



Biotechnology & Biomanufacturing

Use of biological systems for the production, conversion, or functionalization of resources and (raw)materials

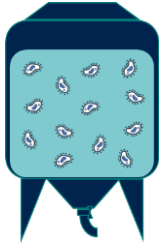
Discover – Develop – Connect

Gefördert durch:



aufgrund eines Beschlusses
des Deutschen Bundestages

Content



Fermentation – Mild reaction to access a wide product range



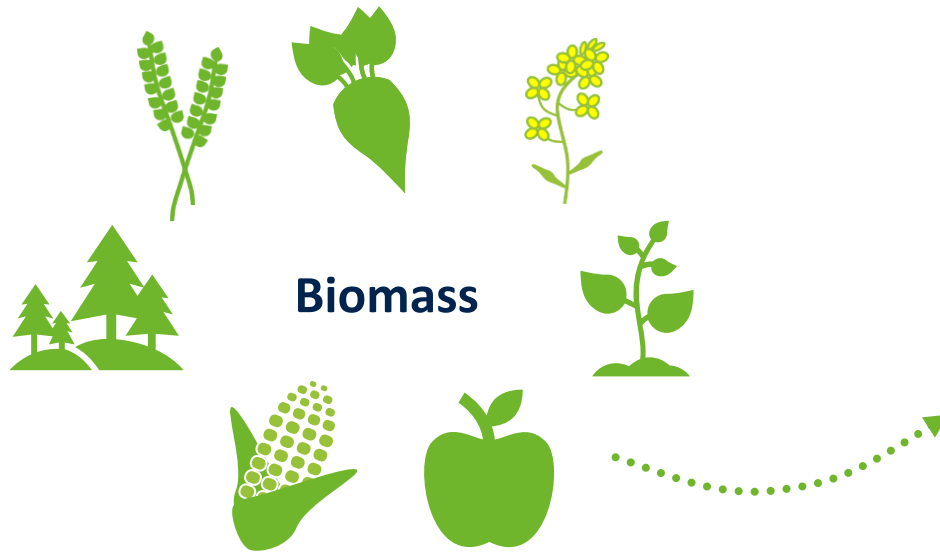
Protein Engineering – Tailored biomolecules



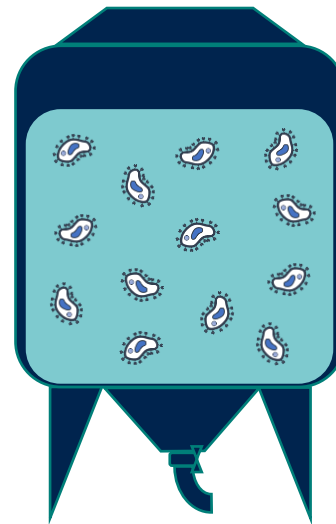
Bifunctional peptides – Biological adhesion promoters

Fermentation

Microbial or enzymatic conversion of organic substances



- Agricultural mass
- Organic waste (residuals, green waste, waste water etc.)



Fermenter (Bioreactor)

- pH value
- Temperature
- Microorganism/enzyme class
- Aerobic or anaerobic

Platform chemicals, building blocks



e.g.

- Ethanol
- Acidic acid
- Lactic acid
- Diacids
- Citric acid
- Butanol



Potential of fermentation



Environment & Safety

- Typically free of toxic solvents/reactants
- Less hazardous side products
- Products are often bio-degradable



Sustainable & renewable sources

- Biomass use
- Reduction of greenhouse gas emission



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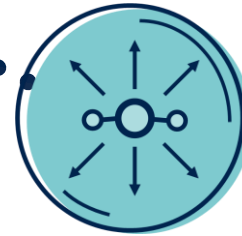
Sustainable & renewable sources

- Biomass use
- Reduction of greenhouse gas emission



Energy efficiency

- Low/ambient temperatures
- Mild conditions
- Ambient pressure



Product variety

- Broad range of platform chemicals accessible
- Access to highly functionalized molecules

Potential of fermentation



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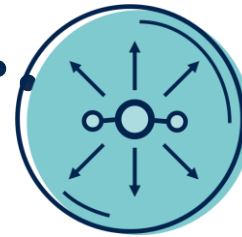
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Energy efficiency

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- Mild conditions
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Product variety

- Broad range of platform chemicals accessible
- Access to highly functionalized molecules



Optimization

- High specificity possible
- Engineering of proteins and microorganisms allows increased selectivity, conversion and yield



Economy

- Cost efficiency
- Scalability



Everyday examples



Pharmaceuticals

Large-scale production of many **medicines and active substances** (e.g., antibiotics, penicillin) with **assured quality**

Everyday examples



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Food

Crucial role in the **production of beverages and food** to generate **texture, flavor, and shelf life** (e.g., baked goods, cheese, yoghurt, beer and wine)

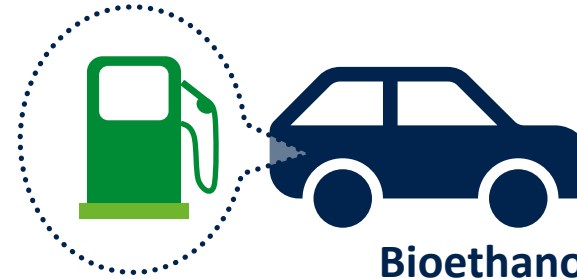


Everyday examples



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Bioethanol

Biorefinery and conversion of sugar from biomass to bioethanol



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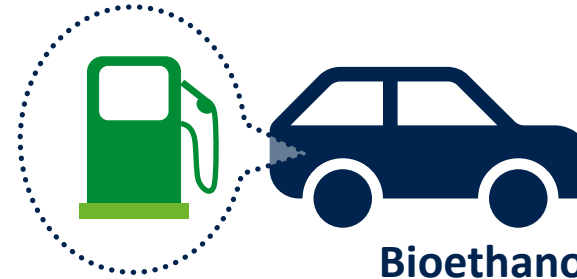


Everyday examples



Pharmaceuticals

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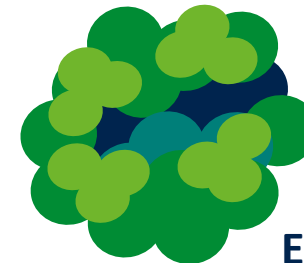
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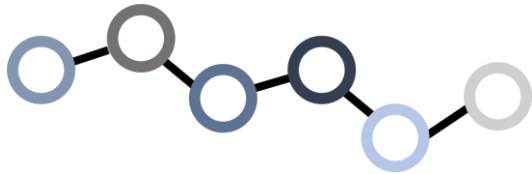
Enzymes

Production of **enzymes** for **industrial applications**, such as proteases, lipases, and cellulases for **detergents**

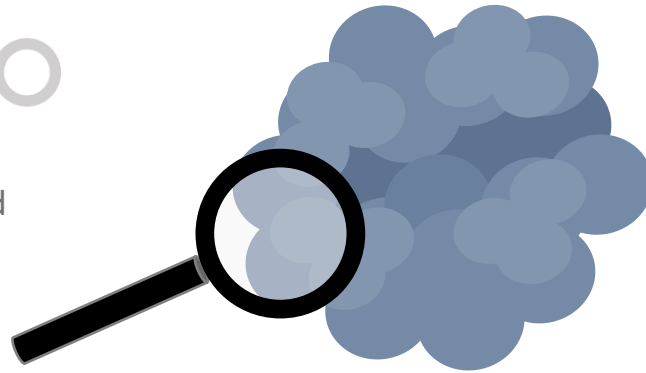


Protein Engineering

Construction, optimization and production of proteins (including enzymes)

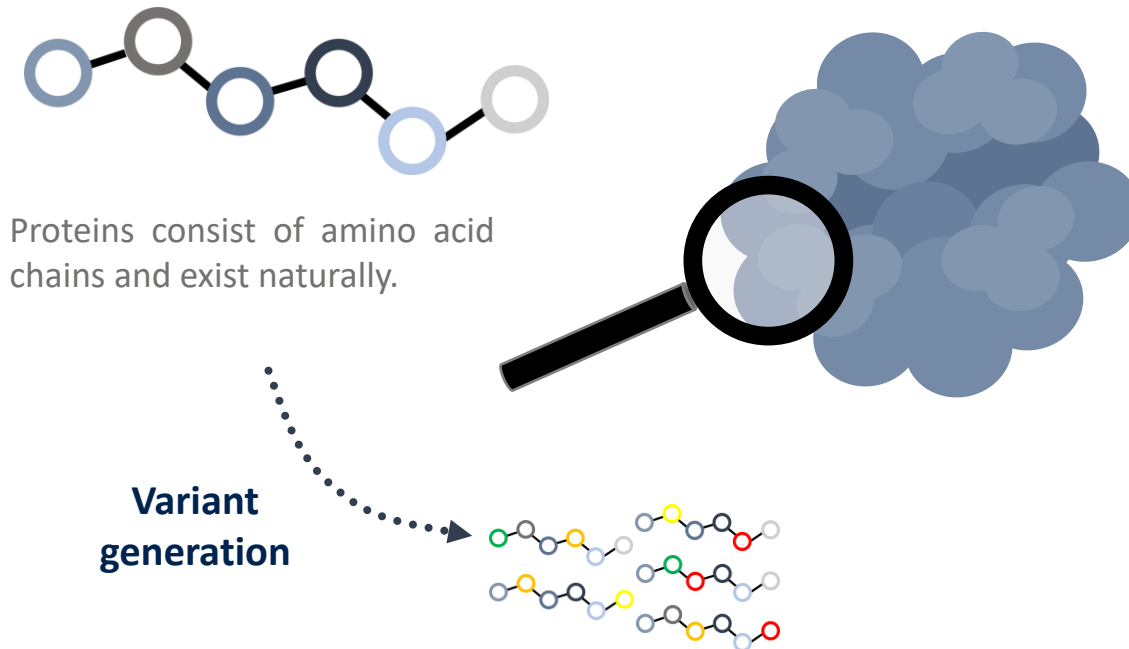


Proteins consist of amino acid chains and exist naturally.



Protein Engineering

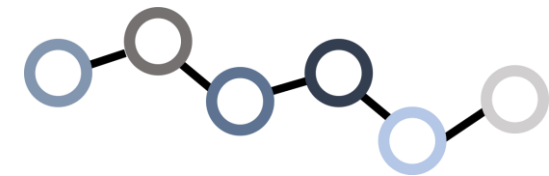
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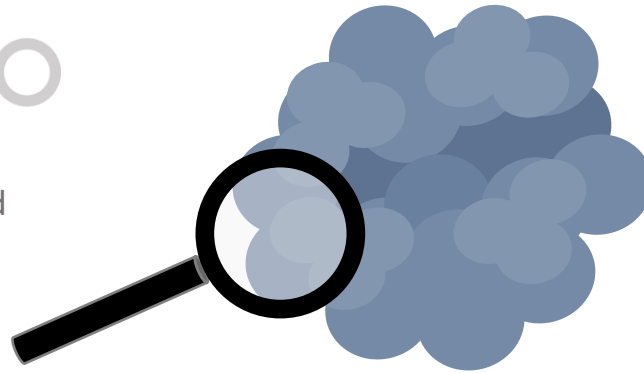
Using biotechnological methodologies, amino acids can be exchanged by others in a side-directed or random fashion.

Protein Engineering

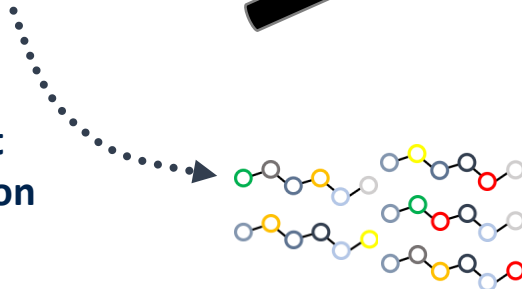
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Variant generation



Using biotechnological methodologies, amino acids can be exchanged by others in a side-directed or random fashion.

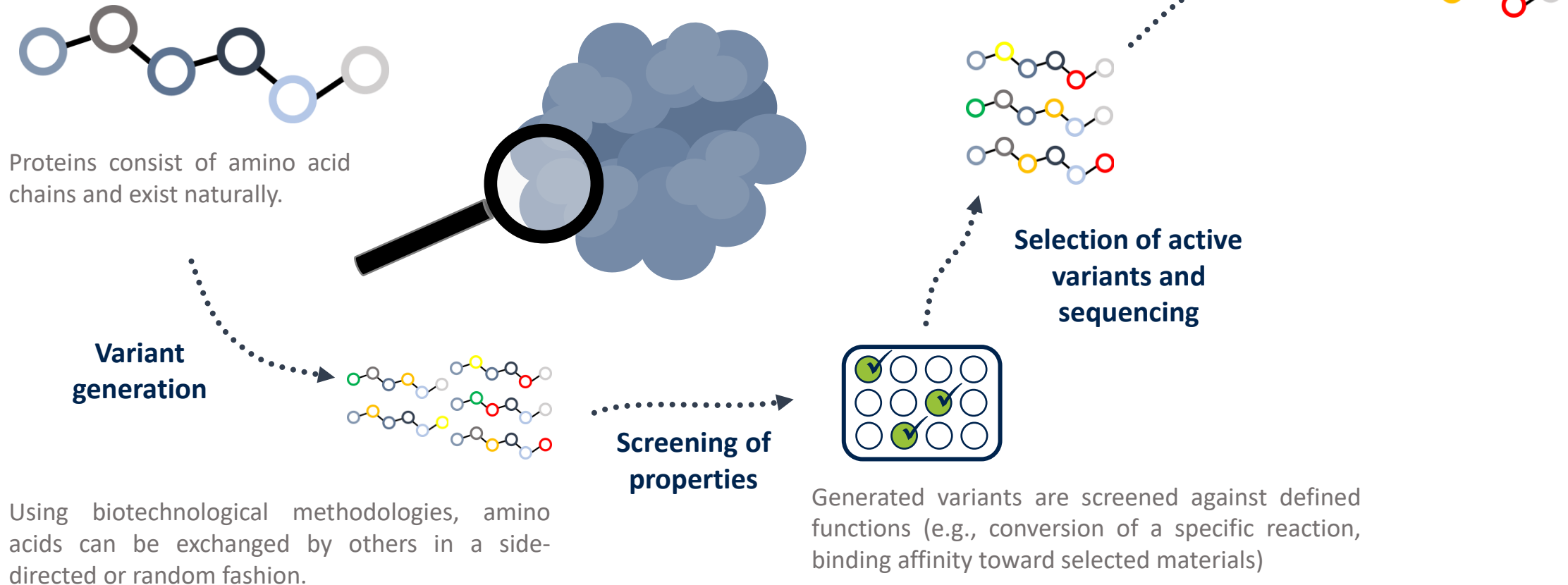
Screening of properties



Generated variants are screened against defined functions (e.g., conversion of a specific reaction, binding affinity toward selected materials)

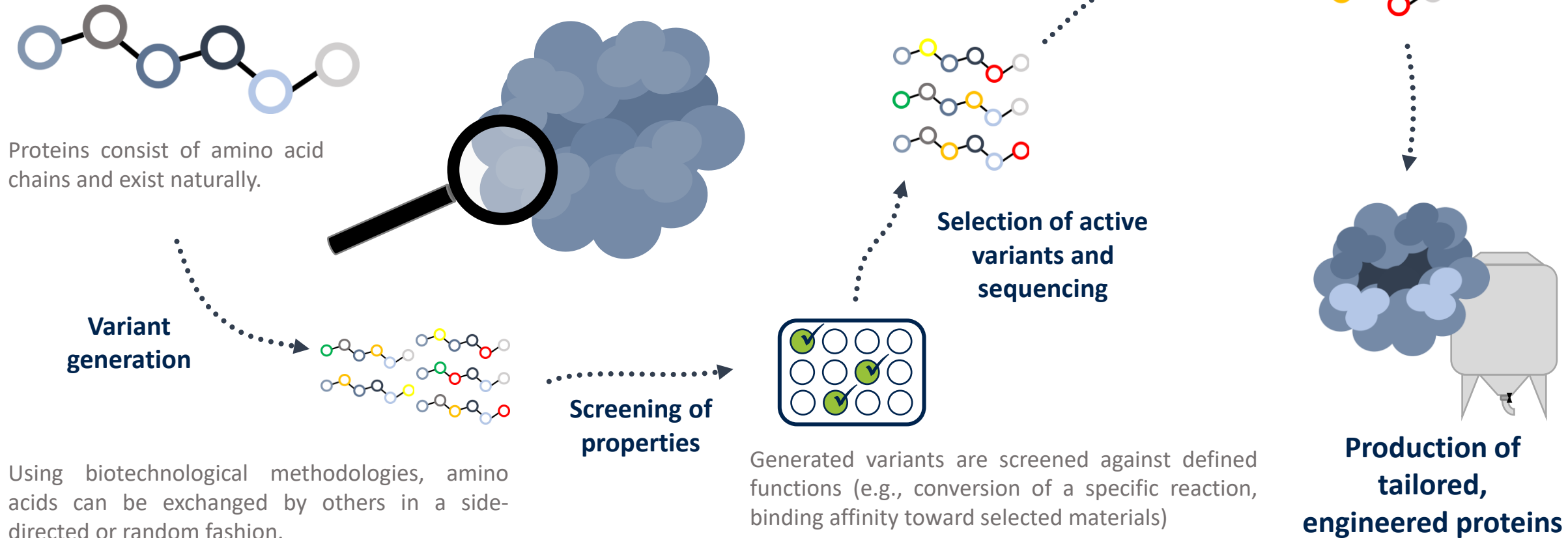
Protein Engineering

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Protein Engineering

Construction, optimization and production of proteins (including enzymes)





Potential for optimization (**stability**)

Tailored function through protein engineering



Thermal stability

The natural **thermal stability** of proteins varies greatly and depends on the natural origin of the protein. It can be **improved compared to the native form** by modifying its structure, which leads to more intramolecular bonds. Typically, a **balance between stability and activity** must be taken into account.

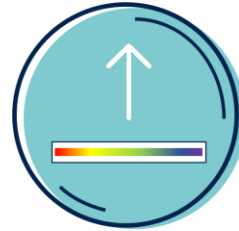
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**pH
stability**

Natural processes such as biocatalysis usually work best within **defined pH ranges**. These can be modified to allow for **improved tolerance to fluctuations, broader pH ranges or specific processing conditions**.

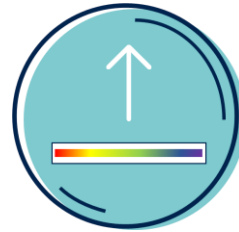
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**Tolerance against
ionic liquids**

The use of ionic liquids (ILs) is playing an increasingly important role in the **processing of biological materials** (e.g., the extraction of cellulose fibers). They are also used as substitutes for conventional volatile organic solvents and compounds. **Tolerance to ionic liquids** can therefore be **particularly important in the industrial use of enzymes**.



Potential for optimization (**stability**)

Tailored function through protein engineering



Activity

Proteins can be **naturally catalytically active** and carry out a specific chemical reaction (e.g., hydrolysis). These types of **reactions can be relevant for industrial processes** and can be used for example for the targeted degradation of substances (e.g., degradation of polymers/plastics). The **conversion, yield and reaction rate can be significantly improved**.



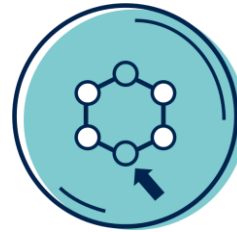
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Regio-selectivity

The regioselectivity of a chemical synthesis plays a **decisive role in the resulting chemical and physical properties**, not only for active (bio)molecules. Enzymes enable **very good regioselectivity at mild/ambient conditions and employing less synthesis steps** than conventional organic chemical synthesis.

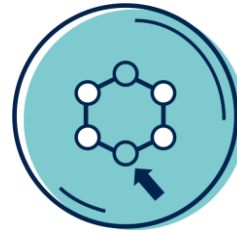
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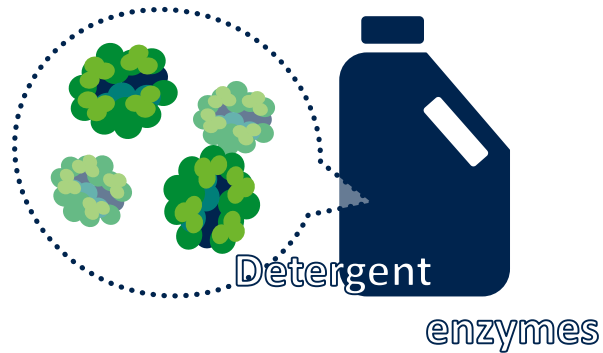
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Material-specificity

Similar to the activity of catalytically active proteins, functionalization on and in materials using proteins can be **tailored by improved specificity toward particular materials**. This can enable, for example, **selective binding/labeling or functionalization in material mixtures**. Both **natural and man-made materials** are applicable.

Everyday examples



Substrate specificity enables high efficiency with small dosages, thereby protecting textiles to preserve durability.



Stability in the presence of surfactants and other components, as well as at different pH values.

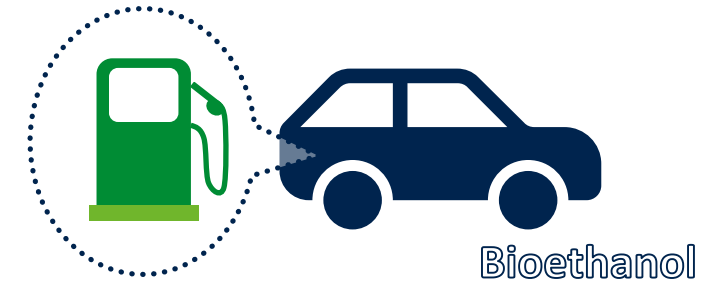
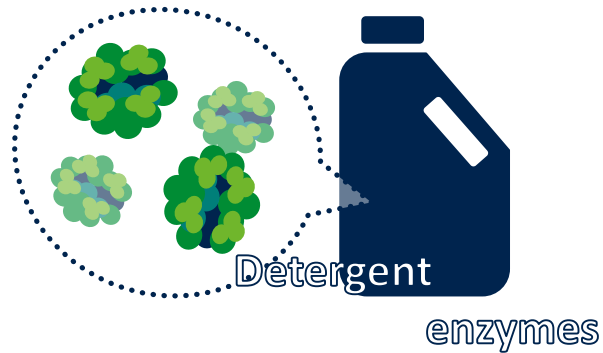


High **efficiency** at low temperatures enables significant energy savings.



Biodegradability of enzymes makes them more environmentally friendly than synthetic chemicals.

Everyday examples



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Stability in the presence of surfactants and other components, as well as at different pH values.



High **efficiency** at low temperatures enables significant energy savings.



Biodegradability of enzymes makes them more environmentally friendly than synthetic chemicals.



Substrate specificity ensures faster and more efficient conversion of complex carbohydrates (e.g., from grains) into sugars.



Stability at elevated temperatures (pre-treatment and hydrolysis of biomass) as well as at different pH values or in presence of larger amounts of ethanol and inhibitors.



More flexible adaptability of processes while reducing toxic chemicals, especially in second-generation biomass.



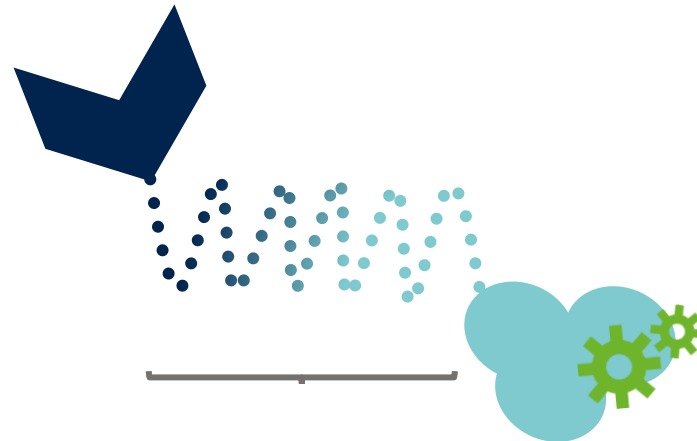
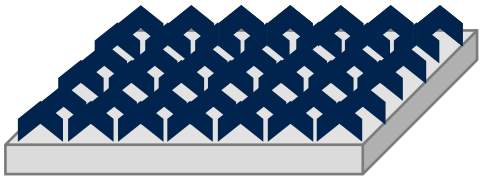
Renewability of the raw materials makes the product more environmentally sustainable.

Bifunctional peptides

Tailored biological adhesion promoters^[1]

Peptide A | ADHESION

- Small domain (ca. 10-50 amino acids)
- Tailor-made for **strong, specific binding** to **Material 1**



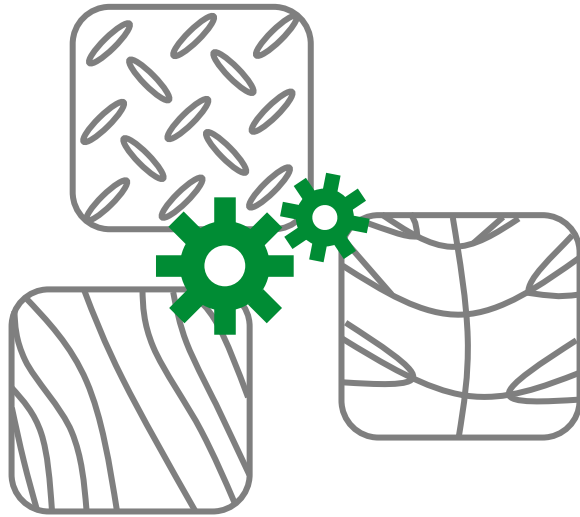
Spacer

- small helix
- separates peptide A and peptide B to prevent undesired interactions

Peptide B | FUNCTION

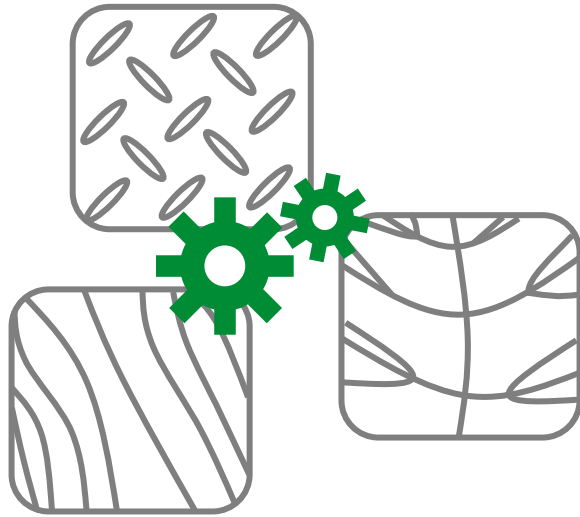
- Small domain (ca. 10-50 amino acids)
- Tailor-made for:
Binding of molecules (e.g. flame retardant)
Function: biological (e.g. anti-microbial), chemical (e.g. anti-corrosive), physical (e.g. hydrophobic, hydrophilic)
strong, specific binding to **Material 2**

Functionalization ON material



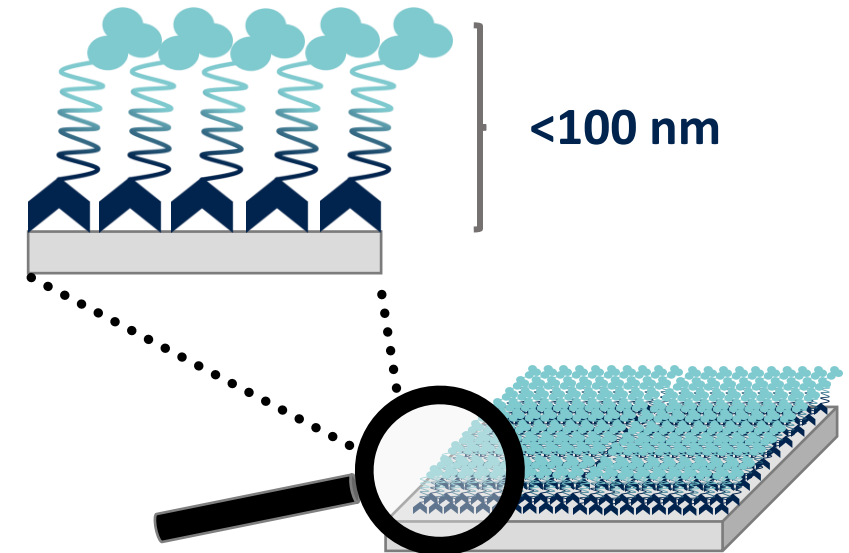
- **Functional coatings** (e.g. hydrophobic, hydrophilic, flame-retardancy or other functions)
- **Switchable adhesives** (e.g. for selective disassembly of multi-component material)
- **Biological cross-linker** (e.g. for fiber extension)

Functionalization ON material

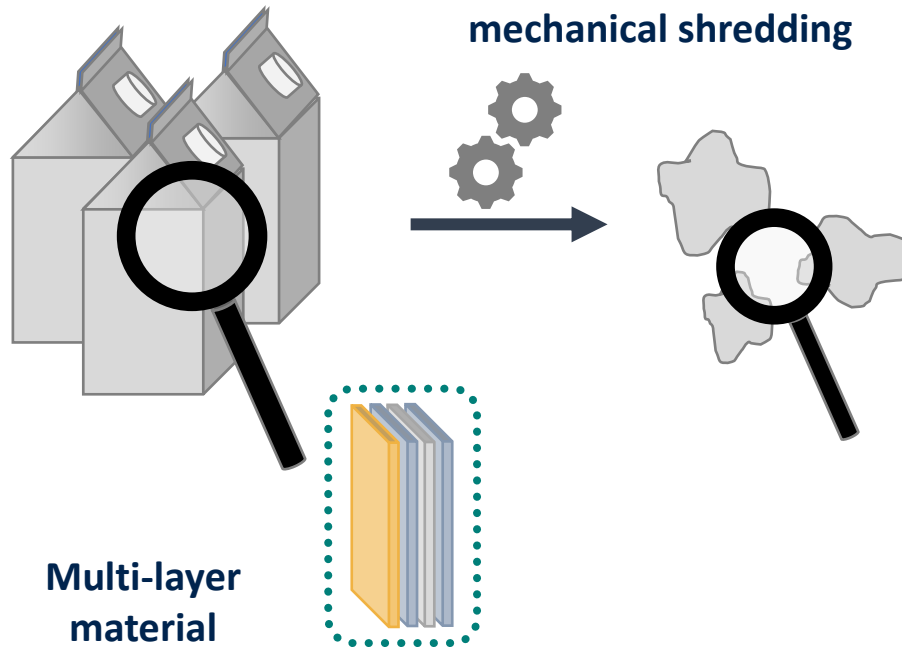


- ✓ material-specific binding (e.g., to PLA, PE, PA)
- ✓ high **mass/area efficiency** (1 g peptide for ca. 250 m²)
- ✓ high **coating density**
- ✓ **scaleable** & cost efficient production
- ✓ compatible with **standard coating methods** (e.g. dip-coating, spray coating)

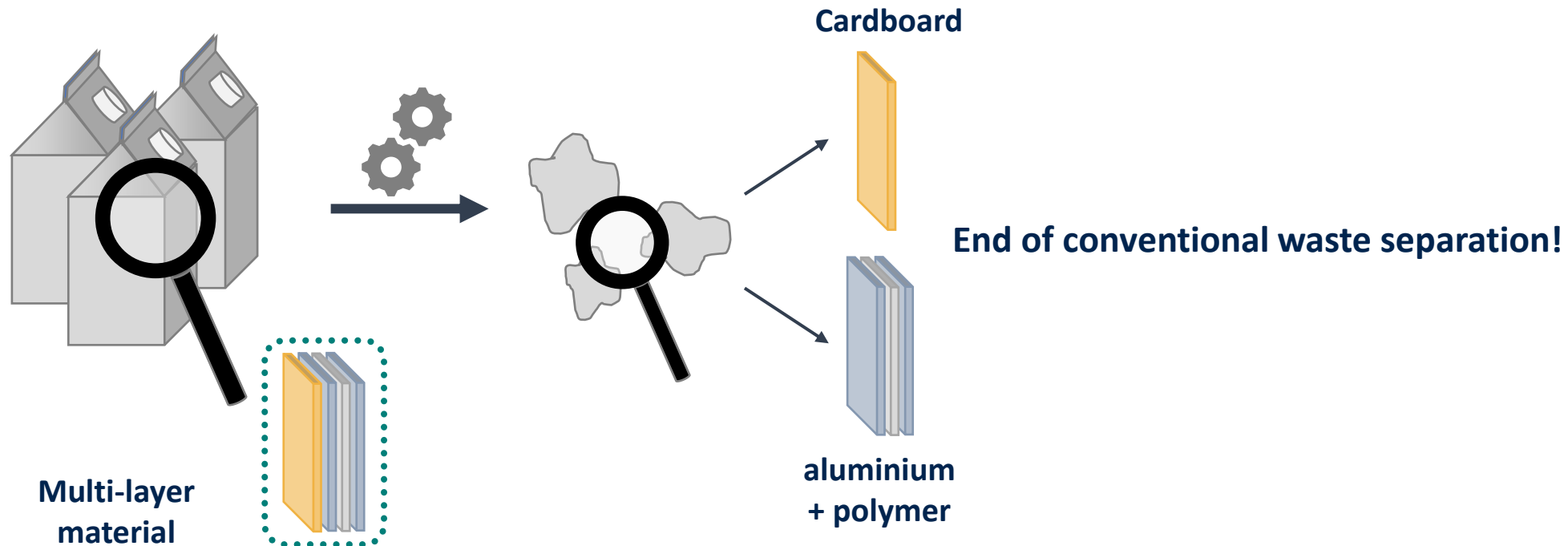
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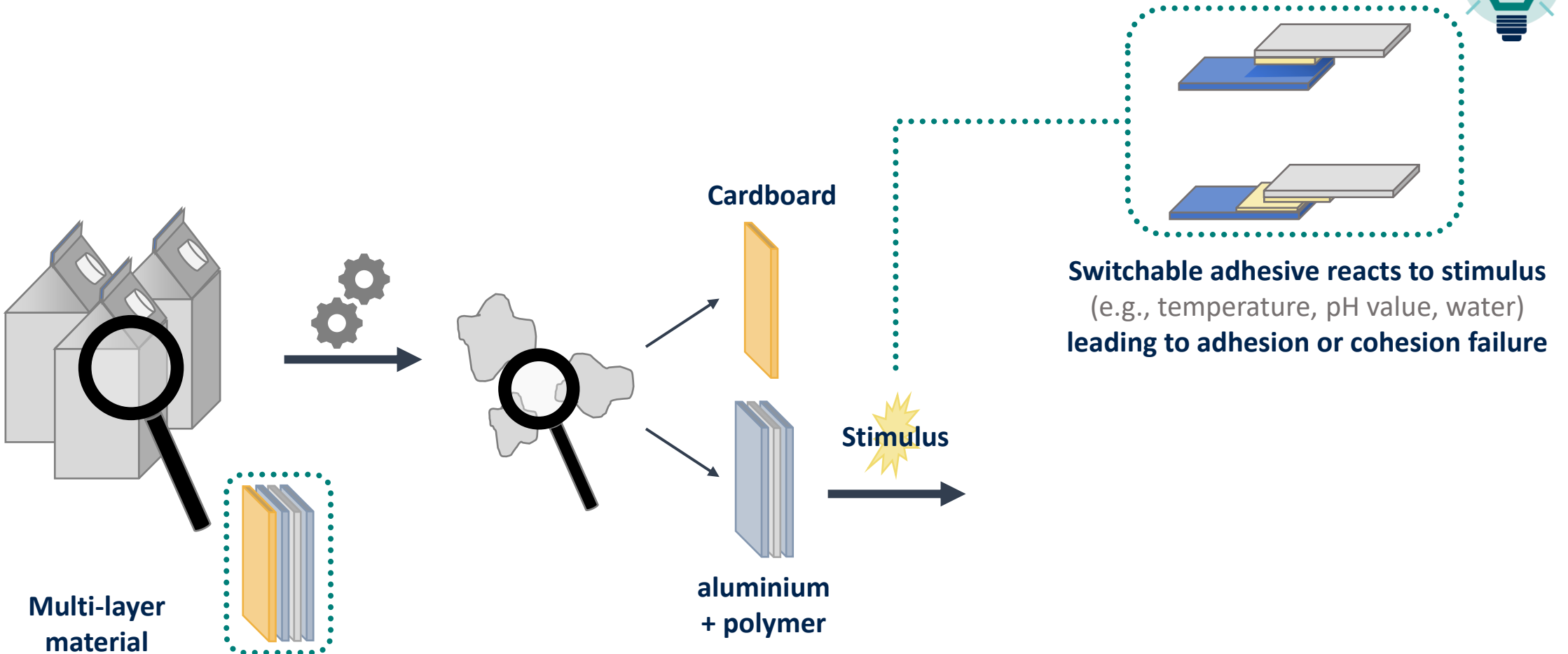
Use Case: Switchable adhesives in multi-layer materials



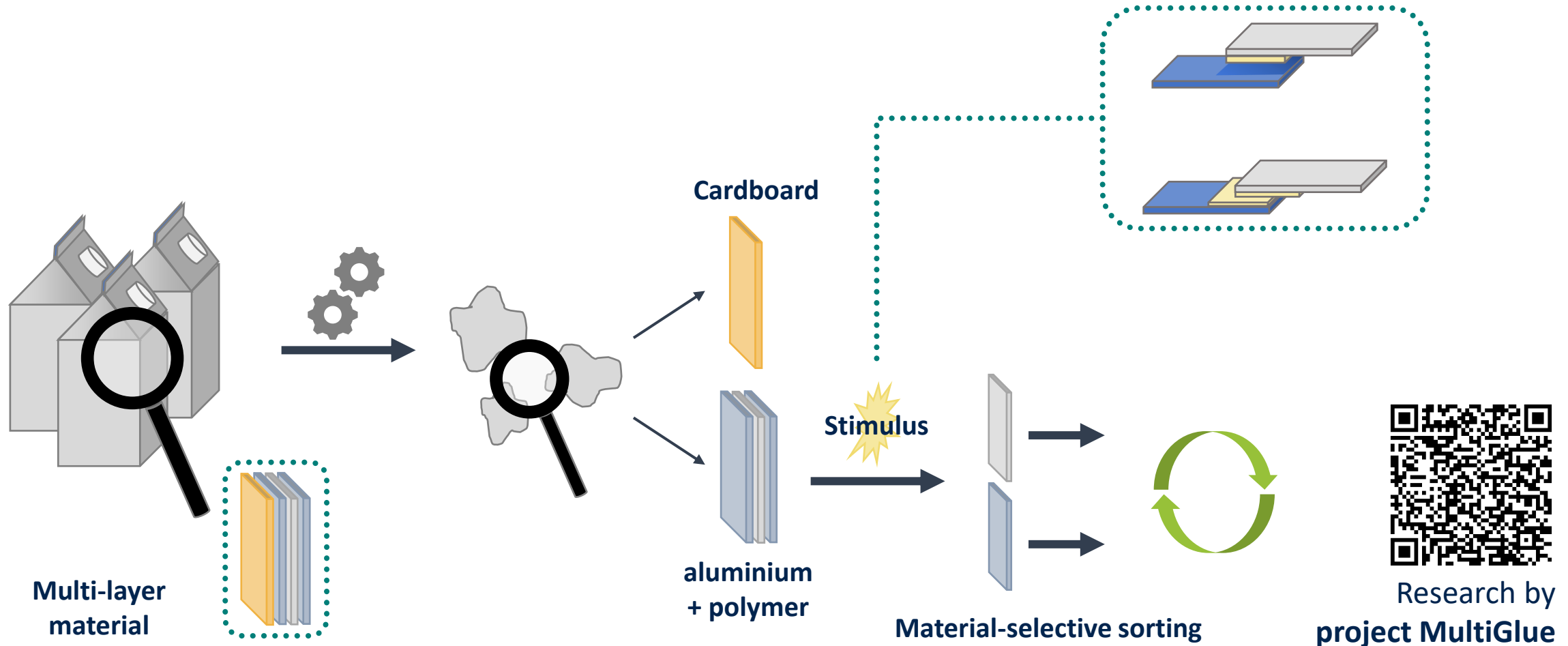
Use Case: Switchable adhesives in multi-layer materials



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Use Case: Switchable adhesives in multi-layer materials



Functionalization IN material



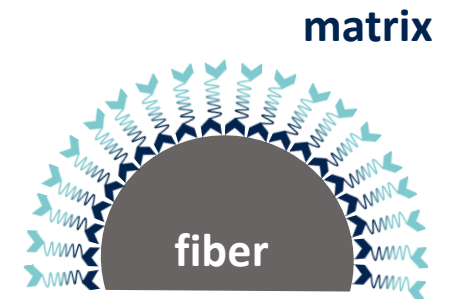
- **(Programmed) degradation** (e.g., of plastics with high littering risk)
- **Compatibility in composites** (e.g., in fiber-reinforced plastics)
- **Material-specific de-polymerization** (e.g., in mixed plastics)

Functionalization IN material



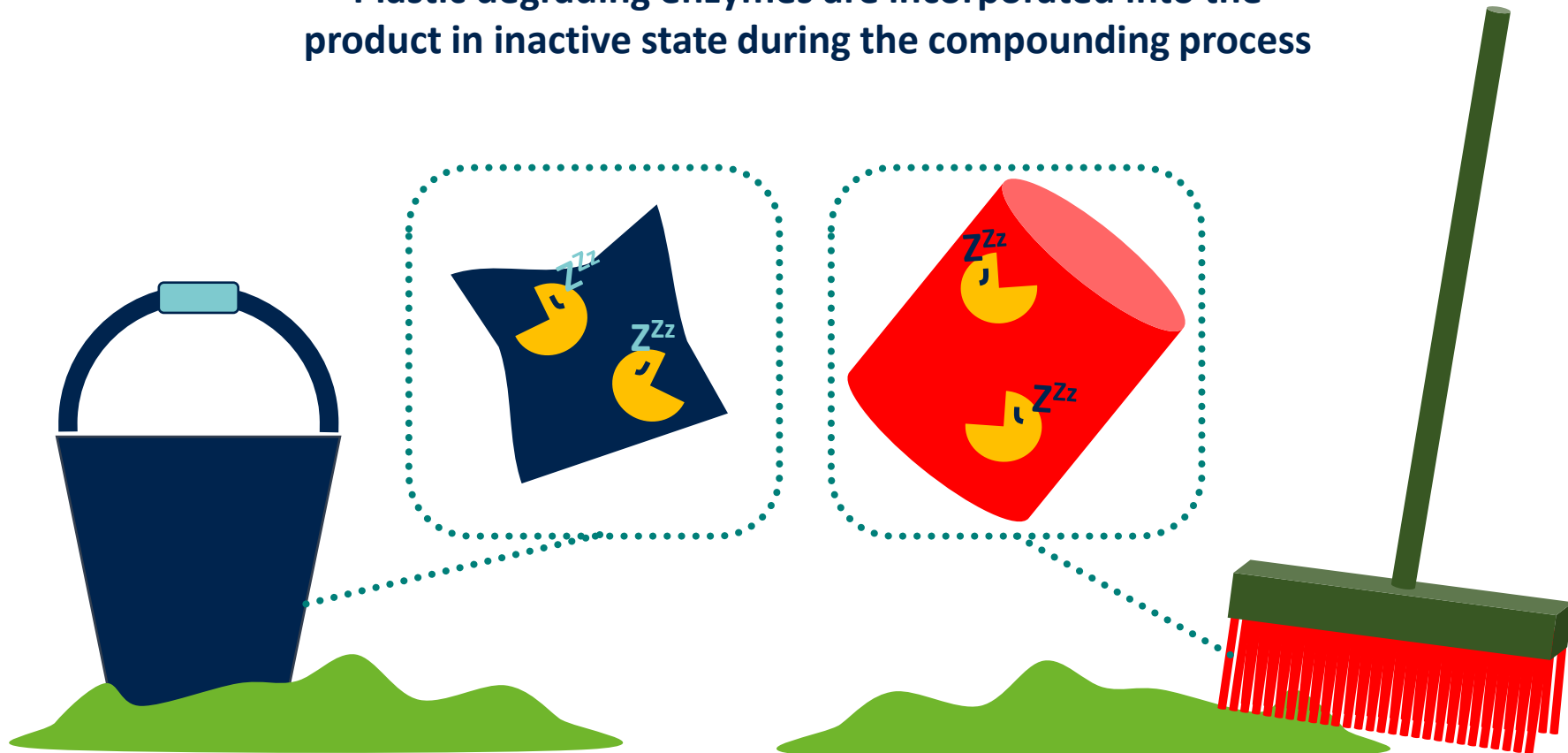
- **(Programmed) degradation** (e.g., of plastics with high littering risk)
- **Compatibility in composites** (e.g., in fiber-reinforced plastics)
- **Material-specific de-polymerization** (e.g., in mixed plastics)

- ✓ material-specific binding,
e.g., to fiber (protein A) and polymer matrix (protein B)
- ✓ high **mass/area efficiency** (1 g peptid can coat ca. 250 m²)
- ✓ **programmable properties**,
e.g., activation of degrading enzymes at defined environmental conditions
- Adjustment of processes might be necessary to ensure **biocompatible production technologies**

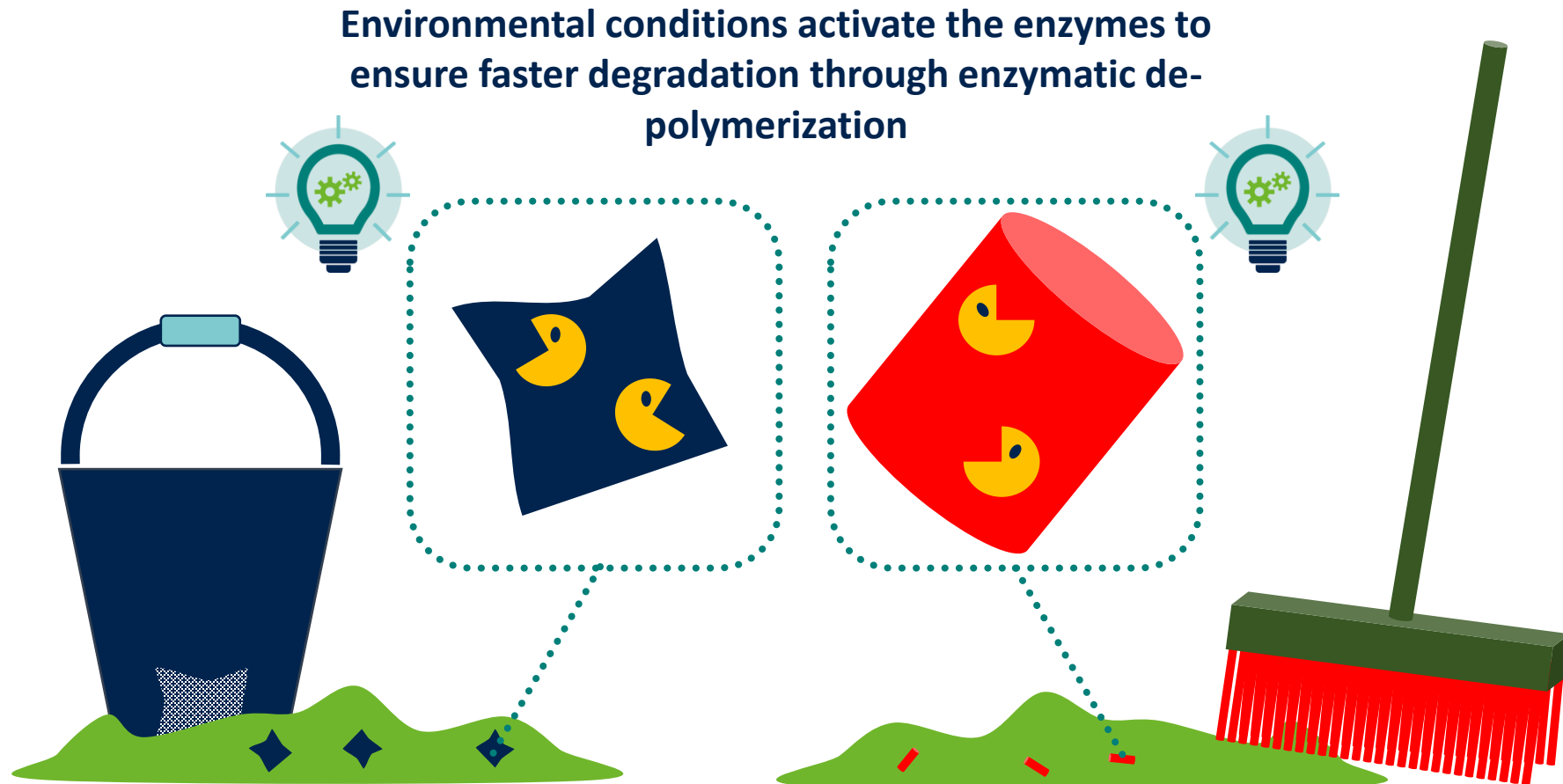


Use Case: Programmed plastic degradation

Plastic degrading enzymes are incorporated into the product in inactive state during the compounding process

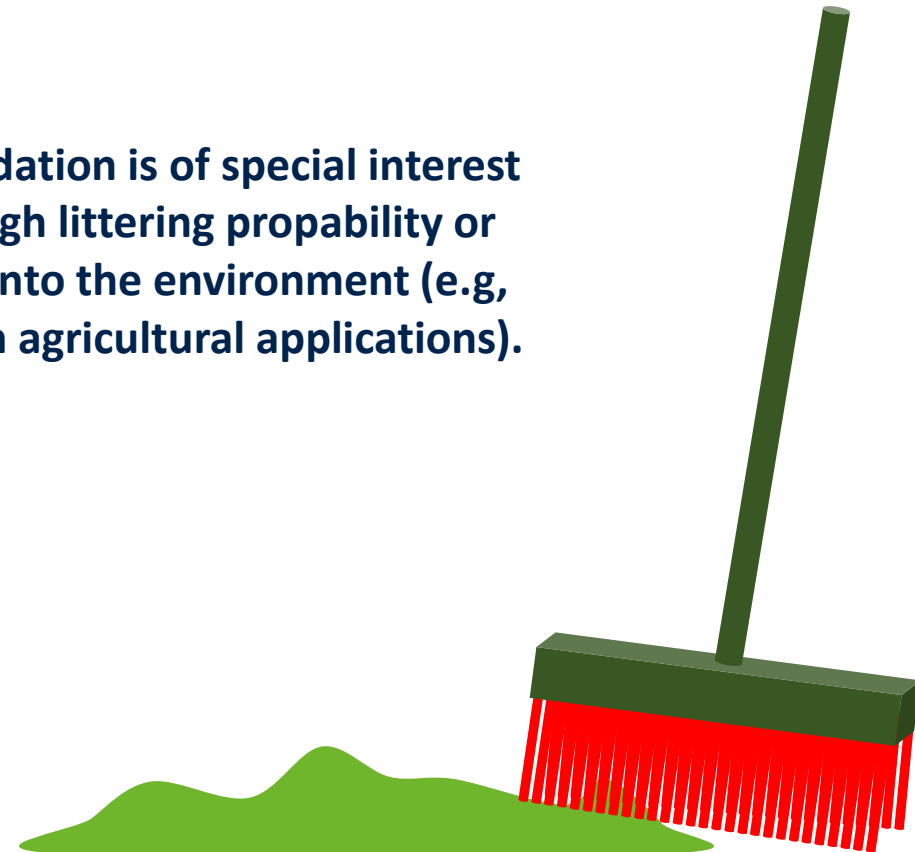
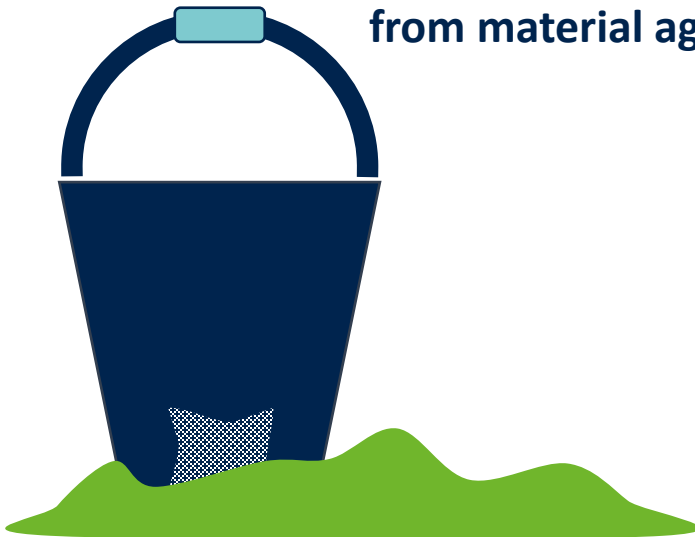


Use Case: Programmed plastic degradation

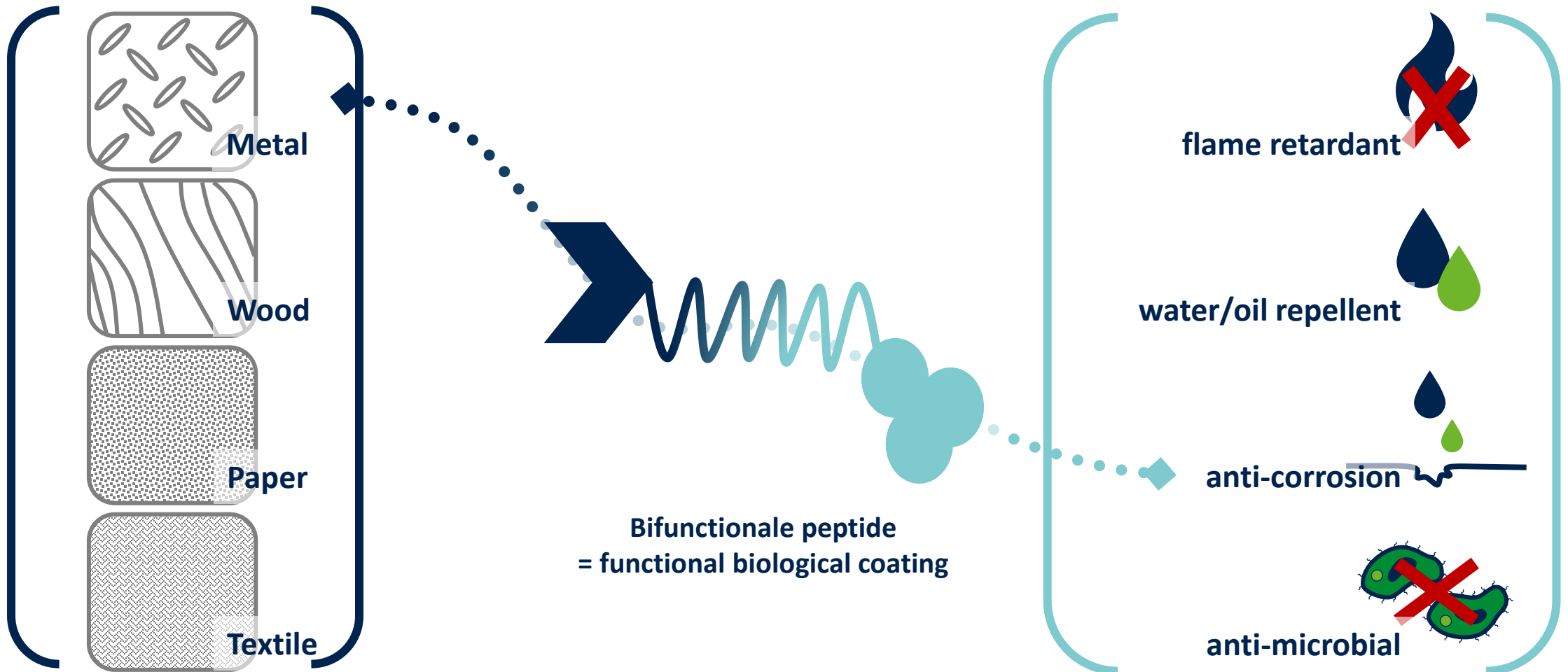


Use Case: Programmed plastic degradation

This programmed degradation is of special interest for applications with high littering propability or exposure of fragments into the environment (e.g, from material aging or in agricultural applications).



Use Case: “Toolbox” for surface functionalization



“Protein Engineering allows us to tailor enzymes to create new materials, drive green chemistry, and make biopolymers that are biodegradable and environmentally friendly.”

Frances H. Arnold, Nobel laureate 2018
“for the directed evolution of enzymes”



Discover – Develop – Connect



Your contact and partner regarding
Biotechnology & Biomanufacturing

Dr. Thomke Berge
Institute of Biotechnology,
RWTH Aachen University

 transbib@rwth-aachen.de

Self-check

True or false? Evaluate the following statements based on the content from the recent chapter (solutions are presented on the following slide).

(1) Proteins can bind in specific fashion to various materials.

(2) Only performance properties found in native proteins can be used or applied.

(3) Apart from biological functions, chemical or physical functionalities can be used in peptide coatings.

(4) Proteins can only be used in surface functionalization and applications.





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Correct! The material selection is not limited to natural (e.g., leaves) or biobased (e.g., paper) materials. Man-made materials such as plastics or metal alloys might as well be coated with functional peptides.





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False! With the help of *protein engineering* i.e., the customized functionalization of proteins, specific properties such as stability towards temperature, pH value, or the enzymatic activity can be tailored for desired applications.



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Correct! By using bifunctional peptides, both the binding to bulk material and the function (e.g., hydrophilic, hydrophobic, anti-microbial) of the coating can be customized.





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False! Functionalization with proteins (including enzymes) can also be used for functionalization IN materials. Examples include adhesion promoters in composite materials or embedded enzymes for improved degradation.



Note

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