Journal of Experimental Botany, Vol. 76, No. 6 pp. 1482–1485, 2025
https://doi.org/10.1093/jxb/eraf060

eXtra Botany

Insight

Redox signalling in the nucleus: shaping the epigenetic code

Luisa Maria Sandalio*

Department of Stress, Development and Signalling in Plants, Estación Experimental del Zaidín, CSIC, Profesor Albareda 1, 18008, Granada, Spain

* Correspondence: luisamaria.sandalio@eez.csic.es

This article comments on:

De Smet B, Yang X, Plskova Z, Castell C, Fernández-Fernández A, Dard A, Masood J, Mhamdi A, Huang J, Vertommen D, Chan KX, Pyr dit Ruys S, Messens J, Kerchev PI, Van Breusegem F. 2025. The nuclear sulfenome of Arabidopsis: spotlight on histone acetyltransferase GCN5 regulation through functional thiols. Journal of Experimental Botany **76**, 1569–1584. https://doi.org/10.1093/jxb/erae514

Hydrogen peroxide (H_2O_2) promotes reversible oxidation of specific cysteines from proteins to sulfenic acid (RSOH), which is considered an important post-translational modification (PTM) of proteins known as sulfenylation. De Smet *et al.* (2025) explored the role of the nucleus in cellular redox homeostasis by analysing nuclear sulfenylated proteins (sulfenome). They identified the histone acetyltransferase GCN5 as a sulfenylated target and deciphered the functional importance of this redox modification of specific cysteine residues. Their findings highlight GCN5 as a key regulator at the intersection of ROS-dependent stress signalling and genetic reprograming.

Epigenetics and histone acetylation

Histone acetylation is an important process in the regulation of chromatin structure, and contributes to epigenetic regulation and gene reprogramming (Shen *et al.*, 2016; Jiang *et al*, 2020; Wu *et al.*, 2023). Epigenetics involves stable chromatin state changes during meiosis or mitosis which can alter gene expression and therefore phenotypic traits independent of DNA sequence changes (Shen *et al.*, 2016). In addition to histone acetylation, other histone modifications such as methylation, ADP-ribosylation, SUMOylation, and phosphorylation, as well as small or long non-coding RNAs, are epigenetic modifications (Shen *et al.*, 2016). The level of histone acetylation is regulated by the activities of histone acetyltransferases (HATs) and histone deacetylases (HDACs; Shen *et al.*, 2016). In general, histone acetylation has been associated with activation of gene expression, whereas histone deacetylation represses it (Jiang *et al.*, 2020; Wu *et al.*, 2023). Histone acetylation relaxes histone–DNA interactions by neutralizing the positive charge of histone tails, thus allowing transcription factors to access chromatin (Jiang *et al.*, 2020; Grasser *et al.*, 2021). This process also serves as a recognition motif for bromodomain-containing proteins, thus allowing the recruitment of downstream transcriptional activators and chromatin remodellers (Mutlu and Puigserver, 2021). GCN5 acetyltransferase mediates histone acetylation and transcriptional activation in eukaryotes, cooperating with HDACs to maintain endogenous histone acetylation homeostasis (Wu *et al.*, 2023). GCN5 is part of the transcriptional co-activator Spt–Ada–Gcn5 Acetyltransferase (SAGA) complex which is organized in four distinct modules: histone acetylation (HAT), deubiquitination (DUB), and a structural core including SPT and TAF (Grasser *et al.*, 2021; Wu *et al.*, 2021, 2023). In addition to the HAT domain, GCN5 contains a bromodomain that recognizes acetylated histone and thereby promotes the histone acetylation activity of SAGA (Wu *et al.*, 2023). Additionally, GCN5 makes up part of the plant-specific PAGA complex which, apart from GCN5, contains ADA2A and four subunits (SPC, ING1, SDRL, and EAF6; Wu *et al.*, 2023). PAGA and SAGA act independently in mediating moderate and high levels of histone acetylation, respectively, thereby promoting transcriptional activation (Wu *et al.*, 2023). PAGA and SAGA display an antagonistic effect, repressing or derepressing gene expression. SAGA is implicated in both developmental and stress programmes, while PAGA regulates plant morphology (Wu *et al.*, 2023).

Reactive oxygen species: oxidative damage versus signalling

Reactive oxygen species (ROS) are by-products of aerobic metabolism and comprise different reduced derivatives

of molecular oxygen, giving rise sequentially to superoxide ($O_2^{\cdot -}$), H_2O_2, and hydroxyl radicals (·OH) (Fig. 1A; Sies *et al.*, 2022). The chemical reactivity and biological functions of the different ROS differ and, according to their concentrations and chemical properties, they can participate in signalling processes, cellular damage, and cell death (Mittler *et al.*, 2022; Sies *et al.*, 2022). ROS production occurs in various cell compartments including mitochondria, chloroplasts, peroxisomes, the plasma membrane, the endoplasmic reticulum, and nuclei (Smirnoff and Arnaud, 2019; Fig. 1B), and their accumulation is controlled by enzymatic and non-enzymatic antioxidants. Under stress conditions, excessive ROS accumulation can overwhelm the antioxidant defences, causing oxidative damage to macromolecules, consequently disrupting metabolism and potentially leading to reduced growth or even plant death. However, H_2O_2 acts as secondary messenger through reversible oxidation of specific cysteines from proteins to sulfenic acid (RSOH), which is considered an important PTM known as sulfenylation, and subsequent redox relay mechanisms (Huang *et al.*, 2018; Smirnoff and Arnaud, 2019; Sies *et al*, 2022). Nowadays, one of the main challenges in redox research is the molecular identification of targets of redox signalling and the redox networks at the cellular, subcellular, and intercellular level, mainly under conditions of oxidative stress imposed by adverse conditions. Therefore, the development of a subcellular atlas of redox network responses as well as the analysis of redox dynamics and redox proteomes are required to understand the mechanisms involved in acclimation to changing environmental conditions. Sulfenylated proteins take part in these redox networks. This PTM is reversible and acts as a dynamic molecular switch that can modulate protein structure, function, stability, and location, thereby amplifying the complexity of the proteome, and enhancing the regulatory potential of cells (Huang *et al.*, 2018; Sandalio *et al.*, 2019; Muleya *et al.*, 2022).

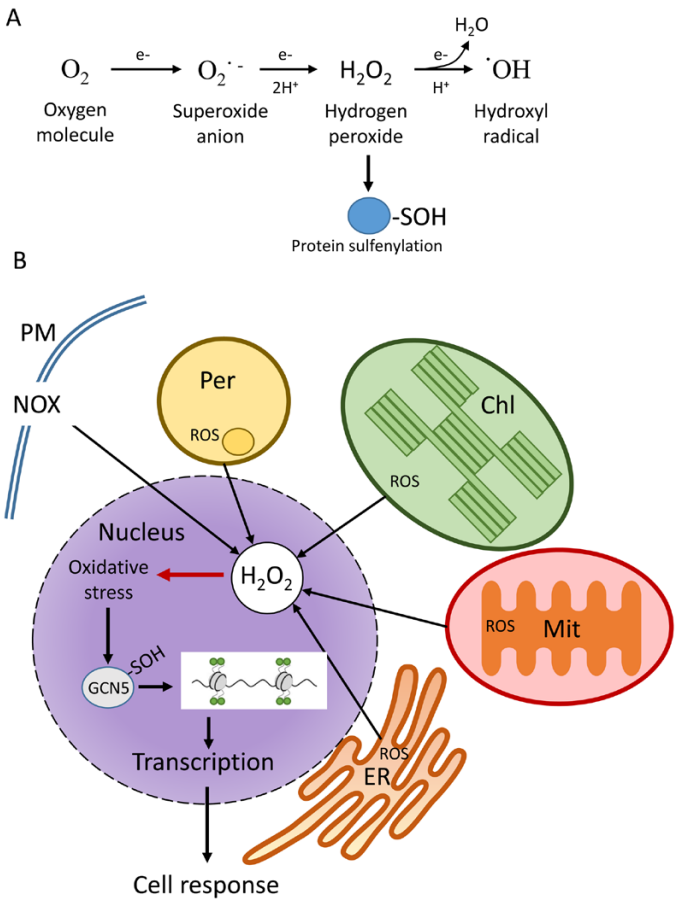

Fig. 1. Scheme of reactive oxygen species (ROS) production and H_2O_2-dependent epigenetic transcriptional regulation. (A) Sequential reduction of O_2 leads to the formation of various ROS, including the superoxide anion, hydrogen peroxide (H_2O_2), and the hydroxyl radical. H_2O_2 can oxidize cysteine residues in proteins, resulting in sulfenylation modifications. (B) H_2O_2 produced by NADPH oxidases (NOXs; also known as RBOHs) in chloroplasts (Chl), mitochondria (Mit), the endoplasmic reticulum (ER), and peroxisomes (Per) can alter H_2O_2 levels in the nucleus under stress conditions. This promotes the sulfenylation of nuclear proteins, such as GCN5, thereby affecting its activity and fine-tuning gene transcription and cellular responses. PM, plasma membrane. Green colour, acetylation.

The nuclear sulfenome and redox regulation of epigenetics

Advances in redox proteome analysis techniques have enhanced our understanding of plant regulation under changing metabolic and environmental conditions. To identify sulfenylated proteins, several redox proteomics methods have been developed, including labelling with dimedone, a nucleophilic reagent that selectively reacts with the electrophilic sulfur atom in sulfenic acid to form a stable thioesther bond (Huang *et al.*, 2018). An alternative approach uses the genetic Yeast Activation Protein-1 (YAP1) probe to identify sulfenylated cysteines which can be targeted to different subcellular locations (De Smet *et al.*, 2019).

Using the YAP1 approach, De Smet *et al.* (2025) carried out a pioneering study identifying the nuclear sulfenome in Arabidopsis plants exposed to H_2O_2, revealing 225 putative redox-active nuclear proteins. Among the enriched Gene Ontology categories, cell cycle processes, nuclear transport, histone methylation, and translational initiation were represented (De Smet *et al.*, 2025). Recent studies suggest that the altered redox status affects epigenetic changes, although the exact mechanisms remained unclear (Shen *et al.*, 2016; Ramakrishnan *et al.*, 2022). However, the interplay of redox regulation with DNA methylation, histone methylation and deacetylation, and miRNA biogenesis has been reported (Auverlot *et al.*, 2024; Plskova *et al.*, 2024). Additionally, epigenetic changes induced by stress conditions are also linked with primary metabolism which supplies intermediates and donor compounds required for these modifications, such as acetyl-CoA, *S*-adenosyl-methionine (SAM), and NAD(P)H (Auverlot *et al.*, 2024; Plskova *et al.*, 2024). Several enzymes

involved in the SAM cycle have been reported to be redox regulated (reviewed in Auverlot *et al.*, 2024; Plskova *et al.*, 2024), as well as some enzymes involved in peroxisomal β-oxidation (Sandalio *et al.*, 2019). Additionally, various HDACs have been reported as targets of redox-dependent PTMs (Auverlot *et al.*, 2024; Dard *et al.*, 2024). Notably, eight HDAs out of the 18 proteins annotated as HDACs in Arabidopsis have been shown to undergo redox-dependent PTMs (Dard *et al.*, 2024). The effects of redox-dependent PTMs in HDACs were first analysed in mammalian cells, where redox-regulated Cys residues have been identified in several HDACs (Auverlot *et al.*, 2024; Dard *et al.*, 2024). Interestingly, these redox-sensitive Cys residues are conserved between mammals and plants (reviewed in Auverlot *et al.*, 2024; Dard *et al.*, 2024). Nitric oxide (NO) donors induce redox changes in various HDACs from both mammals and plants; however, the outcomes of NO-dependent PTMs vary depending on the specific HDAC, with results ranging from activation to inactivation (Dard *et al.*, 2024). Additionally, other enzymes involved in histone acetylation, such as HAG2 and HAG3, have been shown to contain redox-regulated Cys residues (Dard *et al.*, 2024). However, unlike HDACs, direct evidence of redox regulation and its functional significance in HATs remains very limited.

De Smet *et al.* (2025) have now demonstrated that the HAT, GENERAL CONTROL NON-REPRESSED PROTEIN (GCN5), is a target of sulfenylation, and the authors have studied the functionality of this PTM. The acetyltransferase GCN5 is one of the most studied HATs. GCN5 mediates the acetylation of histone H3 at Lys9, Lys14, and Lys27, and also acetylate several non-histone targets such as ADA2 (Wu *et al.*, 2023; Yu *et al.*, 2024). De Smet *et al.* (2025) modellled the three-dimensional structure of Arabidopsis AtGCN5. This model revealed the unstructured, plant-specific N-terminal region, as well as the HAT and bromodomain structures, which are connected via a linker. AtGCN5 contains seven Cys residues, three of which—Cys293, Cys368, and Cys400—were identified as sulfenylated. This PTM negatively affected GCN5 enzymatic activity *in vitro* (De Smet *et al.*, 2025). Furthermore, a mutagenesis approach demonstrated that Cys293, located within the HAT domain, is essential for oxidative stress response induced by paraquat treatment, although it does not appear to play a significant role in plant growth and development under controlled conditions. However, substitution of Cys293 did not significantly alter the GCN5 protein interactome with the complexes SAGA and PAGA. None of the three Cys residues affected GCN5 subcellular localization. Thus, the authors conclude that GCN5 sulfenylation mainly affects GCN5 enzymatic activity specifically under oxidative conditions. In fact, *gcn5* mutants have been reported to be sensitive to other stress conditions, such as heat and salt stress, which involve alterations in redox homeostasis (Hu *et al.*, 2015; Zheng *et al.*, 2019; Sachdev *et al.*, 2021). Additionally, GCN5 modulates the cellular response to oxidative stress in yeast and human cells (Gaupe *et al.*, 2015). Therefore, GCN5 could be considered an ubiquitous cornerstone in the oxidative stress response.

GCN5 acetyltransferase: multilevel regulation?

Studies carried out in mammals and yeast have shown that PTM is crucial for regulating GCN5 activity, and three PTM types have been described so far: phosphorylation, ubiquitination, and acetylation (Xiao *et al.*, 2023). In mammalian tissues, GCN5 is activated by phosphorylation, giving rise to an increase in HAT activity. However, in some cases, the inhibition of GCN5 acetyltransferase activity by phosphorylation of the bromodomain has been reported (reviewed in Xiao *et al.*, 2023). In Arabidopsis, a phosphatase 2C protein interacts with GCN5 (Servet *et al*, 2008), suggesting that phosphorylation/dephosphorylation could regulate GCN5 activity, and putative phosphorylation of GCN5 has been reported using the PTM viewer (Willems *et al.*, 2024), although specific analysis of its functionality has not been addressed. De Smet *et al.* (2025) have identified a phosphatase between the GCN5 interactors which alters the phosphorylation status of GCN5, inhibiting its enzymatic activity. Ubiquitination acts by directing the target protein to the proteasome for further degradation, and Chen *et al.* (2023) have demonstrated ubiquitination of GCN5 in *Fusarium*. De Smet *et al.* (2025) add complexity to the GCN5 regulation by sulfenylation and demonstrate the negative effect of GCN5 oxidation on its activity, indicating the integration of redox signalling mechanisms with chromatin control of transcriptional activity through direct GCN5 redox changes. However, further analysis is required to fully establish the mechanism. ADA2–GCN5 interaction increases GCN5 binding to acetyl-CoA, stimulating its HAT activity, and recent studies show that lysine acetylation of ADA2 (associated with SAGA) by GCN5 affects ADA2 stability (Yu *et al.*, 2024). The potential impact of GCN5 sulfenylation on ADA2 stability and functionality warrants further investigation.

In summary, De Smet *et al.* (2025) provide crucial insights into the regulation of epigenetic processes by redox changes, identifying GCN5 as a key element in the redox epigenetic circuit. This information advances our understanding of plant responses to abiotic and biotic factors, showing how plants and possibly other organisms integrate oxidation processes and epigenetics, thus positioning GCN5 as a key regulator at the intersection of developmental and stress programmes (De Smet *et al.*, 2025). Whether sulfenylation could interact with other PTMs such as phosphorylation and their role in regulating dynamic changes in histone acetylation needs to be explored in depth. Understanding how different modifications work together to regulate spatio-temporal GCN5 functionality deserves further analysis involving GCN5 activity, protein structure, localization, and protein–protein interactome

analysis. This information will provide a framework for understanding gene re-programming regulation under oxidative stress conditions.

Acknowledgements

The author acknowledges Aurelio Collado for editing the figure.

Funding

This work was financed by grants P20_00364, from the Junta de Andalucía, and PID2021-122280NB-I00 from the Spanish Ministry of Science, Innovation and Universities, State Research Agency, and the European Regional Development Fund co-funding (MCIU/AEI/ERDF).

Conflict of interest

The author declares no conflict of interest.

Keywords: Epigenetics, histone acetyltransferases, nuclei, oxidative stress, redox homeostasis, signalling, sulfenylation.

References

Auverlot J, Dard A, Sáez-Vásquez J, Reichheld JP. 2024. Redox regulation of epigenetic and epitranscriptomic gene regulatory pathways in plants. Journal of Experimental Botany **75**, 4459–4475.

Chen A, Zhou Y, Ren Y, Liu C, Han X, Wang J, Ma Z, Chen Y. 2023. Ubiquitination of acetyltransferase Gcn5 contributes to fungal virulence in *Fusarium graminearum*. mBio **14**, e01499-23

Dard A, Van Breusegem F, Mhamdi A. 2024. Redox regulation of gene expression: proteomics reveals multiple previously undescribed redox-sensitive cysteines in transcription complexes and chromatin modifiers. Journal of Experimental Botany **75**, 4476–4493

De Smet B, Willems P, Fernandez-Fernandez AD, Alseekh S, Fernie AR, Messens J, Van Breusegem F. 2019. In vivo detection of protein cysteine sulfenylation in plastids. The Plant Journal **97**, 765–778.

De Smet B, Yang X, Plskova Z, *et al*. 2025. The nuclear sulfenome of Arabidopsis: spotlight on histone acetyltransferase GCN5 regulation through functional thiols. Journal of Experimental Botany **76**, 1569–1584.

Gaupe AC, Begley TJ, Tenniswood M. 2015. Gcn5 modulates the cellular response to oxidative stress and histone deacetylase inhibition. Journal of Cellular Biochemistry **116**, 1982–1992.

Grasser KD, Rubio V, Barneche F. 2021. Multifaceted activities of the plant SAGA complex. Biochimica et Biophysica Acta **1864**, 194613.

Hu Z, Song N, Zheng M, *et al*. 2015. Histone acetyltransferase GCN5 is essential for heat stress-responsive gene activation and thermotolerance in Arabidopsis. The Plant Journal **84**, 1178–1191.

Huang J, Willems P, Van Breusegem F, Messens J. 2018. Pathways crossing mammalian and plant sulfenomic landscapes. Free Radical Biology and Medicine **122**, 193–201.

Jiang J, Ding AB, Liu F, Zhong X. 2020. Linking signaling pathways to histone acetylation dynamics in plants. Journal of Experimental Botany **71**, 5179–5190.

Mittler R, Zandalinas SI, Fichman Y, Van Breusegem F. 2022. Reactive oxygen species signalling in plant stress responses. Nature Reviews. Molecular Cell Biology **23**, 663–679.

Muleya V, Lois LM, Chahtane H, Thomas L, Chiapello M, Marondedze C. 2022. (De)activation (ir)reversibly or degradation: dynamics of post-translational protein modifications in plants. Life (Basel) **12**, 324.

Mutlu B, Puigserver P. 2021. GCN5 acetyltransferase in cellular energetic and metabolic processes. Biochimica et Biophysica Acta **1864**, 194626.

Plskova Z, Van Breusegem F, Kerchev P. 2024. Redox regulation of chromatin remodelling in plants. Plant, Cell & Environment **47**, 2780–2792,

Ramakrishnan M, Papolu PK, Satish L, Vinod KK, Wei Q, Sharma A, Emamverdian A, Zou LH, Zhou M. 2022. Redox status of the plant cell determines epigenetic modifications under abiotic stress conditions and during developmental processes. Journal of Advanced Research **42**, 99–116.

Sachdev S, Ansari SA, Ansari MI, Fujita M, Hasanuzzaman M. 2021. Abiotic stress and reactive oxygen species: generation, signaling, and defense mechanisms. Antioxidants (Basel) **10**, 277.

Sandalio LM, Gotor C, Romero LC, Romero-Puertas MC. 2019. Multilevel regulation of peroxisomal proteome by posttranslational modifications. International Journal of Molecular Sciences **20**, 4881.

Servet C, Benhamed M, Latrasse D, Kim W, Delarue M, Zhou DX. 2008. Characterization of a phosphatase 2C protein as an interacting partner of the histone acetyltransferase GCN5 in *Arabidopsis*. Biochimica et Biophysica Acta **1779**, 376–382.

Shen Y, Issakidis-Bourguet E, Zhou DX. 2016. Redox status of the plant cell determines epigenetic modifications under abiotic stress conditions and during developmental processes. Journal of Experimental Botany **67**, 5291–5300.

Sies H, Belousov V, Chandel NN, Davies MJ, Jones DP, Mann GE, Murphy MP, Yamamoto M, Winterbourn C. 2022. Defining roles of specific reactive oxygen species (ROS) in cell biology and physiology. Nature Reviews. Molecular Cell Biology **23**, 499–515.

Smirnoff N, Arnaud D. 2019. Hydrogen peroxide metabolism and functions in plants. New Phytologist **221**, 1197–1214.

Willems P, Sterck L, Dard A, Huang J, De Smet I, Gevaert K, Van Breusegem F. 2024. The Plant PTM Viewer 2.0: in-depth exploration of plant protein modification landscapes. Journal of Experimental Botany **75**, 4611–4624.

Wu C-J, Liu Z-Z, Wei L, Zhou J-X, Cai X-W, Su Y-N, Li L, Chen S, He X-J. 2021. Three functionally redundant plant-specific paralogs are core subunits of the SAGA histone acetyltransferase complex in Arabidopsis. Molecular Plant **14**, 1071–1087.

Wu CJ, Yuan DY, Liu ZZ, Xu X, Wei L, Cai XW, Su YN, Li L, Chen S, He XJ. 2023. Conserved and plant-specific histone acetyltransferase complexes cooperate to regulate gene transcription and plant development. Nature Plants **9**, 442–459.

Xiao HT, Jin J, Zheng ZG. 2023. Emerging role of GCN5 in human diseases and its therapeutic potential. Biomedical Pharmacotherapy **165**, 114835.

Yu Y, Zhao F, Yue Y, Zhao Y, Zhou DX. 2024. Lysine acetylation of histone acetyltransferase adaptor protein ADA2 is a mechanism of metabolic control of chromatin modification in plants. Nature Plants **10**, 439–452.

Zheng M, Liu X, Lin J, *et al.* 2019. Histone acetyltransferase GCN5 contributes to cell wall integrity and salt stress tolerance by altering the expression of cellulose synthesis genes. *The Plant Journal* **97**, 587–602.