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Integrating Speed Breeding and Genomic Approaches to Mitigate Seed Shattering in Brassica napus in Regenerative Agricultural Systems

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Abstract

Silique shattering is one of the major constrictions in the yield of Brassica napus (rapeseed/canola) and is responsible for pre-harvest yield losses ranging from 10 to 70%, depending on environmental conditions and harvest management. As agriculture moves towards regenerative systems characterized by reduced tillage, diversified rotations, and reduced agrochemical inputs, seed retention is increasingly framed as a resilience trait rather than a yield trait. This review synthesizes current understanding of four interconnected areas: the biological and developmental basis of dehiscence-zone regulation, regenerative agroecosystems considered as a distinct selection environment, genomic strategies for improving seed retention, and ideotype design frameworks for low-input production systems. The multigenic basis of pod shattering, governed by transcription factors including *SHP, IND, JAG* and *TCP* hormonal signaling networks, and cell-wall remodeling and cell-death-associated enzymes, motivates integrated breeding efforts that go beyond single-locus solutions. We also assess the combined, complementary contributions of marker-assisted selection, genomic selection that explicitly models genotype (G) × environment (E) × management (M) interaction, multiplex CRISPR editing, speed breeding, and multi-omics integration as approaches for achieving genetic gain more rapidly than through conventional breeding alone. Critical research gaps include the lack of standardized high-throughput phenotyping protocols, limited data from real-world regenerative field conditions, and the underuse of phenomics and multi-omics integration. Building on this synthesis, we propose, as a conceptual framework rather than a demonstrated pipeline, a self-reinforcing, data-driven breeding concept aimed at the development of shattering-resilient B. napus ideotypes suited to sustainable, low-input agroecosystems.

Keywords: Silique shattering; dehiscence zone; regenerative agriculture; genomic selection; ideotype breeding; seed retention

Introduction: The Need for Integrated Breeding in Regenerative Oilseed Systems

Brassica napus (rapeseed, canola) underpins global edible oil security, providing about 13–16% of global vegetable oil supply and serving as feed, biofuel feedstock, and industrial raw material (Raza et al., 2021; Tan et al., 2024). Genomic and molecular breeding have delivered significant gains in oil quality, seed yield, and stress resistance over recent decades, positioning B. napus as a cornerstone crop for food-energy transition systems (Lu et al., 2019). However, climate variability, labor scarcity, and pressure to produce more with fewer external inputs increasingly challenge these gains. Among the factors limiting realized yield, pod/seed shattering remains one of the most intractable problems in commercial B. napus. Because Silique dehiscence is only partially domesticated in this crop, shattering at maturity, during windrowing, and at mechanical harvest causes losses of 10–25% in favorable environments and 50–70% in unfavorable environments (Mustafa et al., 2022; Zaman et al., 2019). These losses are aggravated by high-throughput mechanical harvesting, and shattered seed can persist as a volunteer weed in subsequent rotations, increasing herbicide dependence (Raman et al., 2014). Traditional breeding has achieved only limited gains in shatter resistance, constrained by the narrow genetic basis for this trait in the species and by loss of some ancestral alleles during domestication (Raman et al., 2023; Stephenson et al., 2019). Introgression and pyramiding of natural quantitative trait loci (QTL) for rupture energy is slow and context-dependent, and even

well-characterized QTL often show inconsistent effects across the diverse, low-input field conditions typical of commercial production. Agriculture is simultaneously shifting toward regenerative, systems-based approaches that emphasize soil health, biodiversity, ecosystem services, and climate resilience rather than short-term, input-driven yield maximization (LaCanne & Lundgren, 2018; Lammerts van Bueren et al., 2018). Reduced tillage, longer rotations, cover crops, and reduced agrochemical reliance can increase profitability and ecological resilience, but may also increase crop exposure to abiotic and biotic stress and increase within-field heterogeneity (Giller et al., 2021; Pathirana & Carimi, 2022). Under these conditions, seed retention becomes an important resilience trait: ideotypes that are shatter-prone, and only marginally acceptable in highly managed, high-input systems, are less likely to perform well under longer harvest windows, greater environmental variability, and reduced access to chemical "rescue" interventions. Recent functional genomics, genome-wide association studies (GWAS), and gene-editing work have begun to resolve the complex genetic architecture of shattering, revealing multiple homoeologous loci and regulatory networks (e.g., SHP, IND, JAG, GH28, TCP8) that control dehiscence-zone differentiation and Silique strength (Gu et al., 2024; Raman et al., 2025; Zaman et al., 2021). These advances show that combining high-resolution mapping, model-to-crop translation, crop-wild-relative introgression, and multiplex genome editing can deliver substantial gains in pod-shatter resistance even for polygenic traits (Stephenson et al., 2019; Tan et al., 2024). However, most of this work has been conducted in high-input or experimental settings and is rarely framed around the performance requirements of regenerative agroecosystems specifically. A knowledge gap therefore remains at the interface of genomic breeding, pod-shatter genetics, and regenerative agronomy: it is not yet established how to design, combine, and deploy shattering-resilient B. napus ideotypes that are specifically suited to low-input, ecologically intensive, regenerative oilseed systems. This review addresses that gap by synthesizing the relevant evidence and proposing, explicitly as a conceptual framework, how these strands could be integrated.

Biological and Developmental Basis of Seed Shattering in *Brassica napus*: In B. napus, pods (siliques) consist of two valves joined to a central replum, with a specialized dehiscence zone (DZ) at the valve margin that determines shattering behavior (Zaman et al., 2019; Zhang et al., 2016). During development, valve-margin cells differentiate into a narrow strip of lignified endocarp-b cells and a non-lignified separation layer; tension between these tissues and the valve wall drives Silique opening at maturity (Raman et al., 2014; Whitelaw et al., 1999). Shatter-susceptible B. napus typically shows a single lignified valve-margin layer and a wider separation layer, whereas more resistant Brassica species develop two lignified layers that mechanically reinforce the suture (Zhang et al., 2016). Developmental studies further resolve this into graded lignification of endocarp a/b cells (L1–L3) toward the replum, correlating specific lignification patterns with shatter resistance (Nichol & Samuel, 2024). A distinct anatomical variant, the "lignified-layer bridge" (LLB), controlled by a single recessive locus on C09, further stabilizes the valve–replum junction and confers strong resistance (Chu et al., 2022). Molecularly, the established Arabidopsis pathway is conserved: MADS-box genes SHP1/2 specify valve-margin identity, IND and NST1/3 promote lignified-layer formation, while FUL and RPL repress valve-margin identity in valves and replum (Pabón-Mora, Wong, & Ambrose, 2014). Homologs of SHP, FUL, IND, ALC, NST, RPL and cell-wall hydrolases (ADPG1/2, PGs) are expressed in the Brassica DZ and associate with shattering variation across species and genotypes (Afridi et al., 2022). Genome-wide surveys identify 30–32 shattering-related homologs in B. napus and B. juncea, reflecting complex duplication and regulatory divergence (Yasin et al., 2024). CRISPR/Cas9 editing of SHP and IND homeologs shows that simultaneous attenuation of multiple copies reduces lignified and separation layers at the DZ and increases pod-shatter resistance, with unequal contributions among homeologs (e.g., BnA03.IND has a stronger effect than BnC03.IND) (Zaman et al., 2021; Zhai et al., 2019). Additional regulators such as JAGGED and TCP transcription factors modulate valve-margin width and replum development; editing BnJAG or tuning miR319–TCP3/BnTCP3.A8 activity alters replum size and DZ geometry, improving resistance while revealing trade-offs with Silique morphology (Cao et al., 2022; Zumajo-Cardona et al., 2025).

Note on terminology: in Table 1, the descriptors "promotes," "represses," and "modulates" summarize the predominant reported effect of each gene on shattering-related traits. These roles are frequently context-, tissue-, and home-log-dependent (e.g., BnA03.IND versus BnC03.IND) and can differ between genetic backgrounds or environments; the table simplifies for readability, and readers requiring stage or tissue-specific detail should consult the primary sources cited

Gene / Homolog	Gene Class	Locus	Role*	Primary function in DZ	Evidence level	Key reference
BnSHP1 / BnSHP2	MADS-box TF	A03, C03	Promotes	Specifies valve margin identity;	CRISPR-validated in B. napus	Zaman et al. (2021)
BnIND (BnA03.IND, BnC03.IND)	bHLH TF	A03, C03	Promotes	Promotes lignified & separation layer formation; BnA03.IND > BnC03.IND effect	CRISPR-validated in B. napus	Zhai et al. (2019)
BnFUL	MADS-box TF	A02, C02	Represses	Represses valve margin identity; antagonizes SHP	Candidate gene / natural variants	Pabón-Mora et al. (2014)
BnRPL	Homeobox TF	A01, C01	Represses	Maintains replum boundary; represses valve margin identity in replum	Candidate gene, QTL co-localization	Afridi et al. (2022)
BnALC	bHLH TF	A06, C06	Promotes	Modulates separation-layer differentiation downstream of IND	Natural variants, partially validated	Yasin et al. (2024)
BnJAG (JAGGED)	C2H2 zinc-fingerTF	A07, C07	Modulates	Controls valve-marginwidth and replum development	CRISPR-edited (BnJAG)	Cao et al. (2022)
BnTCP3 / BnTCP8 / BnTCP10	TCP TF	A08 (TCP3.A8), C08	Modulates	miR319 targets TCP3/8/10. alters replum size and DZ geometry	Candidate genes, QTL co-localization	Zumajo-Cardona et al. (2025)
BnNST1 / BnNST3	NAC TF	A09, C09	Promotes	Drives secondary cell-wall lignification in endocarp-bcells	Candidate genes, QTL co-mapping	Nichol & Samuel (2024)
BnNAC genes (multiple homologs)	NAC TF	A03, A07, C03	Modulates	Regulate cell-wall biosynthesis and senescence in Siliquewall tissue	GWAS-supported, not individually validated	Yasin et al. (2024)
LLB locus (lignified layer bridge)	Anatomical / single recessive gene	C09	Represses	Forms a lignified bridge stabilizing the valve–replum junction	Genetically mapped, phenotypically validated	Chu et al. (2022)
BnADPG1 / BnADPG2	Polygalacturonase (PG)	A03, C03	Promotes	Degrades middle-lamella pectin in the separation layer	QTL co-mapping, expression correlation	Jaradat et al. (2014)
BnGH28 / β-1,4-glucanases	Glycoside hydrolase	A03, C04	Promotes	Degrades hemicellulose in separation-layer cell walls	Candidate gene, QTL co-location	Afridi et al. (2022)
Auxin pathway (ARF3,	Hormonal signaling	A05, C05	Modulates	Localized auxin repression required for	Candidate genes, QTL	Dong & Wang (2015)

GA2OX2)				proper DZ patterning	co-mapping	
ABA / ethylene / jasmonate pathways	Hormonal signaling	Multiple loci	Modulates	Pathway-level shifts delay senescence and reduce mechanical stress on DZ	Transcriptomic association only	Jaradat et al. (2014)

Table 1. Key genes and regulatory factors controlling Siliqueshattering in Brassica napus. DZ = dehiscence zone; TF = transcription factor. *Role terms are simplified and context-dependent (see note above). "Evidence level" distinguishes genes with direct functional validation in B. napus (e.g., via CRISPR knockout/editing or genetic mapping of a phenotype-confirmed locus) from candidate genes supported only by expression, QTL co-localization, or GWAS association, per Reviewer guidance to avoid presenting all candidates with equal confidence.

This is a transcriptional network accompanied by hormonal signaling. Comparative transcriptomics of DZs in B. napus (DZ highly susceptible to shattering) and in more resistant Brassica relatives shows that correct separation and lignification of DZs are correlated with local repression of the biosynthesis, transport, and signaling of auxin in the DZ, whereas altered auxin gradients are correlated with DZ patterning (Dong & Wang, 2015; Jaradat et al., 2014). Up-regulation of ABA signaling and down-regulation of ethylene and jasmonate signaling pathways contribute to reduced shattering, delayed senescence, and an indirect decrease in mechanical damage to the pods. Stress on the DZ. Classical studies also suggested that cell-wall enzymes like β-1,4-glucanases and polygalacturonases are related in the degradation of the middle lamella and final cell separation in the abscission layer, and Brassica homologs of the cell-wall enzymes ADPG1/2 and PG are mapped under the QTL of shatter (Jaradat et al., 2014).

The quantitative genetic architecture of pod-shatter resistance in B. napus is highly polygenic, with both additive and non-additive (epistatic) effects. Early genome-wide QTL mapping identified at least 12 QTL across the A and C genomes (A03, A07, A09, C03, C04, C06, C08) explaining approximately 57% of genotypic variance for rupture energy, a mechanical measure of resistance (Raman et al., 2014). Subsequent analyses in interspecific derivatives and diverse germplasm have revealed additional main-effect QTL on A02, A05, C01, and C09, and multiple epistatic QTL pairs (additive × additive, additive × dominance, dominance × dominance) involving A–C genome interactions (Chu et al., 2022). Many QTL co-locate with candidate genes from the shattering cascade (*SHP1/2, FUL, IND, NST2, ADPG1, RPL, AG, ARF3, GA2OX2, TCP8/10, NAC* genes), supporting predominantly additive control from multiple small- to moderate-effect loci layered on a background of epistasis (Dhaliwal et al., 2021; Raman et al., 2025). GWAS and candidate-gene association studies confirm contributions of NAC and IND homologs and identify natural allelic variants introgressed from B. carinata and B. rapa that increase rupture energy, though often with linkage drag. Shatter resistance is heritable, but genotype×environment interaction is substantial, since temperature, humidity, and harvest conditions alter desiccation, cell-wall formation, and hormone status in developing pods.

Chr.	QTL / Locus name	Trait measured	Variance explained	Effect class	Co-locating candidate genes	pulation/source	Reference
A0 2	*qSH.A 02*	Rupture energy (RE)	~4–6%	Minor/additive	BnFUL, ARF	Interspecific B. napus × B. rapa derivatives	Raman et al. (2023)
A0 3	*qSH.A 03*	RE Siliquewall strength	~8–14%	Major	BnSHP1, BnIND, BnADPG1	Doubled-haploid B. napus mapping population	Chu et al. (2022)
A0 5	*qSH.A 05*	RE shattering index	~3–5%	Minor/additive	GA2OX2, ARF3	Diverse B. napus association panel	Dhaliwal et al. (2021)

A0 6	*qSH.A 06*	RE Siliquewall hemicellulose	~5–9%	Major	BnALC, BnNAC	B. napus × B. rapa introgression lines	Chu et al. (2022)
A0 7	*qSH.A 07*	RE collision test score	~4–7%	Minor/additive	BnJAG, BnNAC	Spring B. napus recombinant inbred lines	Raman et al. (2025)
A0 9	*qSH.A 09*	RE lignin content	~6–11%	Major	BnSHP2, BnNST1, BnIND	Winter B. napus mapping populations; DH lines	Raman et al. (2023)
C0 1	*qSH.C 01*	RE dehiscence index	~3–5%	Minor/additive	BnRPL, BnJAG	Interspecific B. napus germplasm	Dhaliwal et al. (2021)
C0 3	*qSH.C 03*	RE Siliquewall strength	~5–10%	Major	BnSHP1 (C copy), BnIND (C copy), BnADPG2	DH mapping populations; GWAS panels	Chu et al. (2022)
C0 4	*qSH.C 04*	RE collision loss	~4–6%	Minor/additive	BnGH28	B. napus association mapping	Raman et al. (2025)
C0 6	*qSH.C 06*	RE Siliquewall dry weight	~3–5%	Minor/additive	BnALC (C copy)	Diverse B. napus germplasm panel	Dhaliwal et al. (2021)
C0 8	*qSH.C 08*	Shattering index; RE	~4–7%	Minor/additive	BnTCP8, BnTCP10	B. napus RIL and GWAS panels	Cao et al. (2022)
C0 9	*qSH/L LB locus*	RE anatomical score (LLB)	~8–15%	Major	LLB gene (recessive), BnNST3	B. napus biparental crosses; anatomical screening	Chu et al. (2022)
A0 3 × C0 3	*E-QTL pair 1*	RE (additive×additive interaction)	~5–8%	Epistatic pair	BnSHP1/BnS HP2 homeologs	Interspecific B. napus mapping populations	Chu et al. (2022)
A0 9 × C0 9	*E-QTL pair 2*	RE (additive×dominance interaction)	~4–6%	Epistatic pair	BnSHP2 / LLB locus	DH and RIL populations; cross-genome interaction analysis	Chu et al. (2022)
A0 6 × C0 6	*E-QTL pair 3*	RE Siliquewall traits (interaction)	~3–5%	Epistatic pair	BnALC homeologs	Diverse B. napus germplasm; GWAS	Dhaliwal et al. (2021)

Table 2. Reported QTL, candidate genes, and epistatic interactions associated with seed-shattering resistance and related traits in Brassica napus across different mapping populations and germplasm sources. †Variance-explained values are as reported in the cited primary source and are not directly comparable across studies (see inclusion criteria above).

Collectively, current genetics in B. napus is constrained by narrow natural diversity for key dehiscence regulators, sub-functionalization, and redundancy of homeologs in the polyploid genome, and pleiotropic effects of major alleles on fruit morphology and yield components (Zhai et al., 2019; Zumajo-Cardona et al., 2025). Many core network components (e.g., ALC, PGs, additional NACs, and TCPs) remain incompletely validated functionally, and fine dissection of cis-regulatory variation and hormone–gene pathway integration continues to emerge (Dong & Wang, 2015; Yasin et al., 2024). Overcoming these constraints will require systematic multi-homeolog editing, targeted introgression from related Brassica species, and precise

modulation of DZ anatomy (e.g., LLB, controlled lignification gradients) to decouple shatter resistance from undesirable changes in Silique and seed characteristics.

Regenerative Agricultural Systems: A Proposed New Selection Environment

A system of agriculture that integrates reduced tillage, soil-carbon management, crop diversity, and retaining crop residues in a single management strategy that prioritizes soil. The core principles include little or no soil disturbance, continuous soil cover, living roots and high biological diversity, frequently including livestock or trees, and are always aimed at restoring soil organic matter, improving nutrient cycling and holding soil carbon, while maintaining yields (Khangura et al., 2023; Nethra, Rao, & Varshini, 2025). Conservation agriculture-style practices such as reduced or no tillage, diversified rotations, cover crops, and residue retention, and are regularly demonstrated to enhance soil organic carbon, soil structure, water infiltration and microbial activity; the benefit being dependent on climate, soil type and length of the retention period (Al-Shammary et al., 2024; Jordon et al., 2022; Patil et al., 2025). Based on the results of the meta-analysis and modelling, it has been suggested that: The cover cropping, residue retention, organic amendments and (in some systems) agroforestry. can significantly increase soil-carbon stocks on decadal time-scales, while reduced tillage can significantly decrease them. Gains with alone are more moderate and are dependent on context (Patil et al., 2025). The practices may be expected to change the selection environment for silique shattering indirectly, through alteration of canopy microclimate, soil-moisture regime, and plant, for shattering traits this has not been directly measured, but nutrition, nevertheless, is a factor. Increased surface cover and canopy diversification are documented to buffer extremes of temperature, wind, and vapor-pressure deficit, creating more humid, thermally stable microclimates (Al-Shammary et al., 2024; Fahad et al., 2022); we propose that, by analogy, this could slow Silique desiccation and reduce the rapid shrink–swell cycling associated with mechanical failure of dry pods, though this specific link has not been tested in B. napus. Similarly, increased soil organic matter and improved soil structure are established to increase water-holding capacity and infiltration (Al-Shammary et al., 2024); we suggest this could buffer drought-driven fluctuations in root-zone water status and reduce the frequency of abrupt wetting–drying cycles during reproductive development, a period in which water-status instability is known to affect pod and seed development more generally (Kalra, Goel, & Elias, 2024). At the same time, increased biologically mediated nutrient supply and reduced synthetic inputs are expected to shift the timing and intensity of nitrogen and other nutrient delivery to reproductive tissues, a change that could smooth, or in poorly managed systems accentuate, nutrient-related stress during flowering and pod fill, with unknown net effects on pod abortion, lignification, and shattering risk. If this hypothesis holds, regenerative management would be expected to favor ideotypes with pod walls that tolerate a longer standing period before harvest consistent with delayed or variable harvest timing across diversified rotations and greater tolerance of repeated minor wet–dry cycling without crack propagation. Better buffering and microclimatic moderation of soil moisture may suggest that siliques do not need to be so brittle and easily thresh able, but greater exposure to late storms and mechanical disturbance due to dense siliques or intercrops may suggest that pods should deform and twist without dehiscence before harvest. open quickly when combined impact (Hao et al., 2025; Ogwu & Kosoe, 2025). Conceptual work on root and whole-plant stress responses suggests that phenotypes combining high performance with stress-responsive plasticity are generally favored in heterogeneous, biologically active soils (Colombi et al., 2019; Kalra, Goel, & Elias, 2024); whether this generalization extends to shattering-related pod traits specifically remains an open, testable question rather than an established finding. We therefore propose plastic adjustment of reproductive timing, pod-wall composition, and cuticle properties, combined with elastic (rather than brittle) mechanical behavior, as a trait complex worth testing in future breeding for regenerative landscapes, while emphasizing that this integration between shatter-resistant pod architecture and stress-plastic root systems is, at present, a proposed research direction rather than a validated breeding target.

Genomic Approaches to Improve Seed Retention

Genomic approaches offer powerful and complementary avenues for improving seed retention, particularly when breeding for complex environments such as regenerative systems. Marker-assisted selection (MAS) is most efficient once major seed-shattering loci have been mapped and linked to robust DNA markers. In soybean, the shattering-resistance allele pdh1 and tightly linked SNP/InDel markers showed a significant marker-phenotype association ($p < 0.05$) and near-fixation of the resistance allele (96.6% of genotypes) in the validation population reported by (Shimbwambwa et al. 2024); comparable marker systems in B. napus (e.g., KASP assays for BnSHP1-linked markers) have accelerated selection for pod-shattering tolerance, though reported accuracies are

generally population- and marker-specific and should not be assumed to transfer unchanged to other germplasm or crops (Larson et al., 2020).

For more polygenic seed-retention phenotypes and correlated traits such as yield, height, and maturity, genomic selection (GS) using dense genome-wide SNPs is used to predict breeding values. Studies in rice, chickpea, and groundnut show that multi-trait, multi-environment prediction models , especially those that explicitly model genotype×environment (G×E) interaction , improve predictive ability and selection efficiency across contrasting water, temperature, or management conditions (Bhandari et al., 2019; Hosain et al., 2025; Vieira, Nogueira, & Fritsche-Neto, 2025); direct evidence of comparable gains specifically for shattering-related traits in B. napus is still limited, and this remains an area for crop-specific validation. Kernel-based and factor-analytic GS models, as well as deep-learning approaches, exploit correlations between traits and environments; recent reviews propose extending these to full G×E×M (genotype × environment × management) structures by adding Environics and management covariates, which would be particularly relevant for breeding under variable regenerative management , though such G×E×M models remain largely conceptual for this trait and crop combination at present (Crossa et al., 2022; Vieira, Nogueira, & Fritsche-Neto, 2025).

Genome editing complements MAS and GS by directly modifying causal genes for shattering and pod strength. CRISPR/Cas platforms, using multiple guide RNAs, allow precise modification of domestication-related loci and can achieve fine-tuning of seed retention without the linkage drag typical of conventional backcrossing, on shorter timelines (Hanif et al., 2023; Kumar et al., 2024). Seed-fluorescence reporter cassettes, as demonstrated in maize, can simplify segregation of edited alleles from transgenes and the construction of mutant libraries (Yan et al., 2021); careful design remains necessary to minimize off-target edits and vector-backbone insertions, which are detectable only by whole-genome sequencing. As with MAS and GS above, editing efficiencies and outcomes reported in model systems or other crops should be treated as indicative rather than directly transferable to B. napus without crop-specific validation.

Beyond single loci, multi-omics integration, combining genome, transcriptome, metabolome, and, increasingly, epigenome data, provides a systems-level view of seed-shattering biology. Reviews of seed quality and crop improvement note that integrating omics layers can reveal regulatory networks, cell-wall remodeling pathways, hormone signaling, and stress responses relevant to seed retention, potentially enabling discovery of new targets and more informative markers (Kumar et al., 2024; Raman et al., 2023). Multi-omics-powered genomic prediction has begun to show consistent accuracy gains over genomic-only models for complex, multigenic traits, provided that integration uses model-based data fusion rather than simple concatenation (Montesinos-López et al., 2025); however, this evidence base derives mainly from other traits and crops, and its extension to shattering resistance in B. napus specifically remains to be demonstrated. Table 3 summarizes these approaches together with an indication of how far each has actually been validated in B. napus shattering research, as distinct from validation in other crops or traits.

Together, MAS for major shattering loci, GS with multi-trait, multi-environment and (prospectively) G×E×M models, precise CRISPR editing of valve-margin and regulatory genes, and multi-omics integration form a genomic toolbox with real potential to support ideotype design for improved seed retention. At present, however, only MAS for major loci and targeted CRISPR editing have been directly demonstrated for shattering resistance in B. napus; the GS/G×E×M and multi-omics components of this toolbox are extrapolated from other crops and traits and require crop- and trait-specific validation before their predictive accuracy and practical readiness in B. napus can be considered established.

Speed Breeding as a Genetic Gain Accelerator

Speed breeding (SB) accelerates genetic gain by shortening the seed-to-seed cycle through controlled-environment manipulation, enabling more generations and selection cycles per year and tighter coupling with genomic tools. Modern systems have demonstrated 4–6 generations per year in oilseeds, cereals, and legumes; empirical comparisons report that this can double annual genetic gain relative to conventional schemes using off-season nurseries (Ćeran et al., 2024; Samantara et al., 2022; Watson et al., 2018). These figures are established results from replicated experiments in the crops and traits studied, but have not, to our knowledge, been specifically demonstrated for shattering-resistance traits in B. napus.

Controlled-environment optimization involves modifying photoperiod, light spectrum, and temperature to shorten the time to flower and maturity. Optimized day/night temperatures and extended photoperiods (typically 20–22 h light) are now commonplace and six or more generations per year are now possible in long-day or day-neutral crops such as wheat, barley, chickpea, pea, and canola (Williams et al., 2024). LED-based light-spectrum engineering (e.g.,

blue-enriched, far-red-adjusted spectra) further fine-tunes plant architecture and flowering in both long-day and short-day species, while temperature regulation interacts with photoperiod to minimize developmental delays and stress (Pandey et al., 2022).

Rapid generation advancement is achieved by accelerating single-seed descent and inducing earlier flowering. Under SB, plants are grown at high density, seeds are harvested at or before physiological maturity, and methods such as immature-seed germination or embryo rescue are used to achieve 4 to 7+ generations per year in cereals, legumes, and oilseeds (Samantara et al., 2022). Protocols combining early-flowering induction (via photoperiod and temperature) with aggressive single-seed descent have reduced cultivar development time from a decade to 2–4 years in the crops directly studied (Cobb et al., 2019; Kumar & Walia, 2024).

Integration with genomic prediction can, in principle, turn SB into a stronger genetic-gain accelerator by enabling early-cycle genomic selection. Genomics-assisted speed breeding, combining MAS, GS, and genome editing in rapidly advancing populations, has been projected in modelling and simulation studies to reduce generation length 2.5–5-fold with increased selection accuracy (Patel & Gurjar, 2024); this is a projection from simulation rather than an empirical field result for B. napus shattering traits specifically. In canola, multivariate genomic-selection models using SB-measured traits such as height and flowering time have improved yield prediction and supported indirect selection before field trials, an empirically demonstrated result, though again not for shattering resistance itself (Watson et al., 2019). Simulation studies in allogamous crops indicate that combining SB with genomic selection ("Speed GS") could reduce cycle length from 11 years to 4–9 years, with careful management of inbreeding needed especially for low-heritability traits; this projection has not yet been validated empirically in B. napus. The restrictiveness of scaling remains relevant. SB needs to be controlled environment infrastructure and dust control. Light, temperature, humidity, and dust control management. expensive nutrition in many public sector or low resource breeding programs (Watson et al., 2019). It is often necessary to optimize this protocol according to the specific crop and genotype, especially the short-day and photoperiod-sensitive material (Patel & Gurjar, 2024). Besides, not all complex adaptation traits and genotype × environment interactions are completely known. Unfortunately, the expression of these traits under SB conditions should not be taken for granted and the selection of SB lines must be carefully tested in the multi-environment field to avoid the selection of growth-chamber artefacts and to ensure that SB lines grow well under target conditions, such as in the target production environments, including non-target regenerative systems that have not been directly tested under SB protocols.

Designing Shattering-Resistant Ideotypes for Regenerative Systems in Oilseed Rape

In regenerative systems, shattering-resistant ideotypes must both withstand mechanical harvesting and integrate into low-input, residue-retentive, diversified rotations. This requires combining pod, plant, and mechanization traits so that yield, soil cover, and system resilience are improved rather than traded off.

The oilseed-rape ideotype concept is shifting in focus from a 'maximum-yield' plant toward a compact, lodging-resistant, machine-ready architecture with strong pods and efficient canopy light use. High pod-shatter resistance is associated with thicker, heavier pericarps, increased pod-wall dry weight, higher hemicellulose and lignin content, and optimized vascular-bundle traits, while compact plants with upright leaves, greater primary-branch height, and reduced branch angle facilitate low-loss combine harvest and better light interception in dense stands (Liu et al., 2024; Qing et al., 2021). Ideotype proposals for high yield in winter and spring oilseed rape emphasize few but productive inflorescences on the main stem, long pods with high seed number per pod, and limited pod abortion, which can be combined with shatter-resistance traits to define a regenerative ideotype (Siles et al., 2021).

Trait-pyramid strategies draw on the multigenic control of pod-shatter resistance. GWAS and QTL studies in B. napus and B. rapa identify robust loci on A06, A09, and C-genome chromosomes, including BnSHP1/IND-related regions, which can be stacked using marker-assisted and genomic selection (Raman et al., 2025; Zhu et al., 2025). Combined "genotype–phenotype" pipelines using KASP markers (e.g., S12.68 for BnSHP1) alongside standardized collision or rupture-energy assays have produced near-zero-shattering experimental lines while retaining high oil content, disease resistance, and favorable plant height and branching for mechanization. Pyramiding complementary mechanisms, for example, SHP/IND pathway alleles together with vascular-bundle reinforcement or altered downstream regulators such as BnIND, reduces reliance on a single locus and should confer more durable resistance across variable climates (Qing et al., 2021; Raman et al., 2025; Stephenson et al., 2019).

Mechanical-harvesting compatibility is central to ideotype design. Pod shatter-resistance indices (SRI) exceeding

0.58 under low reel speed are needed to withstand reel impact and header vibration, yet most existing commercial varieties fall short of this benchmark, indicating room for breeding improvement (Qing et al., 2021). Field evaluations indicate that compact, high-branch-point varieties with strong pods, combined with appropriate plant-density and row-spacing choices (e.g., approximately 45×10^4 plants ha^{-1} with 15 cm rows), improve yield, pod-shatter tolerance, standability, and combine throughput (Kuai et al., 2015; Qing et al., 2021). Screening and selection therefore need to combine SRI or rupture energy, lodging, yield, soil cover, and system resilience improvements rather than trade-offs, branch architecture, maturity synchrony, and machine-loss measurements under realistic combine settings.

Stability in varying years and weather extremes is also desired for regenerative systems. Better pods reduce pre-harvest and header losses, leading to longer harvest periods. Decreasing volunteer seed banks and weed management also decrease volunteer seed banks (Liu et al., 2024; Zhang et al., 2012). There are, However, compromises and pod strength vs. yield potential could occur: Excessive indehiscence (e.g., the failure of the dehiscence layer (near-complete loss) makes threshing difficult and raises the amount of grain lost in the combine, so intermediate 'robust but openable' phenotypes are preferred (Stephenson et al. 2019). Some studies report that genetic control of shatter resistance and major yield is independent. Furthermore, the results indicated that pod strength could be improved without lowering seed size, indicating potential for higher yields of seeds while maintaining the same size of the pods. number or oil content (Kuai et al. 2016; Morgan et al. 2000). Meanwhile, anatomical, and biochemical changes that make pod walls stiffer, by thickening cell walls and changing cell energy cost at the valve margin, have a cost to the plant that must be traded off against efficient extraction and packing of seed.

Overall, a shattering-resistant regenerative ideotype for oilseed rape would combine multigenic pod strength (cell-wall composition and dehiscence-zone regulators), a compact, lodging-resistant architecture adapted to efficient mechanical harvest, and yield-supporting reproductive traits such as long, well-filled pods on a dominant main inflorescence. Table 4 consolidates the quantitative or semi-quantitative targets discussed above into explicit phenotypic breeding targets, addressing the need for the ideotype framework to move beyond a purely qualitative description

Target trait	Proposed breeding target	Rationale	Key reference
Pod-shatter resistance index (SRI)	≥ 0.58 under low reel speed	Threshold reported to withstand reel/header impact at low reel speed	Qing et al. (2021)
Rupture energy	Above population median for the target environment; exact value population-specific	Mechanical measure of resistance; not yet standardized across studies (see Section 8)	Raman et al. (2023)
Pod-wall composition	Increased lignin & hemicellulose relative to susceptible checks	Associated with thicker, heavier pericarp and shatter resistance	Liu et al. (2024)
Plant density/row spacing	~45×10^4 plants ha^{-1}; 15 cm rows	Combination reported to improve yield, standability, and combine throughput	Kuai et al. (2015)
Branch architecture	Higher primary-branch point; reduced branch angle; upright leaves	Facilitates low-loss combine harvest and canopy light interception	Qing et al. (2021)

Reproductive structure	Few, productive inflorescences on main stem; long pods, high seeds/pod; low abortion	Merges yield ideotype with shatter-resistance ideotype	Siles et al. (2021)

Table 3: Proposed measurable phenotypic targets for a shattering-resistant, mechanization-compatible Brassica napus ideotype, consolidated from the sources discussed in this section. Values are drawn from the specific studies and populations cited and should be treated as illustrative starting benchmarks rather than universal thresholds

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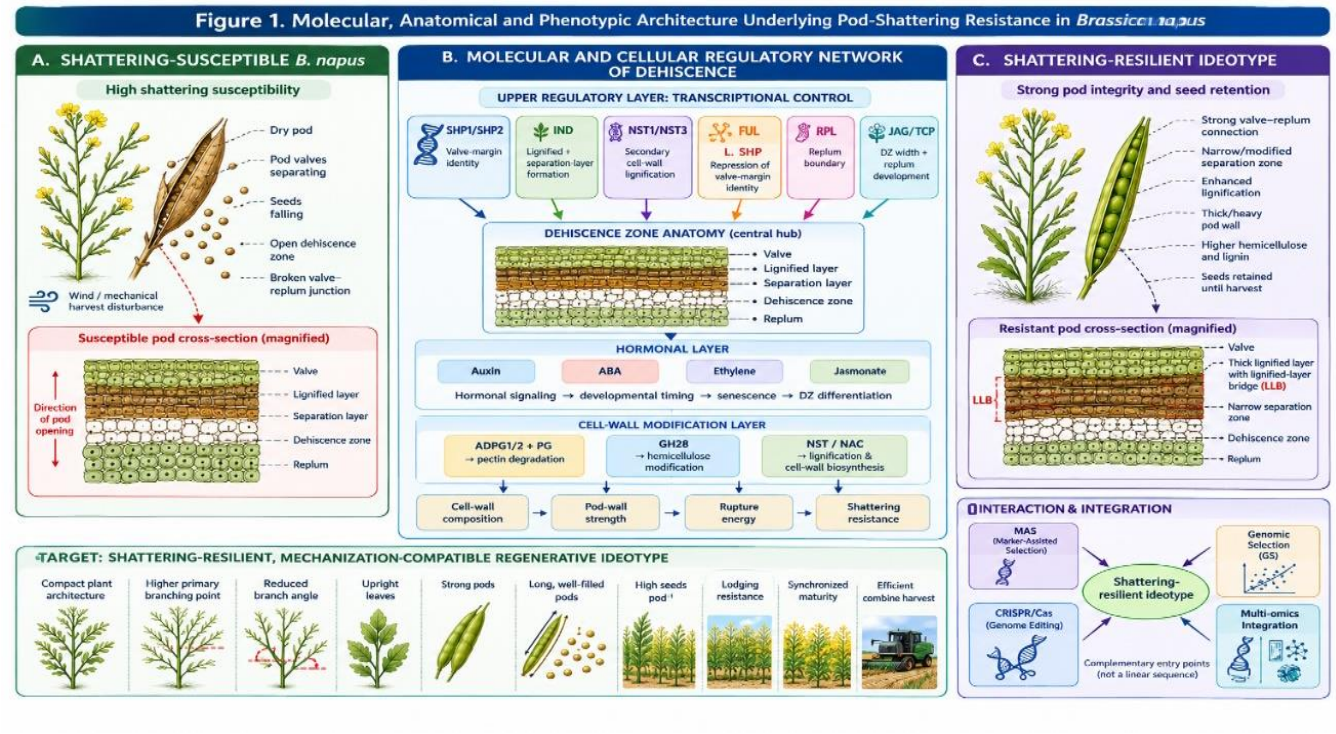


Systems-Level Integration: A Proposed Regenerative Breeding Pipeline Framework

A systems-level regenerative breeding pipeline can be conceptualized as a continuous, data-driven loop linking genomic discovery, accelerated generation turnover, and multi-environment validation in regenerative production systems. In Step 1, genomic discovery would leverage dense genotyping, multi-omics, and genomic prediction to identify alleles and haplotypes associated with complex traits such as stress resilience, resource-use efficiency, and Soil-health contributions, drawing on integrated genomic–environmental data and prediction-based breeding frameworks described in other crops (Fritsche-Neto et al., 2025; Xu et al., 2022). In Step 2, speed-breeding cycle compression would use rapid generation advance and genomics-assisted breeding to fix superior allele combinations and iterate crosses several-fold faster, coupling genomic selection and speed breeding to raise annual genetic gain while keeping costs manageable (Anilkumar et al., 2022; Bhat et al., 2023). In Step 3, regenerative-system field validation would evaluate candidate lines in diverse, management-intentional environments representing contrasting regenerative practices (reduced tillage, cover crops, low synthetic inputs), with environment and high-throughput phenotyping capturing G×E×M responses and system-level outcomes such as yield stability, input efficiency, and resilience. In Step 4, feedback into genomic models would route genomic, phenotypic, environmental, and management data back into updated prediction models, refining trait environment management associations for the next discovery and selection cycle (Messina et al., 2025; Xu et al., 2022).

If realized, such a loop would in principle become self-reinforcing: each deployment in an actual regenerative production system would recalibrate the underlying models and enlarge their domain of validity, progressively

improving data-driven predictions and allowing breeders to design germplasm for emergent, system-level properties rather than yield alone. We emphasize that this is a proposed rationale for why such integration should be worth pursuing, not a demonstrated outcome; the components described (genomic discovery, speed breeding, multi-environment phenotyping, and model updating) have each been demonstrated individually in various crops, but their combination into a closed, self-improving loop specifically for shattering resistance in regenerative B. napus systems remains untested.

Research Gaps and Future Directions

Several areas currently limit progress toward robust, field-relevant shatter resistance. First, there is no universally accepted, standardized protocol for shattering phenotyping. Different studies use rupture energy via pendulum tests, pod shatter-resistance index, collision tests, or categorical visual scores, and differ in maturity stage, environmental conditions, and units of measurement, making results difficult to compare across experiments and crops (Qing et al., 2021; Wang, Ripley, & Rakow, 2007); even within Brassica, pendulum-based rupture energy, dehiscence indices, and field-loss estimates remain unharmonized. Second, most phenotyping and genetics work has been conducted under conventional, well-managed experimental conditions, with little data from low-input or regenerative systems, despite evidence that shattering is strongly modified by weather, management, and harvest method (Cobb et al., 2013; Maier et al., 2022); this limit understanding of the genotype × environment interactions relevant to mechanized, sustainable production. Third, although rupture-energy and collision-test measurements exist, truly high-throughput pod-strength assays remain rare; most current methods are labor-intensive, destructive tests on small samples, which slows genetic mapping, genomic selection, and validation of gene-edited (e.g., CRISPR-derived) material (Tsakirpaloglou, Septiningsih, & Thomson, 2023). A fourth, and currently underexploited, gap concerns the broader integration of modern molecular 'omics' technologies into shattering research. Most existing work has focused on genomics and, to a lesser extent, transcriptomics of bulk dehiscence-zone tissue; several complementary approaches remain untapped for this specific trait:

Transcriptomics beyond bulk profiling could more precisely resolve the timing and tissue context of expression changes in shattering-cascade genes (SHP, IND, FUL, NST, JAG, TCP) across developmental stages and genetic backgrounds, extending the candidate-gene-driven expression studies cited in Section 2 to genome-wide, time-resolved profiling.

Proteomics and post-translational modification profiling could clarify whether transcriptional differences in shattering-cascade genes are reflected at the protein level and identify regulatory steps (e.g., protein stability, modification of cell-wall-remodeling enzymes) not visible from transcript data alone, an angle that has received little attention in Brassica shattering research to date.

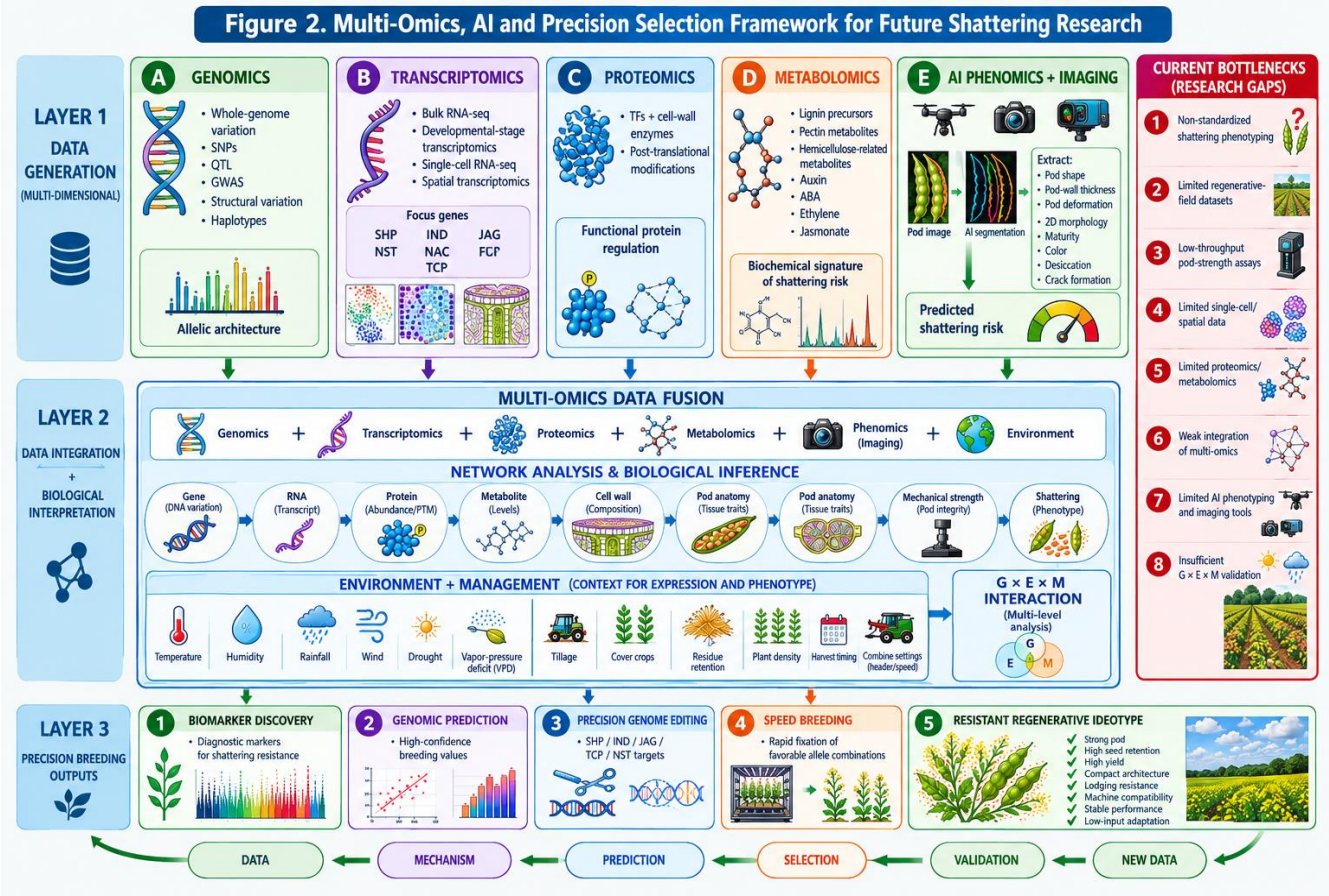
Metabolomics of the dehiscence zone and pod wall could characterize the cell-wall and hormone metabolite pools (lignin precursors, pectin breakdown products, auxin/ABA/ethylene/jasmonate metabolites) underlying the anatomical and hormonal patterns described in Section. **providing a more direct biochemical readout of shattering risk than gene expression alone.**

Single-cell transcriptomics could resolve the distinct cell types within the dehiscence zone, lignified endocarp-b cells, the non-lignified separation layer, and adjacent replum and valve tissue, which are currently studied largely as bulk-tissue averages; this resolution could clarify which specific cell populations drive the graded lignification patterns (L1–L3) described by Nichol & Samuel (2024).

Spatial transcriptomics and imaging-based spatial omics could map gene expression directly onto dehiscence-zone anatomy in situ, preserving positional information (e.g., proximity to the replum, valve margin, or LLB region) that is lost in dissociated single-cell or bulk approaches, and could help explain why anatomical variants such as the LLB locus (Chu et al., 2022) confer strong resistance.

Realizing the value of these approaches will depend less on any single technology than on integrated, multi-omics analysis: combining genomic, transcriptomic, proteomic, metabolomic, and (where available) epigenomic data within a common analytical framework, rather than treating each omics layer in isolation. As noted in Section 4, model-based data fusion across omics layers has begun to outperform single-omics or genomics-only prediction for other complex traits (Montesinos-López et al., 2025); applying this integrative, multi-omics strategy specifically to seed-shattering resistance in B. napus, linking DNA variation, expression, protein, and metabolite data to anatomical and mechanical shattering phenotypes, represents a substantial and currently open research opportunity, and we consider one a priority direction for this field alongside the phenotyping and regenerative-systems testing gaps discussed above. Finally, the rapid development of imaging, sensors, and machine learning in plant phenotyping has not yet been fully applied to

shattering. Integration of AI-driven phenomics, automated imaging of pod deformation, real-time monitoring of maturation, and prediction of field loss from structural and anatomical traits, remains largely a future direction rather than established practice, notwithstanding early work characterizing the structure, histology, and chemical composition of pods that has already identified measurable visual and anatomical correlates of shatter resistance



Conclusion

Pod shattering is an economically significant constraint on Brassica napus yield, but it is also one of the more tractable targets for genomics-enabled breeding, given rapid recent progress in understanding its molecular and developmental basis. To avoid the repetitiveness noted in review, we separate below what this synthesis treats as (i) well-established knowledge, (ii) our proposed integration framework, and (iii) the future research needed to evaluate that framework.

Established knowledge. Pod shattering in B. napus is controlled by a complex, multigenic network of transcription factors, hormonal signals, cell-wall enzymes, and anatomical specializations, compounded by homeolog redundancy in the allopolyploid genome (Section 2). A subset of these genes notably SHP, IND, and FUL pathway members, have direct functional validation in B. napus (e.g., via CRISPR editing), while many other candidates remain supported only by expression association, QTL co-localization, or GWAS (Table 1). No single gene or locus provides a complete solution; durable resistance is expected to require pyramiding of complementary mechanisms. MAS for major loci and targeted CRISPR editing of valve-margin regulators are demonstrated, working approaches in this crop (Sections 2 and **Proposed integration framework.** We propose , as a framework for future work, not as an established result , that regenerative-agriculture management could plausibly shift the selection environment for shattering traits (Section 3, explicitly flagged there as hypothesis-level), that genomic selection and multi-omics integration could extend the demonstrated toolbox of MAS and CRISPR editing once validated for this trait (Section 4), that speed breeding could be coupled with genomic selection to compress breeding cycles (Section 5), and that these elements could in principle be organized into a self-reinforcing G×E×M breeding loop (Section 7). Each component draws on factual evidence from this or other crops, but their combination into an operating pipeline for shattering-resilient, regenerative-adapted B. napus ideotypes (Table 4) has not yet been built or evaluated as such.

Future research needs. Priority gaps include: standardizing shattering-phenotyping protocols across rupture-energy, collision, and visual-scoring methods; generating phenotypic and genomic data under actual low-input and regenerative field conditions rather than only high-input trial environments; developing high-throughput, non-destructive pod-strength assays; and applying integrated multi-omics analysis , transcriptomics, proteomics, metabolomics, single-cell and spatial transcriptomics , to resolve the cellular and biochemical basis of dehiscence-zone differentiation at a resolution current bulk-tissue studies cannot provide (Section Addressing these gaps will require coordinated, open data-sharing across research groups and investment in regenerative-system trial networks.

Taking together, we see incorporation of shattering resilience into a data-driven, iteratively updated breeding framework evaluated, rather than assumed, across genuinely regenerative production environments, as a promising direction for developing B. napus cultivars that can support both grower livelihoods and environmental goals under increasingly variable production conditions in the coming decades.

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