

Impact of mixing pressure and temperature on dough and bread properties: A comparison between Verna wheat flour and refined type “0” bread flour

G. Angeloni^a, A. Spadi^{a,*}, A. Cappelli^b, F. Corti^a, C. Vizzo^a, L. Bruchi^a, A. Parenti^a, P. Masella^a

^a DAGRI, Department of Agriculture, Food, Environment and Forestry, University of Florence, Piazzale delle Cascine 16, 50144, Firenze, Italy

^b San Raffaele University of Rome, Rome Via di Val Cannuta, 247, 00166, Roma, Italy

ABSTRACT

This study investigated the combined influence of mixing pressure (atmospheric vs. −0.4 bar vacuum) and temperature (25 °C vs. 5 °C) on the rheology, bread quality and crust morphology of two wheat flours differing in gluten strength: a refined Type “0” flour (W260) and an ancient Verna flour (W150). Doughs were characterized by alveograph tests, texture profile analysis (TPA), and image analysis of crumb and crust structure.

Low temperature mixing improved dough extensibility and swelling in both flours, demonstrating a consistent thermal effect on gluten development. Vacuum mixing selectively increased deformation energy in the strong flour but had negligible effects in Verna, confirming that pressure-based strengthening requires sufficient baseline gluten potential. In refined flour, vacuum mixing enhanced dough strength but reduced loaf height due to decreased gas nuclei, while low temperature promoted better expansion. Weak Verna doughs were largely insensitive to processing changes, remaining genotype bounded.

Overall, vacuum mixing emerged as a conditional lever—effective only above a gluten strength threshold ($W \geq 220$ –240)—whereas low-temperature mixing proved a robust, flour-independent optimization. These findings define a mechanistic “design rule” linking flour strength to process responsiveness, with practical implications for industrial breadmaking optimization.

1. Introduction

Dough rheology and bread quality are jointly determined by both ingredient composition and process conditions. Prior research has extensively explored ingredient-based strategies, including protein composition, fibre addition, hydrocolloids, and enzymes (Cappelli et al., 2020b).

Parallel, a number of studies have examined individual processing variables such as mixing time, temperature, speed, mixer geometry, temperature, speed, mixer geometry, and dough resting time *e* (Cappelli et al., 2018; Haixi et al., 2025; Venturi et al., 2022; Yue et al., 2020; Cappelli, Guerrini et al., 2020). The need to improve wholemeal and ancient grain formulations has further stimulated interest in the process domain, since bran interferes with gluten network formation, reducing extensibility, increasing tenacity, and lowering bread quality (Guerrini et al., 2020; Noort et al., 2010; Parenti et al., 2020; Venturi et al., 2022). Some process interventions, such as low-temperature mixing or use of solid CO₂, have demonstrated clear benefits in gluten development and final loaf volume (Watanabe et al., 1992). Recent work in Food Chemistry confirmed that thermal history modulates protein–protein interactions and rheological behaviour even before mixing, demonstrating

that temperature directly conditions the molecular state of gluten-forming proteins (Yang et al., 2023). Other interventions, notably vacuum mixing, have shown system-specific effects in noodles and breads (Gao et al., 2017; Liu et al., 2015; Paulik et al., 2021)

However, a critical gap remains, temperature and pressure have been investigated almost exclusively in isolation, and the potential synergy or antagonism between these two fundamental levers has not been systematically quantified. This gap is especially relevant for wholemeal and ancient flours, where the disruptive effect of bran exceeds the leverage typically obtained from individual process interventions. Therefore, the aim of this study was to quantify the combined effect of mixing temperature and pressure on dough rheology, bread quality and crust morphology, using flours with different gluten potential. Our goal was not simply to detect statistical differences, but to understand when process factors remain effective—and when dough behaviour becomes genotype-bounded.

* Corresponding author.

E-mail address: agnese.spadi@unifi.it (A. Spadi).

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2. Materials and methods

2.1. Materials

Two commercial soft wheat flours were used for the experiments: a Type '0' flour (W260; 11.5% protein) and a Type '1' Verna ancient grain flour (9.6% protein). Other ingredients included commercial dehydrated brewer's yeast (*Saccharomyces cerevisiae*), salt (NaCl), and mineral water.

2.2. Experimental design

Two process variables were manipulated during dough mixing: pressure (atmospheric vs. -0.4 bar vacuum) and temperature (25 ± 1 °C vs. 5 ± 1 °C). A full factorial 2×2 design was performed separately for each flour ($n = 4$ conditions $\times$ 3 replicates = $n = 12$ doughs per flour).

Prior to mixing, all ingredients were equilibrated at 25 ± 1 °C. Dough temperature was measured at end of mixing with a thermocouple probe.

2.3. Vacuum and temperature control

Doughs were mixed in the Chopin alveograph mixer. To enable controlled under-pressure mixing, a custom 3D-printed lid was retro-fitted to the mixing chamber, equipped with a pressure gauge and vacuum port. Sealing interfaces were mechanically improved to minimise leakage. A dry vacuum pump (VTS6, Vuototecnica S.r.l., Italy) supplied the negative pressure. Temperature was controlled by circulation of thermostated water (chiller) through the alveograph jacket. Pressure was monitored continuously and remained stable throughout mixing.

Each dough contained 250 g of flour, 2% yeast (flour basis) and a NaCl solution (2.5%) added to achieve a constant final dough hydration of 48%. Mixing was performed as follows.

- 1 min at atmospheric pressure
- removal of residues from bowl walls
- +6 min mixing
- for vacuum treatments only: the custom lid was mounted after minute 1, vacuum was applied and maintained for the remaining 6 min of mixing.

Doughs were extruded and processed according to ISO 27971 (ISO, 2008) prior to alveograph testing.

2.4. Breadmaking

Doughs were hand-shaped into batards, placed in aluminium pans and proofed. For doughs mixed at 25 °C, proofing was carried out at 28 °C for ~70 min; for doughs mixed at 5 °C, proofing was extended to ~130 min due to lower initial dough temperature. Dough temperature was re-evaluated during proofing. Despite the different mixing temperatures (5 °C and 25 °C), doughs reached comparable final temperatures at the end of proofing, immediately prior to baking. Baking was performed in a ventilated oven at 180 °C for 25 min. Loaves were cooled at ambient temperature and stored in plastic bags prior to testing.

2.5. Methods

Dough rheology was quantified by Chopin alveograph (AlveoLab, Chopin Technologies, France) according to ISO 27971. Five dough discs per dough were tested after 20 min rest. The following parameters were extracted: P (tenacity), L (extensibility), G (swelling), W (deformation energy), P/L (equilibrium ratio), Ie (elasticity).

Crust colour was measured in RGB space using a tristimulus colorimeter. Loaf weight was measured with a laboratory scale (Kern, Germany). Bread volume was determined using the millet displacement method (Technical, 2009). Loaf height was measured at mid-length

using a calliper.

Texture Profile Analysis (TPA) was performed on 2×2 cm crumb cubes using a custom-built texturometer equipped with a dual-range load cell (Vernier, USA) and connected to LabQuest 2 (Vernier) for force-time acquisition. Each sample was subjected to double compression. Hardness, cohesiveness, springiness, gumminess and chewiness were calculated following standard definitions (Trinh et al., 2012).

Digital images of central bread slices (3 replicates per condition) were acquired with a smartphone camera using the same imaging setup (camera, distance, and magnification) for all samples. Images were processed using ImageJ software (Ferreira and Rasband, 2012). Morphometric parameters were analyzed in pixel-based units for comparative evaluation across processing conditions. Lighting conditions were kept constant for all image acquisitions.

After conversion to 8-bit and binary thresholding, alveoli were quantified using the "Analyze Particles" function. Output parameters included alveoli count, total alveolar area, and mean alveoli area.

2.6. Data analysis

Data were analyzed by two-way ANOVA (factors: pressure and temperature). Each dough was treated as an experimental unit. Significance threshold was $p \leq 0.05$. Post hoc Tukey HSD tests were performed when appropriate. Statistics were performed in R (v.4.3.3).

In addition to p-values, partial eta squared (η^2_p) was computed as a measure of standardized effect size for each factor and for the interaction term. This metric quantifies the relative amount of variance explained by a factor (0 = no effect; 1 = full determination), thus allowing direct comparison of factor importance across different response variables. Effect sizes were visualised using heatmaps. All analyses were performed in R (v.4.3.3).

3. Results and discussion

3.1. Rheology of refined flour dough (W260) and Verna

In the strong refined flour (W260), mixing temperature was the primary determinant of alveograph performance (Table 1). Low-temperature mixing (5 °C) increased dough extensibility (L) and swelling index (G) and reduced P/L, indicating a more deformable gluten network, consistent with the temperature-dependent tightening of gluten previously reported (Cappelli et al., 2020). Vacuum mixing produced a selective strengthening effect, as deformation energy (W) increased significantly under vacuum while most other parameters remained unchanged.

A plausible explanation is that reduced pressure limits air incorporation and promotes air removal during mixing, resulting in a more compact dough with fewer and smaller gas inclusions. Lower gas entrainment increases the effective contact area between added water and flour particles and reduces interfacial discontinuities, which can facilitate hydration of proteins and starch and enhance water-solid interactions. At the same time, a more continuous and compact gluten phase may develop because the protein network is less disrupted by bubbles and can undergo more effective aggregation during mixing (e.g., via non-covalent interactions and disulfide interchange), leading to a denser gluten matrix. These microstructural effects under vacuum have been reported to strengthen gluten-based systems in related cereal products (e.g., noodle doughs) (Liu et al., 2015). Importantly, no significant temperature $\times$ pressure interaction was found, indicating that the effects of mixing temperature and vacuum acted largely additively rather than synergistically in this strong-gluten system.

In the ancient flour Verna, higher mixing temperature (25 °C) significantly increased both dough tenacity (P) and deformation energy (W) (Table 1), despite the overall weak gluten potential of this flour. This behaviour differs from that observed in the strong refined flour and reflects a different underlying mechanism. In weak-gluten systems,

Table 1

Alveograph test results for W260 flour and Verna flour as a function of pressure temperature, and their interaction. Values are expressed as the mean of three replicates $\pm$ SD. ** ($p < 0.01$); * ($p < 0.05$); ns (not significant).

W260 Flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
P	79.00 $\pm$ 7.87	75.00 $\pm$ 5.44	ns	74.50 $\pm$ 5.72	79.50 $\pm$ 7.13	ns	ns
L	73.67 $\pm$ 11.07	67.83 $\pm$ 5.88	ns	77.50 $\pm$ 7.01	64.00 $\pm$ 4.38	**	ns
G	19.05 $\pm$ 1.46	18.30 $\pm$ 0.79	ns	19.58 $\pm$ 0.90	17.78 $\pm$ 0.59	**	ns
W	217.33 $\pm$ 6.25	197.17 $\pm$ 13.18	**	209.00 $\pm$ 13.83	205.50 $\pm$ 16.07	ns	ns
P/L	1.11 $\pm$ 0.29	1.11 $\pm$ 0.14	ns	0.97 $\pm$ 0.15	1.26 $\pm$ 0.18	*	ns
Verna flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
P	38.50 $\pm$ 12.91	42.00 $\pm$ 9.59	ns	32.50 $\pm$ 7.18	48.00 $\pm$ 8.39	*	ns
L	43.67 $\pm$ 8.62	42.83 $\pm$ 3.66	ns	47.67 $\pm$ 5.65	38.83 $\pm$ 3.06	**	ns
G	14.63 $\pm$ 1.42	14.57 $\pm$ 0.63	ns	15.35 $\pm$ 0.88	13.85 $\pm$ 0.54	**	ns
W	56.50 $\pm$ 15.49	63.67 $\pm$ 13.74	ns	50.33 $\pm$ 9.71	69.83 $\pm$ 11.70	*	ns

increasing temperature can accelerate protein hydration and enhance molecular mobility, promoting faster aggregation of gluten proteins during mixing. However, this aggregation does not translate into a more extensible or well-organised gluten network. Instead, it results in a stiffer and less deformable structure, characterised by higher resistance to deformation (P) and increased energy required for bubble expansion (W).

Therefore, in Verna doughs, the temperature-induced increase in P and W should not be interpreted as an improvement of gluten quality, but rather as a shift toward a more rigid and brittle network, consistent with the observed reduction in extensibility (L) and increase in P/L at higher temperature. This confirms that, in weak-gluten flours, temperature primarily modulates dough stiffness rather than functional gluten development.

The elasticity index (Ie), although measured (Section 2.5), was not significantly affected by either mixing temperature or pressure in Verna doughs ($p > 0.05$) and therefore did not contribute to the interpretation of rheological differences among treatments.

3.2. Bread quality

For W260, vacuum mixing negatively impacted bread development, as shown in Table 2: loaf height decreased under vacuum, despite the higher deformation energy observed in the dough. This apparent paradox is mechanistically coherent: vacuum removes entrapped air nuclei during mixing, leading to fewer sites for CO₂ expansion and thus restricting oven spring (Gao et al., 2017). Temperature also modulated product quality: low mixing temperature generated slightly more extensible doughs, which translated into improved loaf expansion. In Verna, bread quality parameters were globally lower than W260 and largely insensitive to process manipulations. The intrinsic weakness of the gluten network appeared to cap performance, leading to small and statistically unstable shifts in loaf metrics. This finding reinforces the

concept that process levers are constrained by genotype: flours with low gluten quality potential provide limited technological headroom for improvement.

3.3. Crumb structure (image analysis)

In W260, crumb structure metrics (total alveolar area) were remarkably stable across treatments, no significant effects were detected (Table 3; all $p > 0.05$). Neither temperature nor pressure produced meaningful changes. This robustness further confirms the buffering capacity of strong gluten doughs: once a coherent gas cell network is established, moderate shifts in process conditions are insufficient to perturb bubble architecture. In Verna, however, temperature and pressure interacted, producing measurable shifts in alveolar area, as reported in Table 3. This means that, in weak-gluten systems, bubble architecture is more sensitive to upstream conditions, even when macroscopic dough rheology appears inert. Mechanically, this suggests a structural uncoupling: rheology is dominated by gluten polymer characteristics, whereas crumb structure is co-determined by bubble nucleation and stabilisation. This supports the view that platform variables (temperature, pressure) propagate differently across structural scales.

The ridgeline plot, reported in Fig. 1, has been used because blister size distribution carries more physical information than a simple mean: it shows not only how large the blisters are, but how the whole population of blister sizes shifts under different mixing conditions.

As shown in Fig. 1, in Verna doughs vacuum mixing causes a clear rightward shift of the average blister size distribution toward larger values at both temperatures, with broader distributions and the emergence of large-size populations, particularly at 5 °C. This pattern indicates that, when the gluten network is intrinsically weak, the crust regime becomes increasingly bubble-driven, and bubble growth and coalescence are poorly constrained. In contrast, W260 exhibits compact

Table 2

Qualitative characteristics of bread made with W260 flour and Verna as a function of pressure and temperature.

Results are expressed as the mean of three replicates $\pm$ SD ** ($p < 0.01$); * ($p < 0.05$); ns (not significant).

W260 Flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
Weight(g)	343.81 $\pm$ 7.46	347.99 $\pm$ 5.13	ns	347.20 $\pm$ 5.46	344.60 $\pm$ 7.66	ns	ns
Volume (dm ³)	0.68 $\pm$ 0.04	0.72 $\pm$ 0.06	ns	0.71 $\pm$ 0.04	0.68 $\pm$ 0.07	ns	ns
Height (cm)	5.43 $\pm$ 0.37	6.27 $\pm$ 0.39	**	5.72 $\pm$ 0.61	5.98 $\pm$ 0.54	ns	ns
Verna Flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
Weight(g)	355.12 $\pm$ 4.19	355.84 $\pm$ 6.25	ns	357.39 $\pm$ 3.59	353.56 $\pm$ 5.93	ns	ns
Volume (dm ³)	0.65 $\pm$ 0.04	0.68 $\pm$ 0.05	ns	0.67 $\pm$ 0.05	0.66 $\pm$ 0.04	ns	ns
Height (cm)	5.52 $\pm$ 0.50	5.67 $\pm$ 0.19	ns	5.47 $\pm$ 0.34	5.72 $\pm$ 0.39	ns	ns

Table 3
Image analysis results on bread slices made with type 0 W260 flour and Verna as a function of pressure and temperature. Results are expressed as the mean of three replicates $\pm$ SD, ** ($p < 0.01$); * ($p < 0.05$); ns (not significant).

W260 Flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
Number of alveoli. alveoli	$5.4 \times 10^3 \pm 0.6 \times 10^3$	$5.5 \times 10^3 \pm 0.4 \times 10^3$	ns	$5.3 \times 10^3 \pm 0.3 \times 10^3$	$5.6 \times 10^3 \pm 0.6 \times 10^3$	ns	ns
Total area ea totale	$3.6 \times 10^6 \pm 0.7 \times 10^6$	$3.4 \times 10^6 \pm 0.4 \times 10^6$	ns	$3.2 \times 10^6 \pm 0.4 \times 10^6$	$3.7 \times 10^6 \pm 0.7 \times 10^6$	ns	ns
Average alveolar size	$6.5 \times 10^5 \pm 0.7 \times 10^5$	$6.12 \times 10^5 \pm 0.7 \times 10^5$	ns	$6.02 \times 10^5 \pm 0.5 \times 10^5$	$6.7 \times 10^5 \pm 0.8 \times 10^5$	ns	ns
Verna Flour	Pressure (bar)		<i>p</i>	Temperature (°C)		<i>p</i>	TxP interaction
	Vacuum	Control		5	25		
Number of alveoli. alveoli	$5.8 \times 10^3 \pm 3.8 \times 10^3$	$5.9 \times 10^3 \pm 9.9 \times 10^2$	ns	$6.8 \times 10^3 \pm 3.2 \times 10^3$	$4.9 \times 10^3 \pm 1.9 \times 10^3$	ns	ns
Total area ea totale	$3.2 \times 10^6 \pm 1.7 \times 10^6$ a	$1.4 \times 10^6 \pm 2.7 \times 10^5$ b	***	$3.0 \times 10^6 \pm 1.8 \times 10^6$ x	$1.5 \times 10^6 \pm 3.9 \times 10^5$ y	***	***
Average alveolar size	$6.1 \times 10^5 \pm 2.9 \times 10^5$	$2.3 \times 10^5 \pm 0.4 \times 10^4$	ns	$4.8 \times 10^5 \pm 3.5 \times 10^5$	$3.6 \times 10^5 \pm 1.8 \times 10^5$	ns	ns

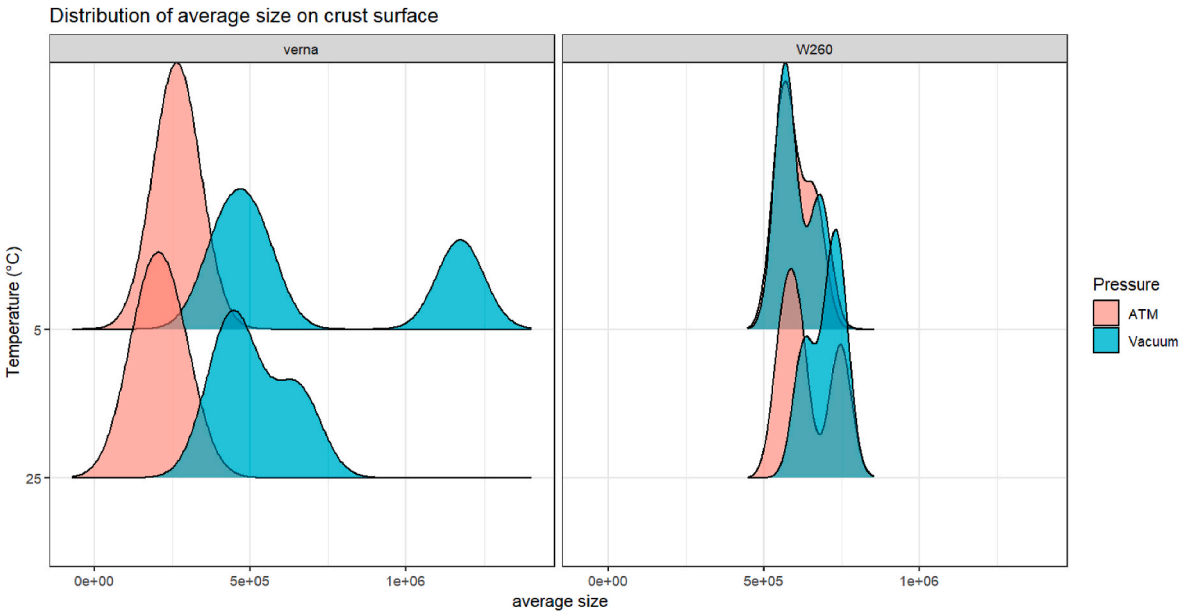


Fig. 1. Ridgeline plots of blister average size for each flour and mixing condition. Distributions were extracted from crust image analysis and represent the full population of blister diameters per loaf.

and largely overlapping distributions across processing conditions, confirming that a stronger gluten backbone limits blister expansion and suppresses pressure- and temperature-induced morphological divergence.

The boxplot on % surface blister coverage, reported in Fig. 2, has been used as a complementary and more intuitive morphological endpoint: this metric quantifies how much of the crust is actually occupied by blisters (i.e., how “bubble-dominated” the crust is). In Verna doughs, blister coverage increased markedly under vacuum mixing and at low temperature (Fig. 2), consistent with the ridgeline evidence of larger and more dominant surface blisters and indicating that weak gluten networks fail to effectively limit surface instability. Under vacuum conditions, blister coverage remained relatively high across temperatures, suggesting reduced temperature sensitivity and a shift toward a pressure-dominated regime of bubble formation. In contrast, under atmospheric pressure, low temperature exerted a stronger effect, highlighting the role of dough viscosity and gas diffusion when air entrainment is not constrained (see Fig. 3).

In W260 doughs, blister coverage remained low and showed limited sensitivity to both pressure and temperature, confirming that strong gluten systems confer a resilient crust micro-architecture that is largely process-insensitive. Overall, the % surface coverage metric translates the ridgeline distribution shifts into a clear structural outcome: weak flours

exhibit high blister dominance, whereas strong flours maintain low blister dominance regardless of processing conditions.

3.4. Texture profile analysis (TPA)

In breads made from refined flour (W260), TPA was more responsive to process variables. Hardness (first bite) and gumminess were significantly increased by both vacuum and high temperature. Secondary hardness, which refers to the hardness value obtained from the second compression cycle of the TPA test, was significantly increased by vacuum, confirming a coherent mechanical response across bites. Chewiness was significantly increased by temperature.

This demonstrates that, for strong flours, vacuum and temperature act as texturising levers: vacuum tends to compact crumb structure (more hardness and gumminess), whereas higher dough temperature tends to generate a firmer and more “chewy” bite profile.

Because multiple TPA endpoints co-vary and operate on different units (N, ratios, derived products), an isolated mean comparison is insufficient to understand which factor actually drives mechanical behaviour. For this reason, we used partial η^2 as a standardized metric and visualised factor importance in a heatmap, allowing direct comparison of how temperature, vacuum and their interaction contribute to textural variance across flours.

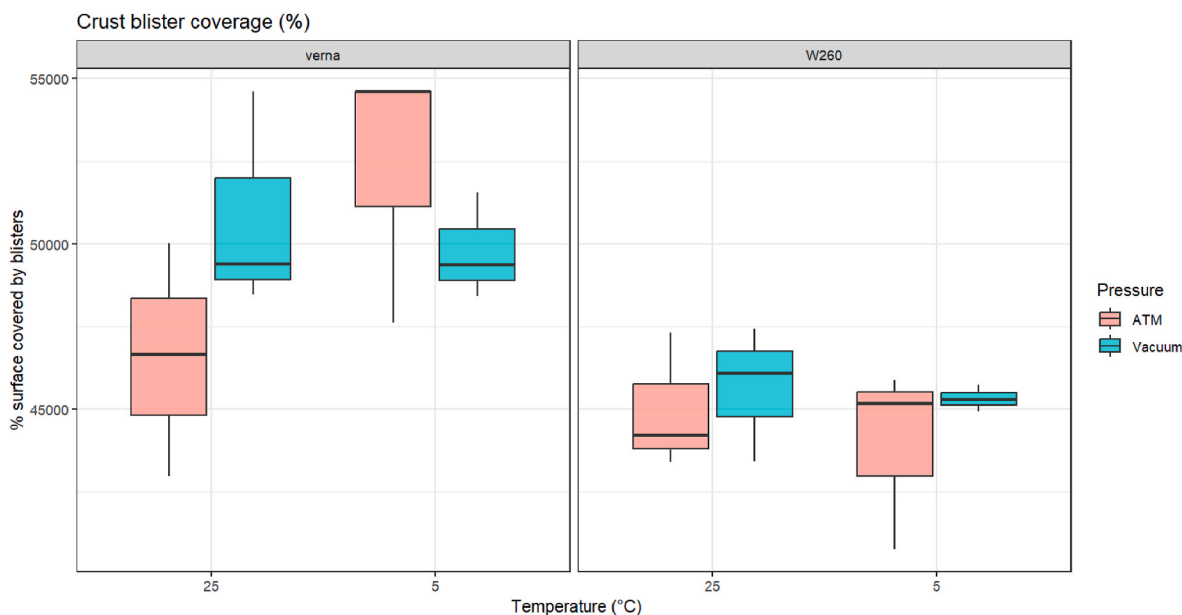


Fig. 2. Boxplots of the percentage of crust surface covered by blisters. Values represent the proportion of surface area segmented as blister.

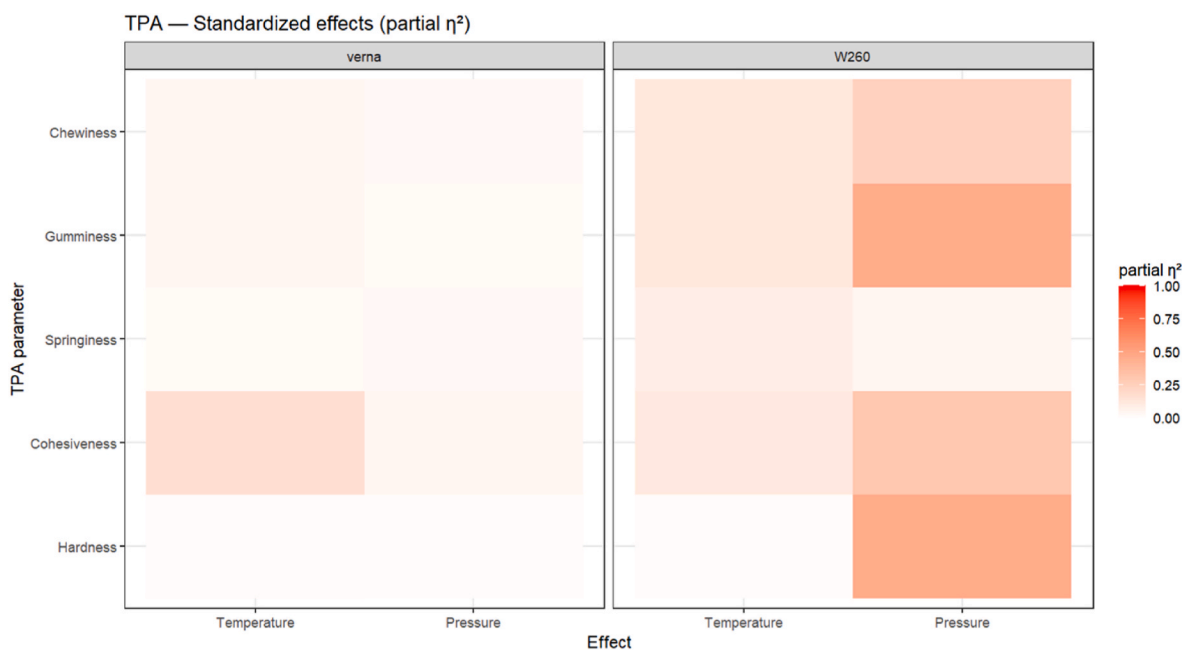


Fig. 3. Partial η^2 heatmap for TPA parameters, showing the proportion of variance explained by mixing temperature, pressure and their interaction, separately for each flour.

In the refined flour (W260) the dominant factor shaping texture was pressure (vacuum). Partial η^2 values indicate large effects of pressure particularly on gumminess, chewiness and hardness. This means that when a strong gluten system is available, vacuum mechanically changes how the gluten/crumb network behaves under deformation: i.e. the matrix becomes texturally stiffer and more energy-demanding to compress.

Temperature was much less important in the W260 TPA response, suggesting that the thermal sensitivity of gluten which was very clear in the alveograph is not equivalently visible at TPA scale. In other words: in a strong flour, process pressure affects final mastication mechanics more than mixing temperature.

Conversely, in Verna the Figure is almost flat. Partial η^2 values are

near zero. Neither temperature nor pressure produces meaningful variance in TPA parameters. This is a key mechanistic finding:

In systems with high gluten potential (W260), process conditions act as true mechanical levers: pressure explains a large share of variance in textural endpoints, indicating that the emerging crumb network remains physically modifiable by the mixing environment. By contrast, in the low-W ancient flour (Verna), process factors lose explanatory power entirely: the system stays locked in a genotype-bounded regime in which no realistic combination of temperature or pressure can meaningfully alter final mechanical properties.

This dissociation between rheological scale (alveograph) and final mastication mechanics (TPA) is mechanistically consistent with a hierarchical gluten model: the alveograph is sensitive to early network

formation kinetics, whereas TPA captures the emergent macro-network once starch gelatinisation and gas expansion have already locked the structure. Therefore, the strong pressure sensitivity observed in W260 at TPA, but not equivalently in Verna, confirms that process levers only propagate through the system if a sufficiently extensible gluten backbone exists. In weak ancestral flour, the system collapses to genotype dominance: the network never reaches a state in which physical processing can produce displacements large enough to be measurable at TPA scale.

Consequently, loaf-level performance mirrors this mechanistic split: in W260, vacuum depresses loaf height because the network is sufficiently developable that vacuum-driven degassing reduces the number of nucleation sites that later seed expansion; in Verna, loaf height remains intrinsically constrained, not because process is ineffective per se, but because the network never reaches a structural regime where gas cell topology can be process-modulated.

3.5. Operation design rule

Our data allow to propose a simple but operational design rule. The ability of pressure to engineer gluten mechanics is not continuous: it is threshold-based. Below a minimum alveograph strength, the system is genotype-bounded and process factors do not propagate to the final mechanics. Above that minimum, the system becomes process-responsive. In practical terms, a W alveograph $\geq \sim 220$ –240 appears to be a plausible lower bound for pressure responsiveness. Below this threshold, vacuum mixing simply cannot impose sufficient mechanical leverage because the elastic backbone of the gluten network is not yet strong enough to sustain energy transfer or to stabilize the microstructure during gas nucleation. Above this threshold, pressure becomes a “true lever”, but with a clear side effect: while gluten becomes mechanically stronger, gas nucleation is suppressed, with negative consequences on loaf volume. This is engineering-relevant: pressure is a selective lever — productive only in flours that already display high inherent strength, and potentially detrimental for volume if not compensated by a gas management strategy. This threshold-based view turns the present findings into a predictive design principle for plant-level decision making: select flours above the W threshold if vacuum is intended as an engineering lever; avoid vacuum in sub-threshold flours.*se genotype-specific strategies and treat “vacuum strengthening” not as a universal solution but as a context-limited tool.

4. Conclusions

Low-temperature mixing emerged as a robust, genotype-independent lever to improve gluten performance. In contrast, vacuum acted as a conditional lever: it strengthened dough only when gluten potential was already high, and this strengthening penalised loaf development by reducing gas nuclei. In weak ancient flour, process variables provided little mechanical leverage: performance was primarily genotype-bounded. These findings resolve an open ambiguity in the literature: the efficacy of vacuum is not flour-agnostic. Process optimization in wholemeal and ancient systems must therefore be genotype-specific, and “vacuum strengthening” should not be treated as a universal intervention. Future work should quantify the gluten threshold above which vacuum becomes beneficial — enabling predictive, not empirical, process design.

CRediT authorship contribution statement

G. Angeloni: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **A. Spadi:** Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation. **A. Cappelli:** Resources, Methodology, Funding

acquisition. **F. Corti:** Investigation, Data curation. **C. Vizzo:** Investigation, Data curation. **L. Bruchi:** Investigation, Data curation. **A. Parenti:** Visualization, Resources, Funding acquisition. **P. Masella:** Writing – review & editing, Visualization, Validation, Supervision, Resources, Methodology, Formal analysis, Conceptualization.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

Data will be made available on request.

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