

MIT Open Access Articles

Dual Roles for PARP1 during Heat Shock: Transcriptional Activator and Posttranscriptional Inhibitor of Gene Expression

The MIT Faculty has made this article openly available. **Please share** how this access benefits you. Your story matters.

Citation: Vyas, Sejal Kamlesh, and Paul Chang. "Dual Roles for PARP1 during Heat Shock: Transcriptional Activator and Posttranscriptional Inhibitor of Gene Expression." *Molecular Cell* 49.1 (2013): 1-3.

As Published: <http://dx.doi.org/10.1016/j.molcel.2012.12.017>

Publisher: Elsevier

Persistent URL: <http://hdl.handle.net/1721.1/84646>

Version: Author's final manuscript: final author's manuscript post peer review, without publisher's formatting or copy editing

Terms of use: Creative Commons Attribution-Noncommercial-Share Alike 3.0



Published in final edited form as:

Mol Cell. 2013 January 10; 49(1): 1–3. doi:10.1016/j.molcel.2012.12.017.

Dual Roles for PARP1 during Heat Shock: Transcriptional Activator and Posttranscriptional Inhibitor of Gene Expression

Sejal Vyas¹ and Paul Chang^{1,*}

¹Koch Institute for Integrative Cancer Research and Department of Biology, Massachusetts Institute of Technology, Cambridge, MA 02139, USA

Abstract

In this issue of *Molecular Cell*, Di Giammartino et al. (2012) identify a new function for PARP1 in the post-transcriptional regulation of mRNAs via ADP-ribosylation of poly(A) polymerase, a mRNA 3' processing enzyme.

Poly(ADP-ribose) polymerase 1 (PARP1, also known as ARTD1) is an abundant nuclear protein that covalently modifies target proteins with poly(ADP-ribose) (PAR) using NAD⁺ as a substrate. It is a multifunctional enzyme and has many DNA-related nuclear functions including DNA damage repair and transcriptional regulation under normal and stressed conditions (reviewed in Kraus, 2008; Luo and Kraus, 2012). During heat shock a small number of genes, referred to as stress-responsive genes, are upregulated, although overall gene expression is downregulated. PARP1 plays a critical role in this upregulation by mediating the transcriptional activation of Hsp70 genes (Tulin and Spradling, 2003). This occurs at Hsp70 loci where PARP1 becomes activated during heat shock and modifies itself and histones, resulting in PARP1 release from chromatin and the loss of nucleosomes within Hsp70 coding regions (Ouararhni et al., 2006; Petesch and Lis, 2008, 2012). In this issue, Di Giammartino et al. show that PARP1 also functions posttranscriptionally during heat shock, downregulating the expression of non-stress-responsive genes by PARylating poly(A) polymerase, the enzyme responsible for poly(A) tail synthesis, thus inhibiting polyadenylation activity (Di Giammartino et al., 2012).

In eukaryotes, polyadenylation of pre-mRNA is required for stability and is mediated by the 3' processing complex (reviewed in Millevoi and Vagner, 2010). Two reactions comprise polyadenylation: endonucleolytic cleavage of the nascent 3' end and poly(A) synthesis onto that end, catalyzed by CPSF73 and poly(A) polymerase, respectively. Polyadenylation is highly regulated by binding proteins and posttranslational modifications (Millevoi and Vagner, 2010). The complexity of 3' processing regulation is illustrated by the authors' previous work identifying ~80 proteins associated with the 3' processing complex, including PARP1 (Shi et al., 2009). To examine PARP1 function at the 3' processing complex, Di Giammartino et al. (2012) used nuclear extracts to assay 3' processing of the exogenous substrate SV40 late RNA, SVL. Induction of PARP enzymatic activity via NAD⁺ addition reduced 3' polyadenylation but not cleavage of SVL, suggesting that poly(A) polymerase but not CPSF73 activity was downregulated. Poly(A) polymerase was PAR modified under these conditions, indicating that regulation was due to PARP activity. Using small molecule inhibitors and siRNAs, they demonstrated that this effect was PARP1 dependent. The authors confirmed that PARP1 can bind and modify poly(A) polymerase in vitro using purified components and showed that such modification results in a dramatic

*Correspondence: pchang2@mit.edu.

decrease in SVL polyadenylation. To determine the mechanism of inhibition, Di Giammartino et al. examined 3' processing complex assembly and SVL RNA binding to poly(A) polymerase with or without PAR modification. When poly(A) polymerase was PARylated, RNA binding to poly(A) polymerase decreased, but no effect on assembly of the 3' processing complex was detected. Together these results identify poly(A) polymerase as a new PARP1 substrate and show that poly(A) polymerase function is regulated by PARylation in a PARP1-dependent manner.

To determine if PAP regulation by PARP1 occurs in cells, Di Giammartino et al. (2012) used nuclear extracts from cells grown under conditions known to activate PARP1 enzymatic activity — DNA damage, oxidative stress, and heat shock. Only heat shock resulted in polyadenylation repression, suggesting that the signal to activate PARP1 at 3' processing complexes is specific. These results were confirmed by examining polyadenylation of newly synthesized β -globin RNA in an inducible cell line and by examining polyadenylation of total RNA during heat shock. The authors demonstrated that poly(A) polymerase is PAR modified in vivo during heat shock and, using chromatin IP/real-time PCR, that poly(A) polymerase association with the 3' end of the reporter β -globin gene and other endogenous genes was reduced. This result suggests that PARylated poly(A) polymerase no longer associates with the newly transcribed mRNA during heat shock. Interestingly, poly(A) polymerase binding to the 3' ends of stress-induced genes was upregulated during heat shock, consistent with their increased expression and suggesting that the 3' processing complex can somehow distinguish between stress-induced and non-stress-induced transcripts.

This work reaffirms the importance of PARP1 and PAR in nuclear stress responses and adds postranscriptional polyadenylation as an additional layer of gene expression regulation during heat shock. Three critical questions are raised: (1) How does this mechanism of postranscriptional regulation differentiate between heat shock and non-heat shock transcripts? (2) How is PARP1 recruited to the 3' processing complex? (3) Is PARP1 at the 3' processing complex regulated independently of PARP1 at Hsp70 loci during heat shock?

How poly(A) polymerase distinguishes between heat shock and non-heat shock-specific transcripts is unknown. One possibility is that PARP1 activity is inhibited by proteins specifically bound to heat shock-induced transcripts, resulting in decreased PARylation of poly(A) polymerase at these transcripts. Alternatively, there could be specific recruitment of PAR binding proteins to PARylated poly(A) polymerase that convey transcript specificity to poly(A) polymerase activity. Comparing 3' processing complex proteins bound to heat shock-induced versus non-heat shock transcripts and identifying nuclear PAR binding proteins from heat-shocked cells could provide insight.

How PARP1 is recruited to the 3' processing complex is currently unknown. While it is unclear whether PARP1 activation precedes recruitment, the low abundance of PARP1 at 3' processing complexes under normal conditions suggests that it is not a constitutive component and that a signal is required for recruitment to the 3' processing complex (Shi et al., 2009). Such recruitment could occur via increased binding affinity between 3' processing components and PARP1 or via binding to the PAR attached to activated PARP1. Examining the binding interactions between PARylated PARP1 and the 3' processing complex could shed significant light on how PARP1 recruitment is regulated.

During heat shock PARP1 enzymatic activity at Hsp70 loci is specifically upregulated in a heat shock factor (HSF)-dependent manner (Petesch and Lis, 2012). Whether or not PARP1 is independently activated at the 3' processing complex during heat shock is unknown. However, either possibility has important functional implications described by the single

pool and multiple pool models (Figure 1). In the multiple pool model, PARP1 is activated at the Hsp70 loci and at the 3' processing complex independently, requiring separate regulatory mechanisms. In the single pool model, HSF is the sole activator of PARP1, and activated PARP1 released from Hsp70 loci is recruited to the 3' processing complex. While this model requires prior activation at Hsp70 loci, it has the benefit of potential crosstalk between the two sites of activity (Figure 1). Clearly, identifying the mechanism of activation of PARP1 in 3' processing is critical to understanding this interesting new PARP1 function and will shed light on how it simultaneously functions as both a transcriptional activator and a posttranscriptional inhibitor during heat shock.

References

- Di Giammartino DC, Shi Y, Manley JL. *Mol Cell*. 2012; 49:7–17. this issue. [PubMed: 23219533]
- Kraus WL. *Curr Opin Cell Biol*. 2008; 20:294–302. [PubMed: 18450439]
- Luo X, Kraus WL. *Genes Dev*. 2012; 26:417–432. [PubMed: 22391446]
- Millevoi S, Vagner S. *Nucleic Acids Res*. 2010; 38:2757–2774. [PubMed: 20044349]
- Ouararhni K, Hadj-Slimane R, Ait-Si-Ali S, Robin P, Miettton F, Harel-Bellan A, Dimitrov S, Hamiche A. *Genes Dev*. 2006; 20:3324–3336. [PubMed: 17158748]
- Petes SJ, Lis JT. *Cell*. 2008; 134:74–84. [PubMed: 18614012]
- Petes SJ, Lis JT. *Mol Cell*. 2012; 45:64–74. [PubMed: 22178397]
- Shi Y, Di Giammartino DC, Taylor D, Sarkeshik A, Rice WJ, Yates JR 3rd, Frank J, Manley JL. *Mol Cell*. 2009; 33:365–376. [PubMed: 19217410]
- Tulin A, Spradling A. *Science*. 2003; 299:560–562. [PubMed: 12543974]

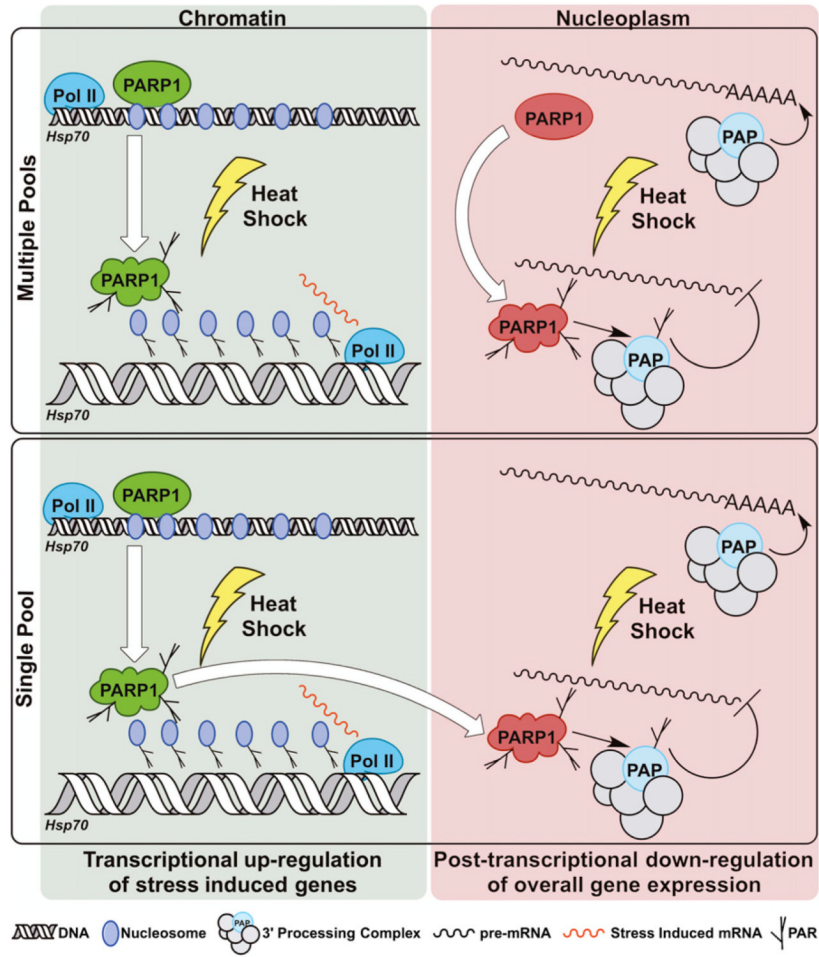


Figure 1. Regulation of PARP1 during Heat Shock

PARP1 has multiple functions during heat shock, acting as both a transcriptional activator and a posttranscriptional inhibitor of gene expression. If and how these functions are related remains unclear. Multiple pools of PARP1 can exist, established by distinct targeting mechanisms, with each pool being activated independently upon heat shock. Alternatively, chromatin-bound PARP1 at stress-responsive genes such as Hsp70 could be recruited to the 3' processing complex upon heat shock, potentially via binding to PAR attached to PARP1. In each of these models, activation of PARP1 during heat shock results in two distinct outcomes: upregulation of stress-induced gene expression through PARP1 bound to stress-induced gene loci such as Hsp70, and downregulation of overall gene expression via inhibition of poly(A) polymerase at the 3' processing complex.