

SELF HEALING BONE CEMENT USING SQUID RING TEETH (SRT) PROTEIN

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The Problem

What is Bone Cement?

Bone cement (BC) is a critical material used in **orthopaedic surgeries**. It anchors artificial joints by filling the space between the prosthesis and the patient's bone.

Limitations of Bone Cement

Mechanical Fatigue

Cyclic loading in the body, leads to microcracks.

Brittle

Prone to fracture under repetitive stress.

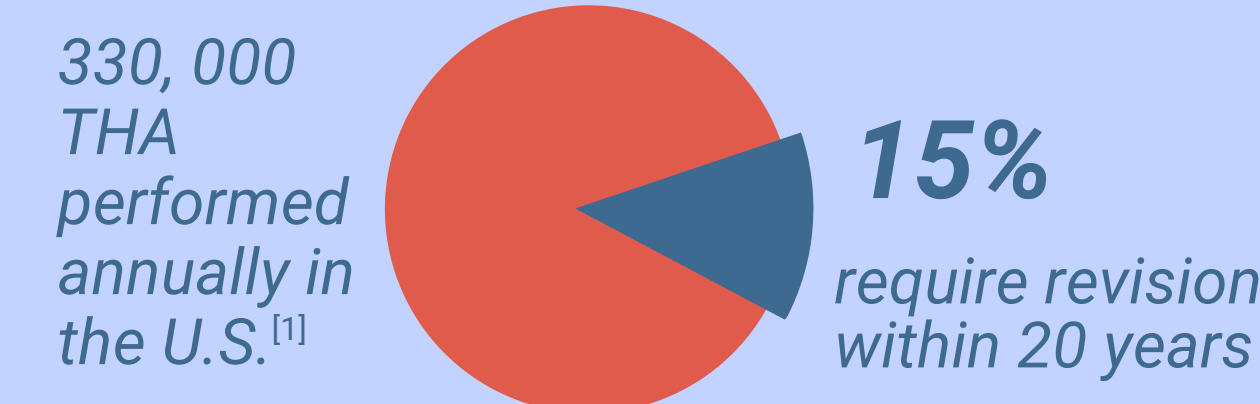
Infection Risk

Cracks in the cement can serve as pathways for bacterial colonisation.

Causing joint implants to fail, resulting in **revision surgery**...

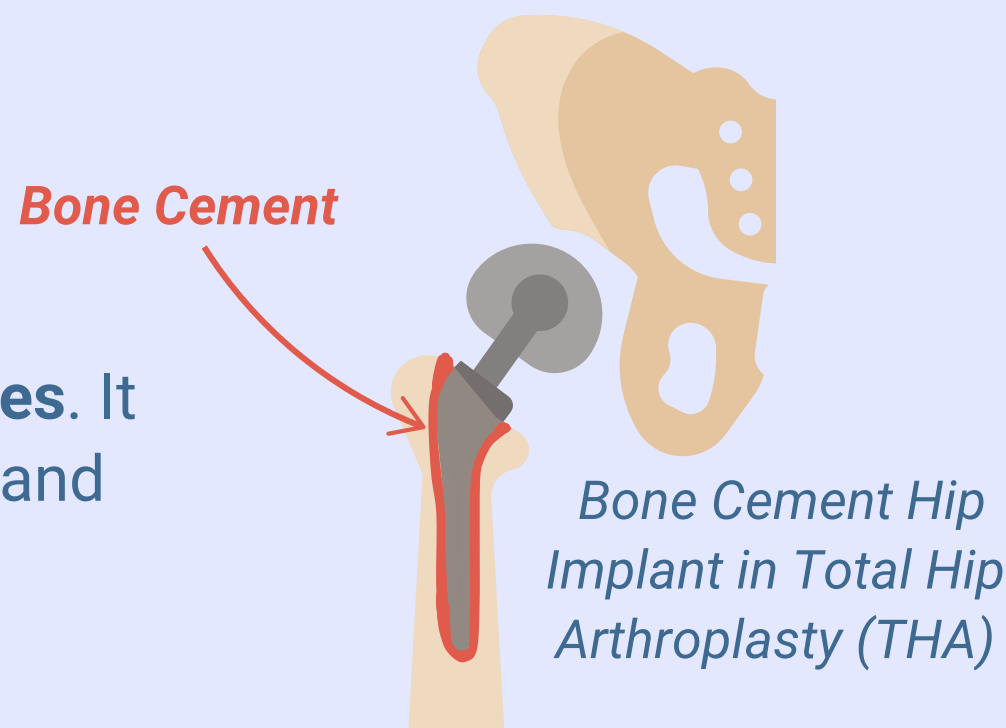
The Need for Innovation

With an aging population, demand for artificial joints continues to rise.



Making bone cement **self-healing** could offer a breakthrough solution:

- ➡ **Reduce** the risk of implant loosening and failure.
- ➡ **Improve** patient outcomes and minimise revision surgeries.



Protein Production

How to obtain SRT proteins in a sustainable way?

Use E.coli bacteria: ideal host as commercially available and has well-understood genetics.

The Method

Objective: Replicate the chemistry, structure and properties of SRT proteins.

1 Create repeating DNA sequence that includes an alternating amorphous and crystalline region.

2 Insert DNA sequence into a cloning vector using restriction enzymes and ligase.

3 Generate multiple copies of DNA template using Rolling Circle Amplification.

4 Cut long products into shorter fragments using restriction enzymes.

5 Separate the fragments by size using electrophoresis.

6 Select and ligate fragments into an expression vector.

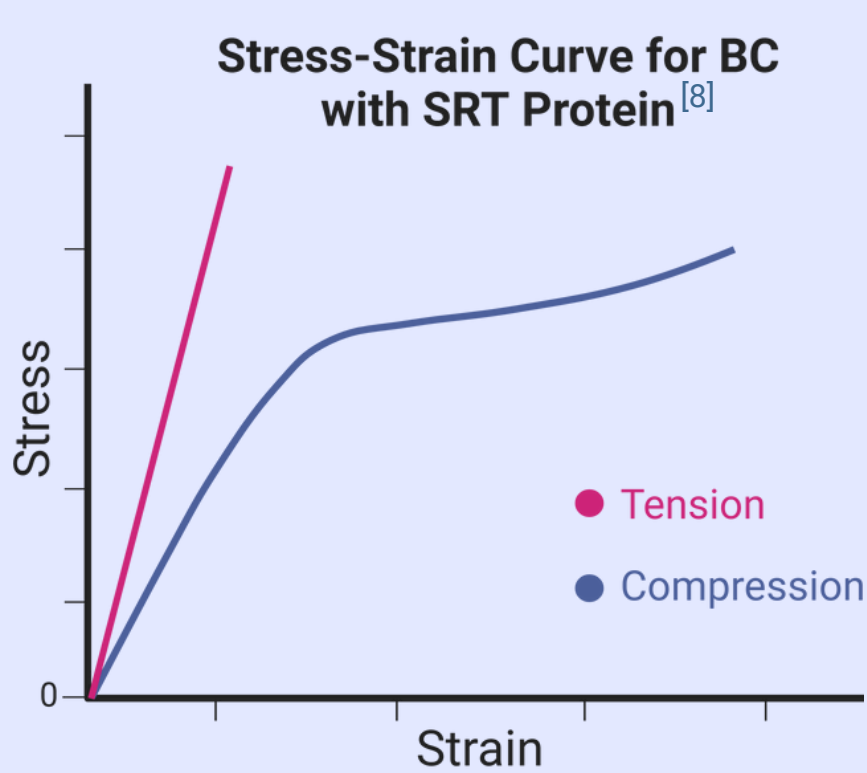
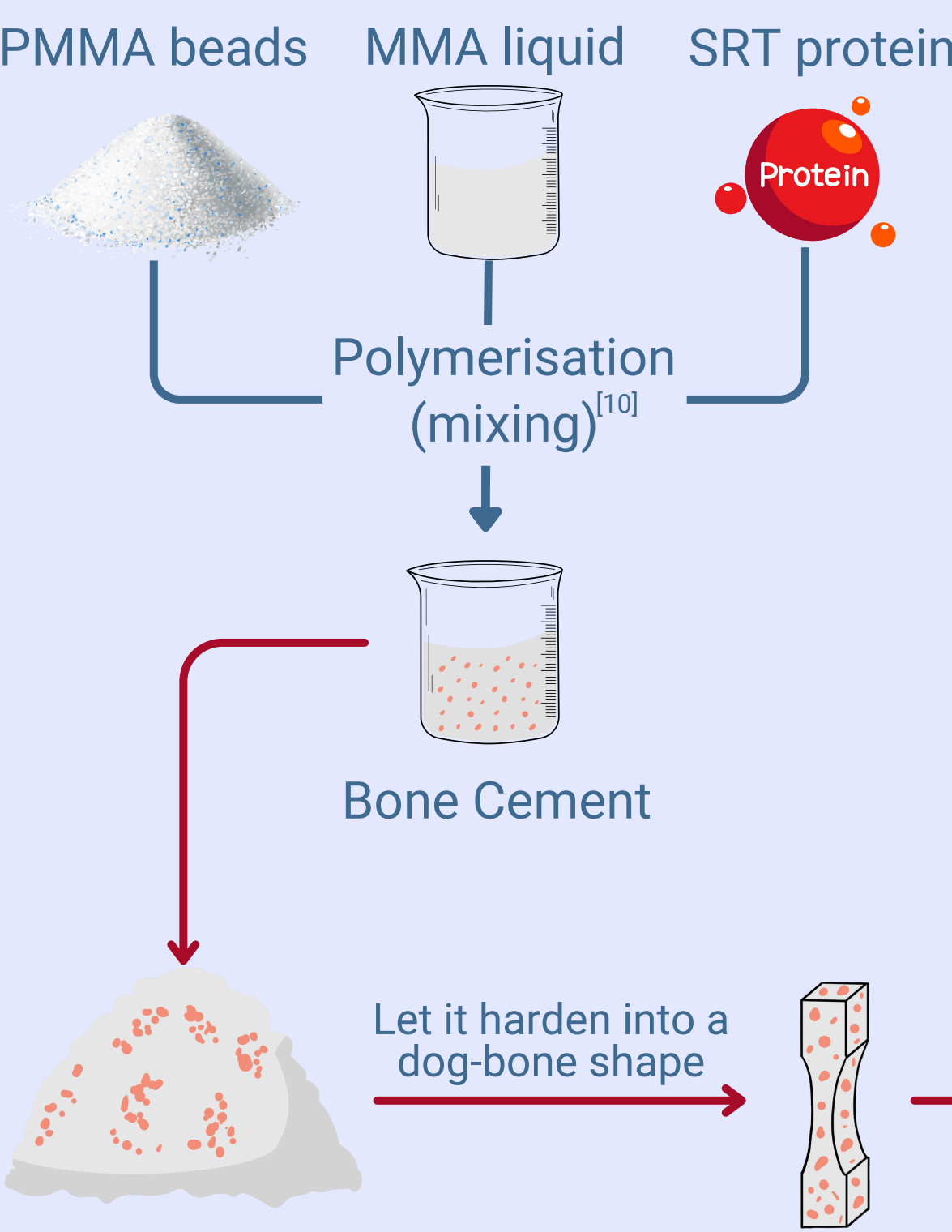
7 Introduce the expression vectors into E.coli for protein synthesis.

Implementation

Testing Methods

Two types of tests:

- **Compression** - Evaluate whether it is as strong and capable of withstanding compressive forces as standard BC.
- **Tensile** - Assess the tensile strength and determine if it can self-heal when subjected to pressure and water.
 - Cyclic loading: Conduct test repeatedly to measure the effectiveness of the self-healing properties.



How does it compare?

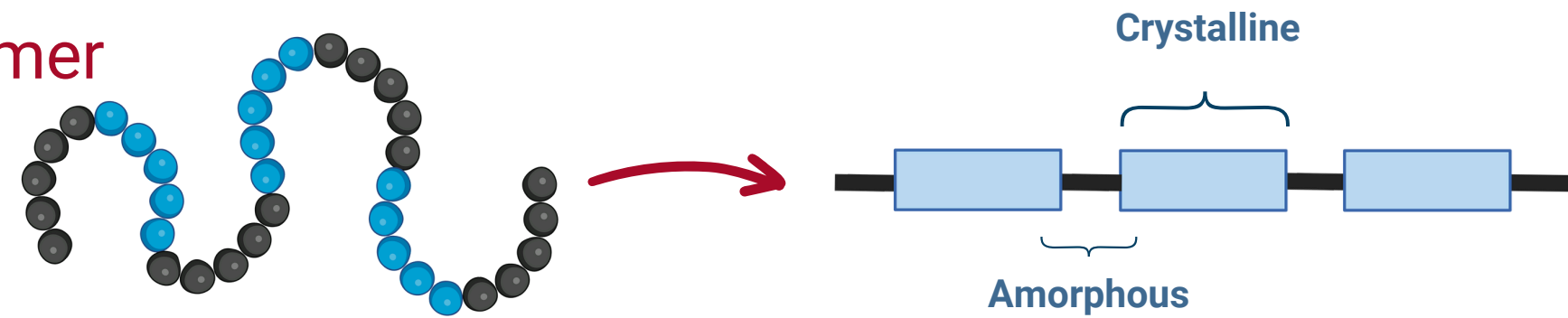
Property	Traditional Bone Cement	SRT Bone Cement
Self-Healing	Once damaged, it cannot repair itself.	Capable of repairing microcracks.
Biocompatibility	May cause inflammation, irritation, or allergic reactions.	Highly biocompatible with low cytotoxicity; biodegrades into natural amino acids. ^[3]
Toughness	Becomes more brittle over time, especially under stress	Improved toughness with isotropically oriented nanocrystals. ^[7]

The Technology: SRT Proteins

Protein Structure

Primary Structure - Heteropolymer

Alternating amorphous and crystalline regions



Secondary Structure - β -pleated sheets and random coils

β -Pleated Sheets provide **strength**

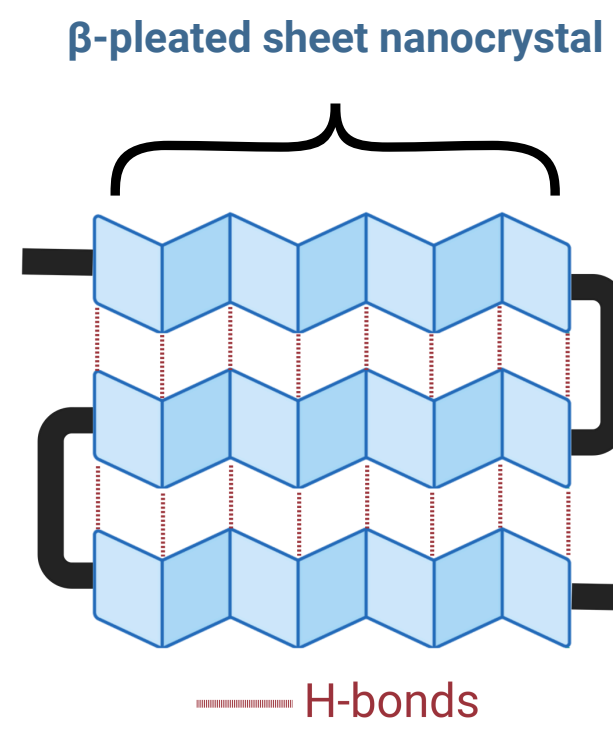
Formed through: H-bonds between crystalline regions drive self-assembly.

Aggregates to form nanocrystals with high tensile strength and rigidity.^[9]

Random Coils provide **flexibility**.

Formed through: Non-covalent interactions, between and within amorphous regions, drive randomly coiled structures.

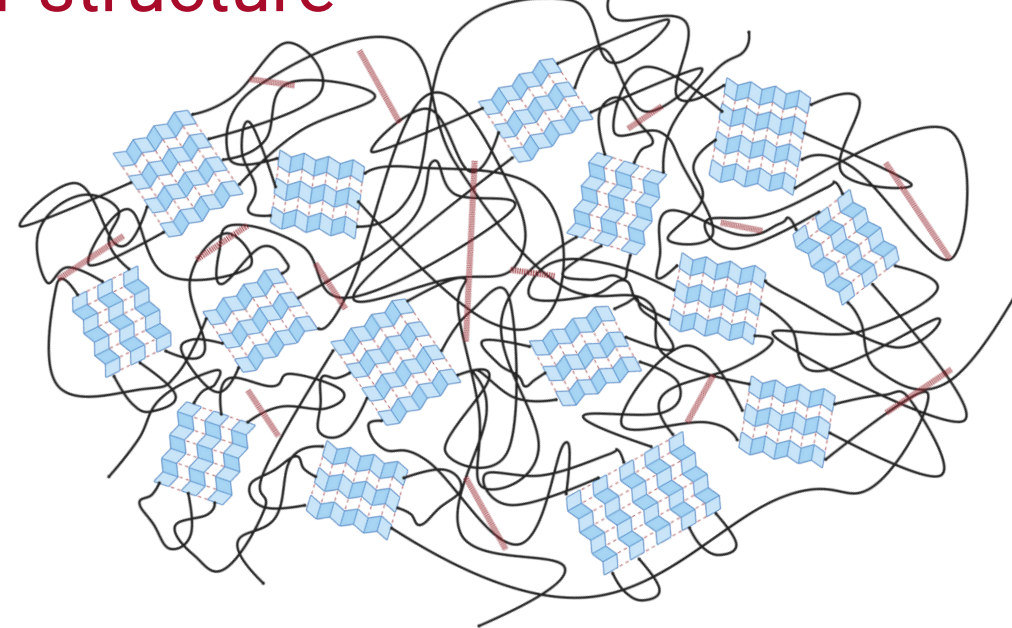
Enables flexibility and mobility, with an elastic modulus of 6-8 GPa in air.^[9]



Tertiary Structure - Self assembly into supramolecular structure

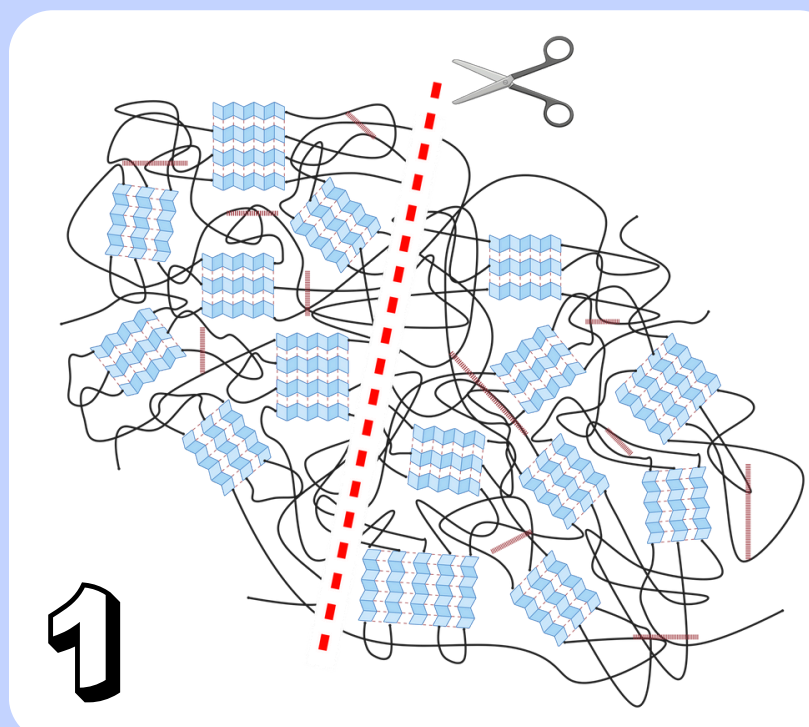
Formed through: Crystalline regions from different polypeptide chains form a single nanocrystal, acting as cross-links.^{[2][5][7]}

Since the structure is held together by non-covalent interactions the structure can self assemble and disassemble reversibly.



How are SRT Self-healing?

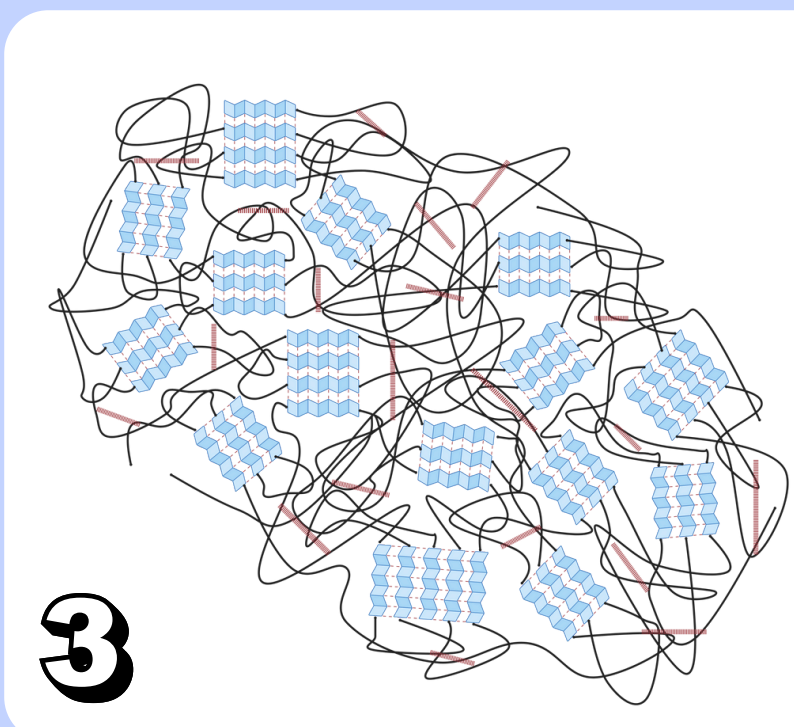
Damage leads to the disruption of H-bonds between the amorphous regions.



Addition of Water decreases the Glass Transition Temperature.^[9] Triggering the rigid to soft transition of amorphous regions.



Plasticisation facilitates **chain diffusion** and entanglement. This allows the realignment of the structure and the reformation of H-bonds.



Applications & Challenges

Challenges

Unknown lifespan of SRT proteins in BC - uncertainty in long-term performance.

Bacterial production of SRT proteins may pose scalability challenges for large-scale manufacturing.

Solutions

Perform **accelerated life span testing** to predict durability.

Optimise bacterial expression systems through genetic engineering to enhance yield and scalability.

Sustainability

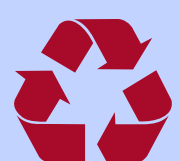
Existing bone cement is not recyclable, so it's more practical to focus on other sustainability aspects, rather than disposing/recycling SRT-based BC. Such as:



• **Animal-Friendly Production:** Utilising bacterial production of SRT proteins eliminates the need to harvest squid, thereby preserving marine biodiversity.



• **Eco-Friendly Manufacturing:** Implement the use of renewable carbon sources, such as plant-based sugars or industrial by-products, during the E. coli fermentation process.

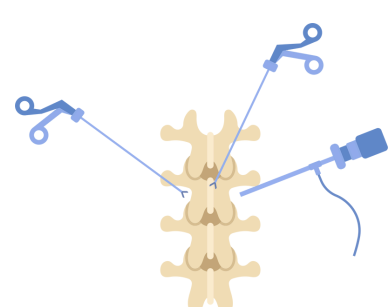


• **Biodegradability:** SRT proteins are biodegradable, which could contribute to making bone cement biodegradable in the future.

Other Applications

Spinal Fusion

Stabilisation of fractured vertebrae or fusion of bones to treat spinal disorders.



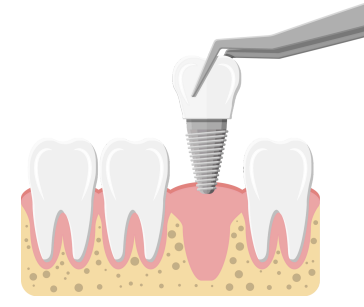
Cranioplasty

Reconstruction of cranial defects following trauma, surgery, or congenital anomalies



Dental Implants

Acting as durable, self-healing base for implants.



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