

Rancidity in pearl millet (*Pennisetum glaucum*): biochemical mechanisms, genetic variability and mitigation strategies

Meghana Singh Rajotia

Department of Genetics and Plant Breeding, College of Agriculture, CCS HAU, Hisar, Haryana

*Corresponding Author's Email: meghanarajotia@hau.ac.in

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ABSTRACT

Pearlmillet is a nutritionally rich and climate-resilient cereal crop widely cultivated in arid and semi-arid regions. However, its utilization in processed food systems is limited by the rapid development of rancidity in the flour after milling. Rancidity in pearl millet flour is predominantly enzymatic, initiated by endogenous triacylglycerol lipases that release free fatty acids, followed by oxidative reactions mediated by lipoxygenase and non-enzymatic pathways, resulting in the formation of volatile compounds responsible for development of off-flavour during storage. Although numerous studies have independently investigated biochemical indicators, genetic variability and processing-based mitigation strategies, the available information remains fragmented and lacks an integrated synthesis linking enzymatic mechanisms with genetic regulation and technological interventions. Recent advances in metabolomics, transcriptomics and functional genomics have improved understanding of lipid catabolism and flavour deterioration in pearl millet flour. However, these findings have not yet been comprehensively evaluated in the context of breeding and industrial processing. Integration of biochemical understanding, genetic improvement and optimized post-harvest processing strategies is essential for enhancing flour shelf stability and expanding the utilization of pearl millet in modern food systems.

Keywords: Lipase activity, lipid oxidation, pearl millet flour shelf life, post-harvest processing, rancidity

INTRODUCTION

Pearlmillet (*Pennisetum glaucum* (L.) R. Br.) is a major cereal crop grown predominantly in the arid and semi-arid regions of Asia and Africa, where it plays a crucial role in sustaining food and nutritional security. The crop is well known for its tolerance to drought, high temperature and poor soil fertility, making it particularly important under changing climatic conditions (Saleh *et al.*, 2013). In addition to its agronomic resilience, pearl millet possesses superior nutritional attributes, including higher lipid content, balanced amino acid composition and greater mineral density compared with several major cereals (Chandrasekara and Shahidi, 2012). Despite these advantages, pearl millet remains underutilized in

processed food systems largely due to the rapid development of rancidity in flour after milling. During storage, pearl millet flour quickly develops undesirable odour and bitterness, resulting in reduced consumer acceptability and limited commercial utilization. Short shelf life is a major bottleneck in the commercialization of pearl millet-based products, particularly the flour-based foods requiring extended storage (Taylor and Duodu, 2017). Rancidity in pearl millet flour is closely associated with lipid degradation processes. Pearlmillet grains contain relatively high lipid concentrations (approximately 5-7.5%), most of which are localized in the germ together with lipid-degrading enzymes (Lai and Varriano-Marston, 1980). Disruption of cellular integrity during milling brings these endogenous lipases

into direct contact with triglycerides, resulting in rapid hydrolysis and accumulation of free fatty acids (Kaced *et al.*, 1984). These free fatty acids not only contribute directly to unpleasant flavours but also act as substrates for oxidative reactions. Oxidative deterioration further accelerates quality loss in pearl millet flour. Lipoxygenase-mediated oxidation and non-enzymatic autoxidation of unsaturated fatty acids lead to the formation of hydroperoxides, which subsequently decompose into volatile aldehydes, ketones and alcohols responsible for off-flavour development (Chandrasekara and Shahidi, 2012). Studies have consistently shown that hydrolytic rancidity precedes oxidative rancidity in pearl millet flour, highlighting the central role of lipase activity in initiating deterioration. Several investigations have demonstrated substantial genotypic variation for rancidity-related traits in pearl millet. Differences among genotypes in acid value, peroxide value and the rate of flavour deterioration during storage suggest that susceptibility to rancidity is at least partly under genetic control. Identification of low-rancidity genotypes provides opportunities for genetic improvement of flour shelf life through conventional breeding and molecular approaches.

Alongside genetic strategies, various processing and storage interventions have been explored to mitigate rancidity in pearl millet flour. Thermal treatments such as dry heating, hot-water blanching and microwave processing have been shown to significantly reduce lipase activity and slow the accumulation of free fatty acids during storage (Kadlag *et al.*, 1995). More recent studies have reported the effectiveness of extrusion cooking and advanced packaging systems in extending flour shelf life without major losses in nutritional quality. Recent advances in metabolomics and transcriptomics have further improved understanding of the molecular basis of rancidity in pearl millet. These studies have identified lipid-derived metabolites and upregulation of lipase and oxidation-related genes as key determinants of flour deterioration, offering new avenues for predictive screening and targeted improvement (Kumar *et al.*, 2025; Yogendra *et al.*, 2024; Aher *et al.*, 2022). Thus, understanding the biochemical mechanisms underlying rancidity, the genetic

variation influencing shelf-life traits and the effectiveness of technological mitigation strategies is essential for improving the storage stability and commercial utilization of pearl millet flour. Thus, in view these backgrounds an attempt was made to bridge the research gaps that helps in improving the shelf life and also opens up new vistas for pearl millet flour utilization for food industry.

Biochemical mechanisms underlying rancidity in pearl millet flour

Rancidity in pearl millet flour arises primarily from lipid degradation processes that occur rapidly after milling, owing to the disruption of cellular compartmentalization and exposure of lipids to endogenous enzymes. Compared with most cereals, pearl millet contains a relatively high lipid fraction (approximately 5-7.5%), with a substantial proportion localized in the germ along with lipid-degrading enzymes, making the flour particularly susceptible to deterioration during storage (Lai and Varriano-Marston, 1980; Chandrasekara and Shahidi, 2012). The germ of sorghum and pearl millet contains a disproportionately high concentration of lipids and associated enzymes, making these cereals particularly susceptible to lipid hydrolysis once milling disrupts cellular compartments (Hoseney *et al.*, 1987).

Hydrolytic rancidity and role of lipases

The initial and most critical step in rancidity development is hydrolytic lipid breakdown catalysed by endogenous triacylglycerol lipases. Endogenous lipases catalyse the breakdown of triacylglycerols into free fatty acids and glycerol immediately after milling, leading to a rapid increase in acid value during early storage (Kaced *et al.*, 1984). Accumulation of FFAs directly contributes to undesirable taste and odour and accelerates subsequent oxidative reactions. Several studies have demonstrated that increases in acid value precede changes in peroxide value, confirming that lipid hydrolysis is the primary initiating event in pearl millet flour rancidity (Mazumdar *et al.* 2016; Sharma *et al.*, 2020).

Recent molecular studies have strengthened this biochemical evidence by identifying specific triacylglycerol lipase genes associated with flour rancidity. Loss-of-function alleles of these lipases

have been linked with reduced accumulation of FFAs and delayed flavour deterioration, providing direct genetic evidence for the central role of lipase-mediated hydrolysis in rancidity initiation (Aher *et al.*, 2022).

Oxidative rancidity and lipoxygenase-mediated reactions

Following lipid hydrolysis, oxidative rancidity becomes the dominant process driving flavour deterioration. Free unsaturated fatty acids generated during lipid hydrolysis subsequently act as substrates for lipoxygenase-mediated oxidation, which catalyses the incorporation of molecular oxygen to form fatty acid hydroperoxides. These primary oxidation products are unstable and readily decompose into secondary volatile compounds such as aldehydes, ketones and alcohols that impart characteristic off-flavours to stored

flour (Chandrasekara and Shahidi, 2012).

Lipoxygenase activity in pearl millet flour has been shown to correlate positively with peroxide value and sensory perception of rancidity during storage. In addition to enzymatic oxidation, non-enzymatic autoxidation of unsaturated fatty acids further contributes to peroxide formation, particularly under conditions of elevated temperature, moisture and oxygen availability. Together, enzymatic and non-enzymatic oxidation processes amplify flavour deterioration once hydrolytic rancidity has been initiated.

Formation of volatile compounds and flavour deterioration

The breakdown of hydroperoxides results in the formation of low-molecular-weight volatile compounds, including hexanal, nonanal, 2-pentanone and related aldehydes and ketones, which are primarily responsible for the characteristic rancid odour of pearl millet flour (Yogendra *et al.*, 2024; Kumar *et al.*, 2025). Recent volatome and metabolomic studies using GC-MS and LC-HRMS have consistently identified lipid-derived aldehydes and alcohols as key markers associated with advanced stages of rancidity (Yogendra *et al.*, 2024; Kumar *et al.*, 2025).

These studies further indicate that the rate and composition of volatile formation vary among genotypes, supporting the hypothesis that biochemical pathways underlying rancidity are modulated by genetic factors. Importantly, metabolomic evidence confirms that lipid catabolism, rather than carbohydrate or protein degradation, dominates the biochemical landscape of flavour deterioration in pearl millet flour during storage (Kumar *et al.*, 2025).

Interaction between hydrolytic and oxidative pathways

Hydrolytic and oxidative rancidity are not independent processes but are tightly interconnected. Lipase-mediated release of free fatty acids increases substrate availability for lipoxygenase and accelerates oxidative reactions, creating a self-reinforcing cycle of lipid degradation. Consequently, interventions that reduce lipase activity at early stages either through genetic selection or processing treatments are more effective

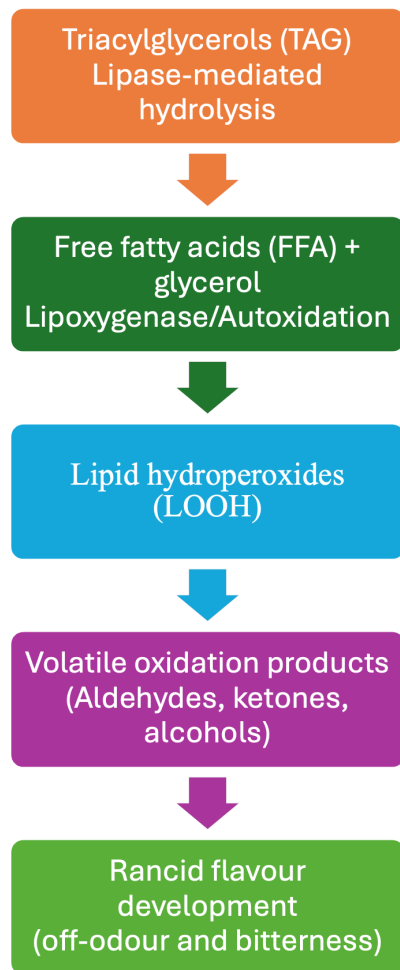


Fig. 1. Biochemical mechanism of rancidity in pearl millet

tive in delaying overall rancidity development than strategies targeting oxidation alone (Kadlag *et al.*, 1995; Yadav *et al.*, 2012).

Understanding this biochemical cascade highlights the importance of targeting the initial hydrolytic step to achieve durable improvements in flour shelf life. The mechanistic linkage between lipase activity, lipid oxidation and volatile formation provides a rational framework for integrating biochemical screening, genetic improvement and processing-based mitigation strategies.

Analytical indicators and methods for assessing rancidity in pearl millet flour

Reliable assessment of rancidity is essential for understanding deterioration mechanisms, comparing genotypes, evaluating processing interventions and predicting the shelf life of pearl millet flour. Rancidity in pearl millet is a complex phenomenon involving hydrolytic and oxidative lipid degradation, and its assessment therefore requires the use of multiple complementary analytical approaches. Chemical indices, enzyme activity measurements, volatile profiling and sensory evaluation have all been applied to characterize different stages of rancidity development in pearl millet flour.

Acid value and free fatty acid content

Acid value (AV), commonly expressed as free fatty acid (FFA) content, is the most widely used indicator of hydrolytic rancidity in pearl millet flour. It reflects the extent of triacylglycerol hydrolysis catalysed by endogenous lipases released during milling and typically increases rapidly during the early stages of storage. Classic studies on pearl millet have demonstrated that disruption of cellular compartmentalization during milling results in rapid accumulation of free fatty acids, making acid value a sensitive early marker of flour deterioration (Lai and Varriano-Marston 1980; Kaced *et al.*, 1984).

In pearl millet, acid value has been effectively used to monitor the progression of rancidity during storage and to differentiate samples with contrasting rates of lipid hydrolysis. However, acid value alone does not adequately reflect oxidative deterioration or flavour intensity at later stages of storage, highlighting the need to interpret this

parameter alongside oxidative and sensory indicators.

Peroxide value and oxidative indices

Peroxide value (PV) is commonly employed to assess the formation of lipid hydroperoxides during the initial stages of oxidative rancidity in pearl millet flour. Following the onset of hydrolytic rancidity, unsaturated fatty acids released from triacylglycerols undergo oxidation, leading to an increase in PV. Studies on pearl millet have shown that PV generally rises after increases in acid value and may subsequently decline as hydroperoxides decompose into secondary volatile compounds responsible for off-flavours (Lai and Varriano-Marston, 1980).

Although PV provides useful information on oxidative progression, its non-linear behaviour during storage limits its usefulness as a sole indicator of advanced rancidity. Therefore, PV is most informative when combined with hydrolytic indices and volatile analysis in pearl millet flour.

Experimental determination of acid value and peroxide value

Acid value and peroxide value of pearl millet flour are commonly determined using standard titrimetric procedures recommended by the AOAC (AOAC1990). These parameters serve as reliable indicators of hydrolytic and oxidative rancidity during storage. Acid value reflects the extent of lipid hydrolysis caused by endogenous lipases, while peroxide value indicates the formation of primary oxidation products such as lipid hydroperoxides.

For determination of acid value, freshly milled flour is extracted with hot neutralized ethanol and titrated against standardized sodium hydroxide solution using phenolphthalein indicator. The appearance of a persistent pink colour indicates the endpoint of titration. Acid value is expressed as the milligrams of NaOH required to neutralize the free fatty acids present in one gram of sample.

$$CAV = 40 \times A \times N / W$$

where

A = volume of NaOH used (ml)

N = normality of NaOH solution

W = weight of the sample (g)

Peroxide value is determined using the iodometric titration method. Flour samples are treated with an acetic acid–chloroform solvent mixture, followed by addition of potassium iodide which reacts with lipid peroxides to liberate iodine. The liberated iodine is titrated with standardized sodium thiosulphate solution using starch indicator until the disappearance of the blue-black colour.

$$\text{CPV} = (A - B) \times N \times 1000 / W$$

where

A = volume of sodium thiosulphate used for sample (ml)

B = volume used for blank (ml)

N = normality of sodium thiosulphate

W = weight of sample (g)

These indices are widely used for evaluating rancidity progression in pearl millet flour and for identifying genotypes with improved flour storage stability (Mazumdar *et al.*, 2016, Sharma *et al.*, 2020). Studies have shown that increases in acid value generally precede increases in peroxide value during storage, indicating the sequential occurrence of hydrolytic followed by oxidative rancidity.

Enzyme activity assays

Direct measurement of lipase and lipoxygenase activities provides mechanistic insight into the biochemical drivers of rancidity in pearl millet flour. Early biochemical studies established that endogenous lipases present in the germ play a central role in initiating lipid hydrolysis immediately after milling (Kaced *et al.*, 1984). More recently, genetic evidence has confirmed the importance of lipase activity in determining flour rancidity behaviour in pearl millet. Loss-of-function alleles of triacylglycerol lipase genes have been associated with reduced free fatty acid accumulation and delayed flavour deterioration during storage (Aher *et al.*, 2022).

Measurement of lipoxygenase activity further contributes to understanding oxidative rancidity, as this enzyme catalyses the oxidation of unsaturated fatty acids released during hydrolysis. Although fewer studies have quantified lipoxygenase activity directly in pearl millet flour, available evidence suggests that enzymatic oxidation amplifies quality loss once hydrolytic ran-

idity has been initiated.

Volatile profiling and metabolomic approaches

Instrumental analysis of volatile compounds has emerged as a powerful tool for characterizing flavour deterioration in pearl millet flour. Gas chromatography-mass spectrometry (GC-MS) studies have consistently identified lipid-derived aldehydes, ketones and alcohols as major contributors to rancid odour during storage. Recent metabolomic investigations in pearl millet have confirmed that compounds such as hexanal and related aldehydes accumulate progressively with storage time and are closely associated with sensory rejection of flour (Yogendra *et al.*, 2024).

Non-targeted metabolomic approaches have further demonstrated that lipid catabolism dominates the biochemical landscape of deteriorating pearl millet flour. These studies have revealed genotype-specific metabolic signatures linked to differences in rancidity behaviour, highlighting the potential of metabolomics-based markers for predicting shelf life and supporting breeding for improved flour stability (Yogendra *et al.*, 2024).

Sensory evaluation and integrated assessment

Despite advances in chemical and instrumental analyses, sensory evaluation remains indispensable for assessing the practical significance of rancidity in pearl millet flour. Sensory attributes such as off-odour intensity, bitterness and overall acceptability ultimately determine consumer rejection and may not always correlate linearly with individual chemical indices. Studies on pearl millet have emphasized the importance of integrating sensory evaluation with biochemical measurements to obtain a realistic assessment of flour quality during storage (Lai and Varriano-Marston, 1980).

An integrated assessment approach combining acid value, peroxide value, enzyme activity measurements, volatile profiling and sensory evaluation is therefore recommended for comprehensive characterization of rancidity in pearl millet flour. Such multi-parameter frameworks improve the reliability of conclusions regarding genotype performance and the effectiveness of processing and storage interventions.

Genetic variation for rancidity-related traits and breeding implications in pearl millet

Genetic improvement represents a sustainable and long-term approach to addressing flour rancidity in pearl millet, particularly under storage conditions where post-harvest interventions are limited. The feasibility of such an approach depends on the presence of heritable variation in lipid metabolism and enzyme-mediated degradation processes that govern rancidity development.

Direct evidence for genetic control of rancidity in pearl millet has been provided by functional genomic studies identifying endogenous triacylglycerol lipases as key determinants of flour shelf life. Loss-of-function alleles in specific lipase genes significantly reduce free fatty acid accumulation and delay the development of off-flavours in flour, establishing rancidity as a genetically tractable trait (Aher *et al.*, 2022). This represents a major advance in linking post-milling quality directly to defined genetic factors.

The availability of a high-quality reference genome has substantially strengthened the prospects for incorporating such quality traits into pearl millet breeding programmes. Genome-scale resources facilitate the identification of candidate genes, development of molecular markers and application of marker-assisted and genomic selection strategies for complex traits (Varshney *et al.*, 2017).

Experience from pearl millet improvement programmes in India demonstrates that complex traits can be improved without compromising adaptation to harsh environments. Genetic improvement efforts have successfully combined yield enhancement, stress tolerance and grain quality within elite genetic backgrounds, providing confidence that rancidity-related traits can also be incorporated into breeding pipelines (Yadav and Rai, 2013).

The increasing adoption of genomic tools in pearl millet breeding further supports the feasibility of improving post-harvest quality traits. Genomic-assisted breeding approaches, initially developed for abiotic stress tolerance, provide transferable methodologies for incorporating rancidity-related traits once reliable phenotyping and marker-trait associations are established (Serba and Yadav, 2016).

Overall, recent advances in functional genomics and breeding frameworks provide a realistic pathway for improving flour shelf stability in pearl millet. Strategic integration of validated low-rancidity alleles with modern breeding approaches is expected to facilitate the development of cultivars with improved storage stability and enhanced utilization.

Processing and technological strategies for mitigating rancidity in pearl millet flour

Technological interventions play an important role in mitigating rancidity in pearl millet flour, particularly in situations where genetic solutions alone may not provide sufficient shelf-life extension. Processing-based approaches primarily aim to reduce the activity of lipid-degrading enzymes, limit exposure to oxygen and moisture or slow oxidative reactions during storage (Pathare *et al.*, 2025; Yarrakula *et al.*, 2024). Given the rapid onset of rancidity after milling, such interventions are especially relevant for enhancing the usability of pearl millet flour in traditional and commercial food systems. However, the adoption of such interventions in millet-based food systems is often constrained by cost, scale and infrastructural limitations, particularly in semi-arid regions (Taylor *et al.*, 2006). Major analytical indicators used for assessing rancidity in pearl millet flour are summarized in Table 1.

Thermal treatments for enzyme inactivation

Thermal processing has been widely investigated as an effective strategy to reduce lipase activity in pearl millet flour. Early studies demonstrated that heat treatment of pearl millet grains or flour prior to storage significantly reduced the rate of free fatty acid accumulation by partially inactivating endogenous lipases located in the germ (Kaced *et al.*, 1984). These findings established the principle that targeting enzyme activity at the initial stage can delay rancidity development.

Among thermal treatments, microwave processing has received particular attention due to its rapid heating and minimal impact on nutritional quality. Microwave-treated pearl millet flour has been shown to exhibit improved storage stability, with lower increases in acid value

and reduced development of off-odours during storage compared with untreated flour (Yadav *et al.*, 2012). Such treatments offer practical advantages for small-scale and decentralized processing systems.

Role of moisture control and storage environment

Moisture content and storage conditions strongly influence the rate of rancidity development in pearl millet flour. Increased moisture availability enhances enzyme activity and accelerates both hydrolytic and oxidative lipid degradation. Studies on pearl millet have demonstrated that maintaining low moisture content during storage slows free fatty acid accumulation and delays flavour deterioration (Lai and Varriano-Marston, 1980).

Temperature and oxygen availability also interact with moisture to influence flour stability. Storage under cool and dry conditions has been consistently associated with reduced rancidity development, highlighting the importance of appropriate storage practices alongside processing interventions. These findings emphasize that technological mitigation must be supported by proper post-processing handling and storage management.

Milling practices and fractionation effects

Milling practices influence the extent of rancidity development by determining the degree of contact between lipids and lipid-degrading enzymes. Fine milling increases surface area and promotes rapid interaction between enzymes and substrates, thereby accelerating rancidity. Conversely, partial decortication or controlled milling that reduces germ content has been reported to slow lipid degradation, although such approaches may also affect nutritional quality (Goswami *et al.*, 2024; Kaced *et al.*, 1984).

Optimizing milling conditions therefore rep-

resents a trade-off between flour stability and nutrient retention. Careful selection of milling intensity and particle size can contribute to improved shelf life without excessive loss of germ-associated nutrients.

Integration of processing with genetic strategies

While processing interventions can effectively delay rancidity, their impact is often temporary and dependent on storage conditions. Integration of technological approaches with genetic improvement offers a more durable solution. For example, flours derived from genotypes with inherently lower lipase activity exhibit improved storage stability even under minimal processing, suggesting synergistic benefits when low-rancidity genotypes are combined with mild thermal treatments (Aher *et al.*, 2022).

Such integrated strategies are particularly relevant for enhancing the commercial viability of pearl millet-based products, as they reduce reliance on intensive processing and extend shelf life under real-world storage conditions.

Practical implications for pearl millet value chains

From a practical perspective, simple and low-cost processing interventions such as microwave treatment, moisture control and optimized milling practices offer immediate opportunities to improve the storage stability of pearl millet flour. These approaches are particularly relevant in rural and semi-arid regions, where access to advanced storage infrastructure is limited and flour is often stored under ambient conditions. Adoption of such interventions can significantly reduce post-milling quality losses and enhance consumer acceptability of pearl millet-based products.

However, the effectiveness of processing-based mitigation strategies is influenced by storage environment and initial grain quality. Therefore, technological interventions should be viewed as complementary measures that enhance, rather

Table 1. Major indicators used for assessing rancidity in pearl millet flour

Indicator	Parameter measured	Stage of rancidity
Acid value	Free fatty acids	Hydrolytic rancidity
Peroxide value	Lipid hydroperoxides	Early oxidative rancidity
Lipase activity	Lipid hydrolysis enzyme	Initiation stage
Volatile compounds	Aldehydes and ketones	Advanced rancidity

than replace, intrinsic grain stability. Integrating appropriate processing practices within existing pearl millet value chains can improve flour shelf life and support wider utilization of this climate-resilient cereal in traditional and emerging food systems.

Role of lipase and lipoxygenase in rancidity development

Endogenous enzymes play a critical role in the development of rancidity in pearl millet flour. Among these enzymes, lipases and lipoxygenases are considered the primary drivers of lipid degradation during storage. Lipases catalyse the hydrolysis of triacylglycerols released during milling, resulting in rapid accumulation of free fatty acids (Kaced *et al.*, 1984; Kumar *et al.*, 2025). This enzymatic hydrolysis results in accumulation of free fatty acids that subsequently promote oxidative deterioration during storage.

Pearl millet contains relatively high lipid content (approximately 5-6%) and greater lipase activity compared with many other cereal grains (Lai and Varriano-Marston 1980; Chandrasekara and Shahidi, 2012). This combination accelerates lipid hydrolysis and contributes to the rapid deterioration of flour quality during storage (Selvan *et al.*, 2025). Following lipid hydrolysis, the released unsaturated fatty acids serve as substrates for lipoxygenase enzymes. Lipoxygenase catalyses the oxidation of these fatty acids to form hydroperoxides, which subsequently decompose into volatile aldehydes, ketones and alcohols responsible for the characteristic rancid odour of stored flour (Chandrasekara and Shahidi, 2012).

Recent molecular studies have further demonstrated that variations in lipase gene expression significantly influence rancidity behaviour in pearl millet genotypes, highlighting the potential of enzyme-related traits as targets for genetic improvement (Aher *et al.* 2022; Kumar *et al.*, 2025).

Metabolomic and lipidomic approaches for understanding rancidity

Recent advances in metabolomics and lipidomics have significantly improved understanding of rancidity mechanisms in pearl millet. These approaches enable comprehensive profiling of metabolites associated with lipid degrada-

tion and flavour deterioration during flour storage.

Metabolomic investigations have revealed the accumulation of lipid-derived compounds such as aldehydes, ketones and alcohols during storage, which are responsible for the development of undesirable flavours in pearl millet flour (Yogendra *et al.*, 2024; Kumar *et al.*, 2025). Comparative metabolomic studies have also demonstrated substantial differences among genotypes in metabolite accumulation patterns associated with rancidity development.

Lipidomic analyses further provide insights into the changes in fatty acid composition and lipid oxidation products during storage (Selvan *et al.*, 2025; Yogendra *et al.*, 2024). Such studies have confirmed that lipid catabolism is the dominant biochemical pathway underlying flour deterioration and have identified several metabolites that can serve as markers for predicting rancidity susceptibility.

Integration of metabolomic tools with conventional biochemical assays offers new opportunities for identifying low-rancidity germplasm and improving flour shelf life through breeding programmes.

Influence of storage conditions on rancidity development

Storage conditions play a significant role in determining the rate of rancidity development in pearl millet flour. Environmental factors such as temperature, moisture content and oxygen availability strongly influence enzymatic activity and lipid oxidation processes.

Higher storage temperatures and moisture levels enhance lipase and lipoxygenase activities, thereby accelerating lipid hydrolysis and oxidation reactions (Kaced *et al.*, 1984; Mazumdar *et al.* 2016). Conversely, maintaining low moisture content and storing flour under cool conditions can significantly slow rancidity development.

Studies have also demonstrated that storage duration directly affects rancidity indices such as free fatty acid content and peroxide value, with both parameters increasing progressively during storage (Sharma *et al.*, 2020).

Packaging conditions also influence flour stability (Taylor *et al.*, 2006). Storage in moisture-bar-

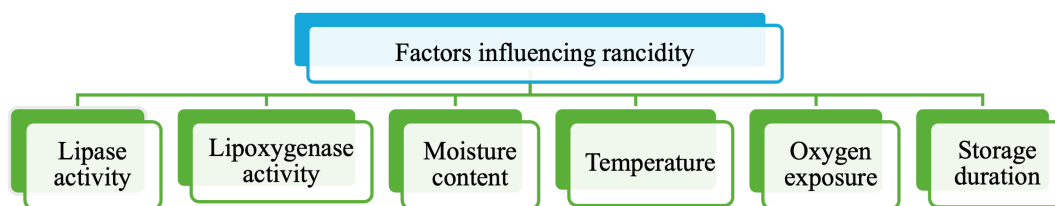


Fig. 2. Factors influencing rancidity in pearl millet

rier packaging materials or vacuum conditions can reduce oxygen exposure and delay oxidative reactions, thereby improving shelf life.

Genetic improvement strategies for reducing rancidity

Genetic improvement represents a sustainable approach to mitigating rancidity in pearl millet flour. Identification of genotypes with reduced lipase activity and improved lipid stability can significantly enhance flour shelf life without requiring extensive processing interventions.

Recent genomic and transcriptomic studies have identified several candidate genes associated with lipid degradation pathways in pearl millet. Functional variations in triacylglycerol lipase genes have been linked with reduced accumulation of free fatty acids and delayed rancidity development in flour (Reddy *et al.*, 2026; Aher *et al.*, 2022).

In addition, breeding programmes have reported significant genetic variation among pearl millet genotypes for rancidity-related traits such as acid value, peroxide value and enzyme activity (Mazumdar *et al.*, 2016; Sharma *et al.*, 2020).

Utilization of molecular markers and genomic selection approaches can therefore accelerate the development of pearl millet cultivars with improved flour storage stability.

Conclusions and future research directions

The rapid development of rancidity remains a major constraint limiting the wider utilization of pearl millet flour in processed food systems despite the crop's nutritional value and resilience to harsh environments. Rancidity in pearl millet

flour is largely driven by enzyme-mediated lipid degradation, in which hydrolytic processes initiate quality deterioration and oxidative reactions further amplify flavour loss during storage.

Advances in biochemical, analytical and molecular studies have improved understanding of the mechanisms underlying rancidity development. Functional genomic studies have demonstrated that variations in lipase-associated genes influence the accumulation of free fatty acids and subsequent flavour deterioration, indicating that flour stability can be targeted through genetic improvement. At the same time, emerging analytical tools provide improved opportunities for characterizing rancidity-related traits and supporting objective phenotyping.

Future progress will depend on integrating biochemical and molecular phenotyping with breeding programmes to accelerate genetic gains for flour shelf life. Development of high-throughput screening approaches for enzyme activity and lipid-derived metabolites will be essential for evaluating large germplasm collections and identifying genotypes with improved storage stability. In addition, genomic approaches such as association mapping and genomic selection can facilitate the incorporation of post-harvest quality traits alongside yield and stress tolerance.

Improving the utilization of pearl millet flour will also require alignment between breeding objectives, processing practices and value-chain requirements. Development of cultivars with reduced lipase activity, combined with optimized processing and storage practices, will contribute to improving flour stability and expanding the use of pearl millet in diverse food applications.

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