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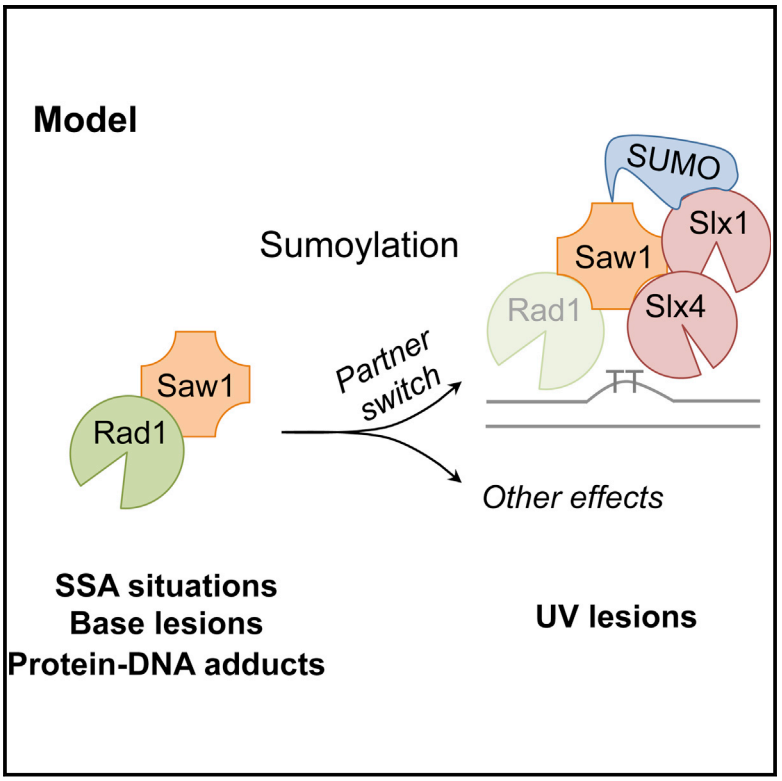
A Versatile Scaffold Contributes to Damage Survival via Sumoylation and Nuclease Interactions

Graphical Abstract



Highlights

The Saw1 scaffold has multiple roles and copes with diverse types of DNA lesions

Saw1 assists the Rad1-Rad10 nuclease in a range of DNA damage conditions

Sumoylation of Saw1 facilitates its interaction with another nuclease Slx1-Slx4

Saw1 sumoylation promotes UV resistance independently of two repair pathways

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In Brief

Scaffold proteins are not DNA repair enzymes themselves but make important contributions to DNA repair by regulating and coordinating various enzymes with their DNA substrates. Sarangi et al. reveal the versatility of the Saw1 scaffold by identifying how it copes with several types of DNA damage that depend on its nuclease interactions and sumoylation. These findings highlight the diverse ways in which multifunctional scaffolds can operate under genotoxic stress and how this is directed by protein modification.

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A Versatile Scaffold Contributes to Damage Survival via Sumoylation and Nuclease Interactions

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SUMMARY

DNA repair scaffolds mediate specific DNA and protein interactions in order to assist repair enzymes in recognizing and removing damaged sequences. Many scaffold proteins are dedicated to repairing a particular type of lesion. Here, we show that the budding yeast Saw1 scaffold is more versatile. It helps cells cope with base lesions and protein-DNA adducts through its known function of recruiting the Rad1-Rad10 nuclease to DNA. In addition, it promotes UV survival via a mechanism mediated by its sumoylation. Saw1 sumoylation favors its interaction with another nuclease Slx1-Slx4, and this SUMO-mediated role is genetically separable from two main UV lesion repair processes. These effects of Saw1 and its sumoylation suggest that Saw1 is a multifunctional scaffold that can facilitate diverse types of DNA repair through its modification and nuclease interactions.

INTRODUCTION

Timely repair of the large number of DNA lesions occurring in the genome is critical to prevent mutations and other alterations of the genetic information. This task requires collaborations between individual DNA repair enzymes, as well as with scaffold proteins that aid some of these enzymes. In particular, DNA nucleases that remove damaged sequences from the genome often carry out their functions in conjunction with scaffold proteins (e.g., Guzder et al., 2006; Hammel et al., 2011; Prolla et al., 1994; Vidal et al., 2001).

Most repair scaffolds are thought to assist a particular repair process (Guzder et al., 2006; Hammel et al., 2011; Prolla et al., 1994; Vidal et al., 2001). The budding yeast scaffold protein Saw1 was recently shown to support single-strand annealing (SSA) repair of double-strand breaks (DSBs) (Li et al., 2008, 2013). SSA entails the annealing of resected DNA at repeat sequences adjacent to the break, the subsequent removal of nonhomologous flaps, and final ligation (Fishman-Lobell et al., 1992; reviewed in Heyer et al., 2010; Krogh and Symington, 2004). In SSA, Saw1 recruits the Rad1-Rad10 nuclease to the break sites for flap removal (Li et al., 2008, 2013). This recruitment requires the coordinated interactions of Saw1 with the nuclease, the flap DNA, and upstream SSA factors (Li et al., 2008, 2013). SSA is considered error-prone repair as it leads to deletions or translocations (Fishman-Lobell et al., 1992; Heyer et al., 2010; Krogh and Symington, 2004).

Although Saw1 is thought to be an SSA-specific scaffold, Rad1-Rad10 is involved in processes that repair other types of DNA lesions (Figure 1A). These include the repair of UV lesions via the nucleotide excision repair (NER) pathway (reviewed in Schärer, 2013), as well as backup repair of base lesions and protein-DNA adducts (Guillet and Boiteux, 2002; Vance and Wilson, 2002). Compared with error-prone SSA repair, these processes contribute to cellular survival in specific genotoxic environments. It has not been explored whether Saw1 can aid Rad1-Rad10 in these repair contexts, nor is it known if Saw1 has Rad1-independent roles in DNA repair.

Here, we show that Saw1 promotes survival in different genotoxic environments that generate base lesions, protein-DNA adducts, and UV lesions. Saw1 interactions with Rad1 and DNA