infection on a (removed) medical implant**

- **confirm the presence of bacteria on the surface**

Fluorescence Microscopy (Cell imaging), SEM (surface)

- **How were bacteria able to populate the surface? (surface chemistry)**

SEM (surface, bacteria) Fluorescence Microscopy (live cells and dynamic processes)