Available online at www.sciencedirect.com
ScienceDirect
Mathematical Biosciences 210 (2007) 60–77
Mathematical Biosciences
www.elsevier.com/locate/mbs

HAUSP-regulated switch from auto- to p53 ubiquitination by Mdm2 (*in silico* discovery)

Paul Brazhnik [a,*], Kurt W. Kohn [b]

[a] *Department of Biological Sciences, Virginia Tech, Blacksburg, VA 24061, United States*
[b] *NCI, NIH, Bethesda, MD 20892, United States*

Received 20 June 2006; received in revised form 19 December 2006; accepted 15 May 2007
Available online 23 May 2007

Abstract

Stability of the 'guardian of the genome' tumor suppressor protein p53 is regulated predominantly through its ubiquitination. The ubiquitin-specific protease HAUSP plays an important role in this process. Recent experiments showed that p53 demonstrates a differential response to changes in HAUSP which nature and significance are not understood yet. Here a data-driven mathematical model of the Mdm2-mediated p53 ubiquitination network is presented which offers an explanation for the cause of such a response. The model predicts existence of the HAUSP-regulated switch from auto- to p53 ubiquitination by Mdm2. This switch suggests a potential role of HAUSP as a downstream target of stress signals in cells. The model accounts for a significant amount of experimental data, makes predictions for some rate constants, and can serve as a building block for the larger model describing a complex dynamic response of p53 to cellular stresses.

Keywords: p53 Ubiquitination; HAUSP; Mathematical models; Mdm2; Stress response

1. Introduction

Cell fate is largely determined under conditions of diverse stresses by the tumor suppressor protein p53 [1–3]. Activity of this transcriptional regulator is controlled by different post-transcriptional

* Corresponding author. Tel.: +1 540 231 6398.
E-mail address: brazhnik@vt.edu (P. Brazhnik).

0025-5564/$ - see front matter
doi:10.1016/j.mbs.2007.05.005

modifications (phosphorylation, ubiquitination, acetylation, etc.) [1,4–6], by interactions with other proteins, including Mdm2 [7,8] and HAUSP [9,10], and by changes in a subcellular localization [1,11–15]. Stabilized upon exposure of a cell to the stress, p53 accumulates in nucleus and activates genes arresting a cell division cycle or guiding the cell to apoptosis, therefore precluding propagation of genetic instabilities [2,16].

p53 is one of the most connected genes, a hub in the cell regulatory network [2,17]. Its responses are cooperative (involve a number of other proteins) [18,19], dynamically complex (from switching to oscillatory or pulsatile) [20–24], and are not easy to comprehend. In this situation a consistent quantitative analysis and computational modeling becomes paramount. A feasible strategy to address complexity of the p53 regulatory network is to divide it into smaller subnetworks (modules) that can be quantitatively modeled and analyzed independently, and then to integrate the modules into larger networks [25]. This work reports a quantitative computational model of the Mdm2-mediated p53 ubiquitination module with the emphasis on the role of the deubiquitinating enzyme HAUSP in p53 stability.

The postranscriptional modification by ubiquitination is an important mechanism regulating protein activities in cells [26–28]. Ubiquitination of p53 may interfere with its activation (by competing for binding sites with acetylation [4,29,30]) and leads to the protein degradation through the ubiquitin proteolysis pathway [31]. It can be accomplished by several proteins, Mdm2, COP1, Pirh2 [1,32], among which Mdm2-mediated ubiquitination is considered to be the most important mechanism of regulation of p53 abundance [1,33,34]. This mechanism is responsible for maintaining low levels of p53 in a normal physiological state of a cell as well as for the rapid increase of p53 after genotoxic stress. Many tumors with wild type p53 exhibit supra-physiological levels of Mdm2 [7]. Importantly, Mdm2 itself is a transcriptional target of p53 [2]. The gene is activated during a stress [35] closing thereby the p53–Mdm2 regulatory negative feedback loop which significance has not been yet sufficiently understood.

The issue of regulation of the p53 protein stability via Mdm2-mediated ubiquitination has recently received a new twist with the discovery that the ubiquitin-specific protease HAUSP (also known as human USP7) [36] can deubiquitinate both, p53 and Mdm2, *in vitro* and *in vivo* [9], suggesting thus a potential tumor-suppressive role of this enzyme [37], Fig. 1. HAUSP is co-localized with a p53 protein at the PML-NB (promyelocytic leukaemia nuclear body) and thus can be an important regulator of p53 stability *in vivo* [38]. It was shown to be crucially involved in both the regulation of p53-dependent apoptosis and the inhibition of a cell growth [9].

Two different, seemingly conflicting experimental results on the effect of HAUSP on p53 levels have been reported. First Li et al. showed that HAUSP, when overexpressed, increases p53 amount in cells [9]. Then Cummins et al. observed [39], that the disruption of the HAUSP gene in human cells lead to the substantial increase in the steady-state level of a p53 protein and its functional activation. Finally, in the most recent work [20], it was demonstrated that when a partial reduction of the HAUSP level (by RNAi) destabilized (reduced) p53, nearly a complete HAUSP ablation stabilized and activated it.

The data-driven mathematical model of the p53 ubiquitination reported here addresses the aforementioned differential response of p53 to changes in HAUSP concentrations. The model shows, using current experimental data, that p53 ubiquitination can be caste in a consistent chemical kinetics model exhibiting naturally a HAUSP-regulated switch from auto- to p53 ubiquitination by Mdm2. This HAUSP-dependent switching behavior explains both mentioned above