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Harnessing Plant Trypsin Inhibitors for Crop Security and Human Health

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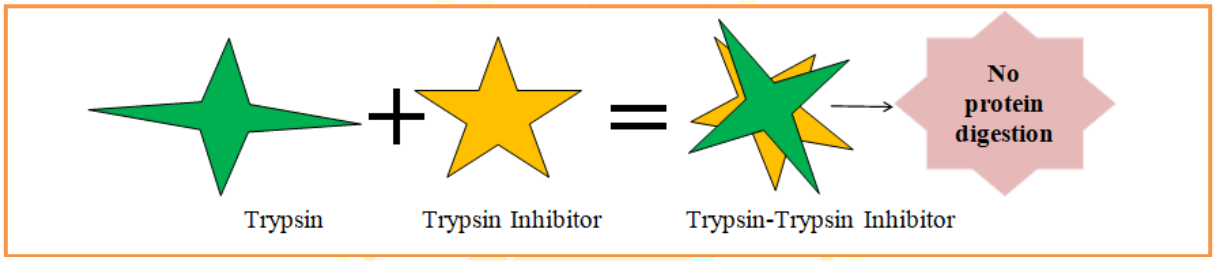
Trypsin inhibitors are anti-nutritional proteins found widely in plants—including legumes (soybeans, beans, peas), cereals (wheat, barley), potatoes, and cucurbits. A trypsin inhibitor is a type of protein that binds to trypsin—a crucial digestive enzyme in our small intestine—and stops it from breaking down proteins. By blocking this enzyme, these inhibitors prevent trypsin from doing its regular job, which has major implications ranging from nutrition issues to breakthrough in managing agriculture challenges and medical treatments.



Basic mechanism of action

Introduction

Trypsin inhibitors are widespread in nature and are found abundantly in various plants. Plants use them as a defense mechanism against insects and pests. Plant-derived trypsin inhibitors (TIs), predominantly categorized into the Kunitz and Bowman-Birk families, act as vital chemical defense mechanisms evolved by plants to deter herbivory (Mehmood et al., 2024; Sultana et al., 2023). Kunitz Type: This is a relatively large, globular protein. It typically weighs around 20 to 22 kDa (kiloDaltons) and consists of a single polypeptide chain of roughly 170–200 amino acids.Contains only 2 disulfide bonds (cross-linkages between cysteine amino acids). Because it has fewer of these rigid internal bridges, its large structure relies more on general protein folding.It has one active binding site. It binds specifically to trypsin in a 1:1 ratio. Once it blocks a trypsin enzyme molecule, the inhibitor is fully occupied.Bowman-Birk Type: This is a tiny, highly compact polypeptide. It is a fraction of the size, weighing only 7 to 8 kDa with just 60–90 amino acids. Contains a massive concentration of 7 disulfide bonds. For a protein so small, this creates an incredibly dense, rigid, cage-like framework that locks the molecule into shape. It features two independent active binding loops located on opposite sides of its tiny structure. Typically, one loop binds and inhibits trypsin, while the second loop targets a completely different digestive enzyme, chymotrypsin. This allows a single Bowman-Birk molecule to deactivate two enzyme molecules simultaneously.

Their deployment in agriculture has moved from exploiting natural host-plant resistance to active genetic engineering, positioning them as a major frontier in eco-friendly

pest management (Mehmood et al., 2024). Beside this trypsin inhibitors are important target source for the management of various health problems

Applications

Transgenic Crop Engineering

Modern biotechnology is relocating plant defenses from seeds to foliage. While wild plants naturally express TIs in their seeds to shield the embryo, engineering crops to overexpress soybean TI genes puts the protection right in the leaves and stems where pests feed. This approach has successfully thwarted defoliation by major pests like the corn earworm (*Helicoverpa zea*) without hurting the plant's overall yield (Birdwell et al., 2025; Sultana et al., 2023).

Post-Harvest Storage Protection

Globally, stored grain losses driven by key pests like the red flour beetle (Tribolium castaneum) represent a severe commercial threat (Kalpna et al., 2022). Although plant-derived trypsin inhibitors (TIs), notably from Albizia procera, can induce high mortality rates throughout every developmental stage of the insect, their raw delivery is often hindered by physical instability (Luneja, 2025; Mehmood et al., 2024). Recent breakthroughs are addressing this vulnerability by using nanotechnology to shield the proteins. Encapsulating these active TIs within zinc oxide or graphene nanoparticles prevents leaching, optimizes environmental stability, and ensures targeted release once the compound reaches the alkaline environment of the insect digestive tract (Mehmood et al., 2024).

Molecular Farming Support

A major hurdle in biopharming—the genetic modification of plants to synthesize high-value vaccines or therapeutic proteins—is the unintended degradation of the target product by endogenous host proteases prior to harvest (Grosse et al., 2018). To mitigate this internal proteolysis, researchers frequently employ the strategy of co-expressing plant serine protease inhibitors alongside the recombinant protein. This dual-expression approach successfully shields the biopharmaceuticals from host enzymes, optimizing overall accumulation yields (Clemente et al., 2019; He et al., 2021).

Cancer Prevention and Therapy: Malignant cells frequently secrete serine proteases to break down the surrounding extracellular matrix, a crucial step that enables tumor cells to invade local blood vessels and metastasize. To disrupt this pathway, plant-derived trypsin inhibitors (TIs)—most notably Bowman-Birk Inhibitors (BBI)—are being intensively investigated for their potent anti-carcinogenic properties (Clemente, 2014). By blocking the localized proteolysis required for cell proliferation and angiogenesis, BBIs downregulate critical cancer-associated signaling pathways. In vivo models have confirmed that this targeted inhibition significantly suppresses tumor growth and reduces the incidence of various malignancies, including reproductive (prostate and breast), colon, and skin cancers (Cristina et al., 2019).

Inflammation and Tissue Damage: Chronic tissue damage in inflammatory disorders like arthritis and Crohn's disease is often a direct result of unchecked enzyme release, particularly neutrophil elastase. To combat this, plant trypsin inhibitors—specifically Bowman-Birk Inhibitors (BBIs)—act as potent regulatory agents. Thanks to a dense network of disulfide bonds forming a resilient, cage-like framework, BBIs naturally resist degradation in the acidic human stomach (Clemente, 2014). This ensures they arrive in the intestinal tract fully active, where they systematically inhibit the over-activated inflammatory proteases of neutrophils and reduce downstream pro-inflammatory cytokines (Cristina et al., 2019).

Cardiovascular and Coagulation Disorders: Specific plant-derived proteins, including the Kunitz family and corn trypsin inhibitor, show immense therapeutic potential in preventing strokes, myocardial infarctions, and venous thromboembolism. They achieve this by directly blocking the precise proteolytic pathways responsible for platelet aggregation and blood coagulation, offering a natural mechanism to safely modulate thrombus formation.

Oral Delivery of Macromolecules: The oral delivery of peptide and protein-based drugs is heavily restricted by rapid enzymatic degradation in the digestive tract. To overcome this

challenge, plant-derived trypsin inhibitors can be utilized as safe, biodegradable barriers that protect delicate macromolecular drugs from premature breakdown, ensuring they safely navigate the gastrointestinal environment.

Antimicrobial and Antifungal Agents: Plant-derived trypsin inhibitors are increasingly recognized for their powerful, broad-spectrum antimicrobial properties, demonstrating significant efficacy against human pathogenic bacteria like Staphylococcus aureus and opportunistic fungi like Candida species. These inhibitors function by aggressively disrupting fundamental biochemical pathways and structural components within invading microbes. Remarkably, this targeted antimicrobial action does not compromise host safety; the proteins exhibit negligible cytotoxicity toward healthy human cells. As conventional antibiotics continue to lose efficacy due to global resistance trends, these stable botanical molecules provide a promising, non-toxic alternative for clinical infectious disease management.

Metabolic Syndrome and Obesity Management: Plant trypsin inhibitors offer a unique, non-stimulant approach to combating obesity and metabolic syndrome through their action as safe appetite suppressants. In the small intestine, the inhibition of active trypsin creates a biochemical illusion of incomplete protein breakdown. This physiological trigger signals the neuroendocrine system to prompt the sustained secretion of cholecystokinin (CCK), a primary gut hormone that communicates satiety directly to the brain. By strategically manipulating these natural energy-balance pathways, plant TIs present an exciting pharmacological frontier for controlling addictive eating behaviors and mitigating chronic overweight conditions (Cristina et al., 2019).

Conclusion

Mostly plant trypsin inhibitor are considered as anti nutritional compounds but due to their multifacet applications they are now recognizing as highly versatile macromolecules. In agriculture their ability to ward off pests and fungi naturally makes them ideal candidates for developing eco-friendly, bio-sustainable alternatives to chemical pesticides. Plant trypsin inhibitor remarkable structural stability, low mammalian toxicity, and high specificity toward dysregulated proteases make them ideal bio-scaffolds for the next generation of therapies targeting cancer, viral infections, and chronic inflammatory disorders.

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