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MATHEMATICAL MODEL IN FUNCTION OF *p*53 GENE IN *p*53-*mdm*2 NETWORK IN APOPTOSIS

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The *p*53 network is the most important pathway involved in preventing the initiation of cancer. More than half of all human cancers contain mutations in the *p*53 gene and in almost all cancers, the *p*53 regulatory circuit is functionally inactivated. *mdm*2 gene plays a very important role in *p*53 network. It regulates the levels of intracellular *p*53 protein concentration through a feedback loop. *p*53 levels and activity are up regulated in response to various stresses including DNA damage, hypoxia, and oncogene activation. Active *p*53 initiates different transcriptional programs that result in cell cycle arrest, cellular senescence or apoptosis. Under normal conditions the *p*53 levels are kept very low. When there is DNA damage, the levels of p53 protein rise and if there is a prolonged elevation the cell shifts to apoptosis, and if there is only a short elevation the cell cycle is arrested and the repair process begun. In this paper, the function of *p*53 gene in *p*53-*mdm*2 module and the associated pathways for apoptosis are studied. A number of key predictions, and experiments to specific aspects of cell cycle arrest and cell death are discussed.

INTRODUCTION

In every normal cell, there is a protective mechanism against tumoral degeneration. This mechanism is based on the *p*53 network. *p*53 is a gene that codes a protein in order to regulate the cell cycle. *p*53 also known as "the guardian of the genome" [1]. The name is due to its molecular mass: it is a 53 kilo Dalton fraction of cell proteins. Due to its major implication in cancer prevention and due to the actions described above, *p*53 has been intensively studied in the last two decades. The tumour suppresser gene *p*53 and the mdm2 oncogene have important role in cell cycle checkpoints, apoptosis, growth control and oncogenesis. The *mdm*2 protein promotes the rapid degradation of the *p*53 protein, while *p*53 protein activates the transcription of the *mdm*2 gene [2].

The *p*53 gene has been mapped to chromosome 17. In the cell, *p*53 protein binds DNA, which in turn stimulates another gene (the *p*21 gene) to produce the *p*21 protein, a protein that interacts with a cell division-stimulating protein (cdk2). When *p*21 forms a complex with cdk2 the cell cannot pass through to the next stage of cell cycle. A mutant *p*53 can no longer bind

DNA in an effective way, and as a consequence the *p*21 protein is not made available to act as the 'stop signal' for cell division. Thus cells divide uncontrollably, and form tumors. It is clear that *p*53 is just one component of a network of events that culminate in tumor formation.

Numerous *p*53-dependent target genes have been identified that play a role as downstream effectors of each *p*53 function. For example, the cyclin dependent kinase inhibitor *p*21Waf1/Cip1 is a direct *p*53 target and deletion of this gene significantly reduces the cell cycle arrest response to *p*53. More recently, the ability of *p*53 to participate directly in DNA repair has been shown to be associated with the activation of a ribonucleotide reductase, named *p*53*R*2 [5]. In general, each *p*53 response appears to reflect activation of several genes. *p*53 can induce expression of proteins that target both the mitochondrial and the death-receptor-induced apoptotic pathways. *p*53-mediated induction of proteins that localize to the mitochondria, such as Bax , NOXA , PUMA and p53AIP1, triggers cytochrome c release and activation of the Apaf-1/caspase-9 apoptosome [4]. *p*53 also induces the expression of death receptors, such as Fas and Killer/DR5, and the death-domain-containing protein PIDD, which are likely to participate in death-receptor-mediated apoptotic signaling [4]. In many cases, inhibition of expression of these *p*53 target genes has been shown to be associated with a decrease in *p*53-induced apoptosis, although in most cases this reduction is incomplete, consistent with the existence of multiple *p*53-inducible apoptotic target genes. One of the key regulators of *p*53 is the *mdm*2 protein, which can both inhibit *p*53's transcriptional activity and target *p*53 for degradation. *p*53 function depends on nuclear localization, and both nuclear import and nuclear export of *p*53 are tightly regulated. The biological consequences of *p*53 activity include cell-cycle regulation, induction of apoptosis, development, differentiation, gene amplification, DNA recombination, chromosomal segregation, and cellular senescence [2].

In this paper, the reaction between the *p*53 and *mdm*2 protein are converted as differential equations. The rate of change of *p*53 protein and *mdm*2 protein with respect to time are taken as the primary data. The correlation between the data gives the binding of *p*53 and *mdm*2 gene in the network. The activation of *p*53 gene in *p*53-*mdm*2 network will decide the fate of the cell. That is either the cell cycle will be arrested or the cell will goes to apoptosis.

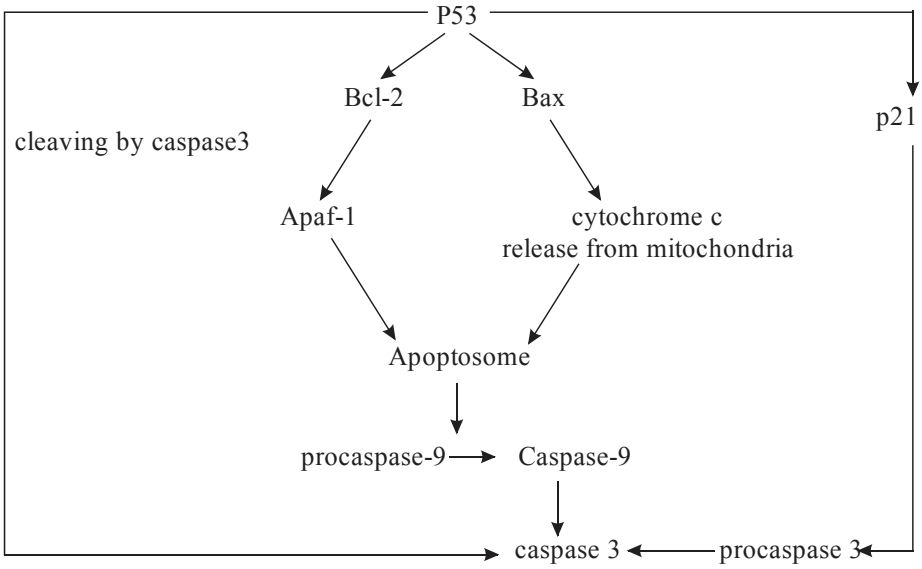


Fig. : The *p*53-mediated apoptotic pathway.

The above figure shows a schematic diagram of the *p*53-mediated apoptotic pathway which is activated when the *p*53-*MDM*2 interaction is inhibited by nutlin molecules. The *p*53 proteins up regulate the synthesis of the pro-apoptotic Bax proteins and down regulate that of the anti-apoptotic Bcl-2 proteins [2]. The Bcl-2 binds the Bax protein to form a heterodimer and consequently suppress the pro-apoptotic activity of Bax. The Bcl-2 further binds the apoptotic protease-activity factor 1 (Apaf-1) and thereby excludes the binding of procaspase-9 to Apaf-1. The Bax and other pro-apoptotic proteins help in the release of cytochrome c proteins from the mitochondria. Apaf-1 binds cytochrome c to form the apoptosome complex which in turn binds procaspase-9. A family of proteases termed caspases play a central role in bringing about apoptosis. The blocking of caspase activity can slow down or prevent apoptosis. Caspases selectively cleave a set of specific target proteins at one or a few positions, always after an asparate residue. The cleavage results in non-functional target proteins. The binding of the procaspase-9 to the apoptosome leads to initiator caspase-9 activation through cleavage of the bound procaspase-9. The initiator caspases activate the executioner or effector caspases. This is followed by the cleavage of procaspase-3 to yield the active effector molecule, caspase- 3. Executioner caspases like caspase-3 cleave key cellular proteins and dismantle the cell resulting in apoptosis. The end comes provided the executioner caspases such as caspase-3 are activated to sufficiently high levels. The level of activation depends on the competition between the pro- and anti-apoptotic agents/processes constituting the apoptotic pathway. The apoptotic network further includes two important processes, namely, the cleavage of the anti-apoptotic Bcl-2 by caspase-3 and the inhibition of the processing and activation of procaspase-3 by *p*21[2]. These two processes provide direct links between the events upstream and downstream of cytochrome c release.

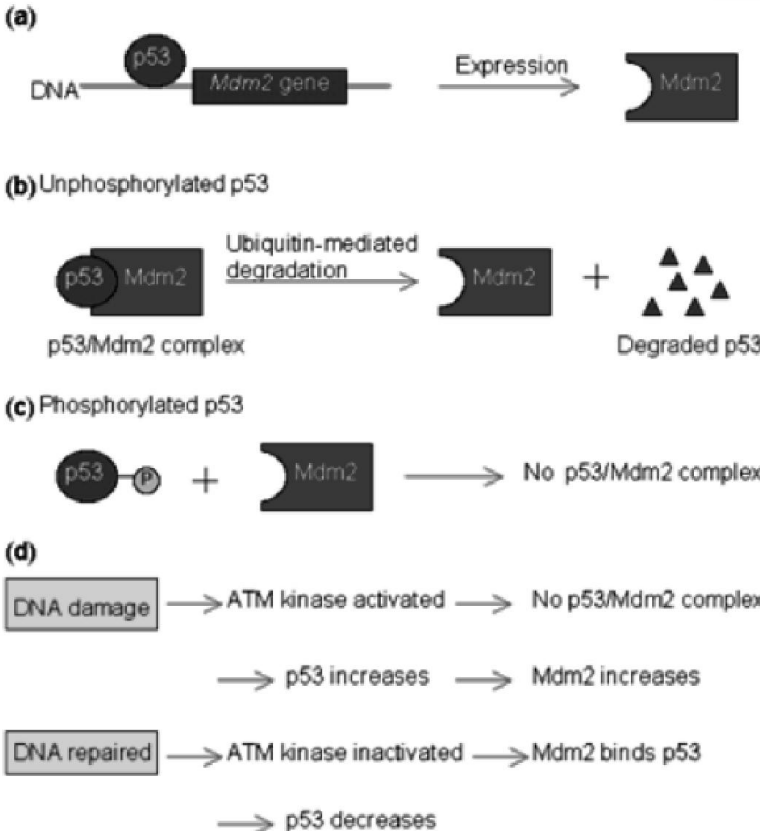
***p*53 and *mdm*2 network parameters:**

The simplified version of the network consists of only 9 representative candidate proteins; the entire network of hundreds of *p*53 regulators is expected to be much more complex. As described above, numerous proteins interact with *p*53 and mdm2.

The parameters which are included in the network and their values are as follows [5]:

Table 1.

1.	*e*1	*p*53-dependent *mdm2* transcription rate	0.02 μM min^{-1}
2.	*kb*1	*p*53-*Mdm*2 complex dissociation rate	123.6 min^{-1}
3.	*Km*	Michaelis constant of *p*53-dependent *mdm2* transcription	0.16 μM
4.	*rp*53	Translation rate of *P*53	0.01 min^{-1}
5.	rmdm2	Translation rate of *Mdm*2	0.04 min^{-1}
6.	*p*53	Basal degradation rate of *p*53	0.03 min^{-1}
7.	*mdm*2	Basal degradation rate of *Mdm*2	0.033 min^{-1}
8.	*k*1	*Mdm*2-dependent *P*53 degradation through *C*1	0.2 min^{-1}
9.	*kf*1	*P*53-*Mdm*2 complex association rate	552 μM min^{-1}
10.	τ1	*mdm*2 transcription	30 min
11.	τ5	*p*53 transcription delay	30 min
12.	τ6	*P*53 translational delay	10 min
13.	τ8	*Mdm*2 translational delay	10 min
14.		*P*53 protein	0.0258
15.		*Mdm*2 protein	0.15



Various activation of p53 gene in DNA damage, DNA repair, Ubiquitin –mediated degradation and the rate of increase and decrease of p53 and mdm2 are shown in the above figure.

The p53 –mdm2 complex c1 formation and dissociation is also included in this model to represent the binding and unbinding between mdm2 and p53 protein molecules.

The reaction between p53 and mdm2 protein can be written as

$$\left.\begin{array}{l} p53 + Mdm2 \xrightarrow{k_{f1}} p53Mdm2 \\ p53Mdm2 \xrightarrow[kb_1]{} p53 + product \end{array}\right\} \quad \ldots (1)$$

Here p represents the product. The general expression for the velocity (rate) of this reaction is

$$v = \frac{d[p]}{dt} = kf_2[p53Mdm2] \quad \ldots (2)$$

The overall rate of production of [p53mdm2] is the difference between the rates of elementary reactions.

Michaelis and Menten assumed that $kf_1 >> kf_2$ so that the first step of the reaction achieves equilibrium.

$$ks = \frac{kb_1}{kf_1} = \frac{[p53][Mdm2]}{[p53Mdm2]} \quad \ldots(3)$$

where ks is the dissociation constant.

The reaction between $p53$ and $mdm2$ can be written as a set of differential equations

$$\frac{d[p53Mdm2]}{dt} = kf_1[p53][Mdm2] - (kb_1 + kf_2)(p53Mdm2) \quad \ldots(4)$$

$$\frac{d[Mdm2]}{dt} = -kf_1[p53][Mdm2] + kb_1(p53Mdm2) \quad \ldots(5)$$

$$\frac{d[p53]}{dt} = -kf_1[p53][Mdm2] + (kb_1 + kf_1)(p53Mdm2) \quad \ldots(6)$$

$$\frac{d[p]}{dt} = kf_2[p53Mdm2] \quad \ldots(7)$$

With initial conditions $p53(0) = p_0$, $Mdm2(0) = M_0$, $p53Mdm2\ (0) = PM_0$ and Product $(0) = P_0$

Combining the first three equations we get

$$\frac{d[p53]}{dt} = -kf_1[p0][Mdm2] + kf_1P53 + kb_1(p53Mdm2) \quad \ldots(8)$$

and $$\frac{d[p53Mdm2]}{dt} = kf_1[p0][Mdm2] - (kf_1p53 + kb_1 + kf_2)(p53Mdm2) \quad \ldots(9)$$

With the initial conditions $P53(0) = p_0 = 0.0258$ μM, $Mdm2\ (0) = M_0 = 0.15$ μM $p53\ Mdm2\ (0) = PM_0 = 0.06$ μM, $Kf_1 = 0.2\ \text{min}^{-1}$, $kb_1 = 123.6$ m,

Assume that $kf_2 = 0.4\ \text{m}^{-1}$,

Result

We can calculate the values of $\frac{d[p53]}{dt}$ and $\frac{d[p53mdm2]}{dt}$ from equations (8) and (9). Assume that these two set of values be $x\ (t)$ and $y\ (t)$ which are listed below.

$X\ (t)$	$Y\ (t)$
7.420	–7.485
7.425	–7.436
7.428	–7.337
7.433	–7.335
7.437	–7.33
7.441	–7.326
7.440	–7.320
7.44	–7.316

From this table of values the mean, standard deviation and the correlation coefficient are calculated. The mean value of rate of change of p53 and mdm2 are as follows.

$$\bar{x} = \frac{\sum X(t)}{n} \quad \text{and} \quad \bar{y} = \frac{\sum Y(t)}{n}$$

$$\bar{x} = \frac{59.464}{8} \quad \text{and} \quad \bar{y} = \frac{-58.835}{8}$$

$$\bar{x} = 7.433 \quad \text{and} \quad \bar{y} = -7.3544.$$

The standard deviation of *p*53 and mdm2 protein and correlation are as follows

$$\sigma_x = \sqrt{\frac{1}{n}\sum (x - \bar{x})} \qquad = 0.0.070356$$

$$\sigma_y = \sqrt{\frac{1}{n}\sum (y - \bar{y})^2} \qquad = 0.04585$$

correlation coefficient $r = \dfrac{\frac{1}{n}\sum (x - \bar{x})(y - \overline{y})}{\sigma_x . \sigma_y}$

$$r = \frac{\frac{1}{8}(0.002485)}{(0.0070356)(0.04585)} = 0.9629$$

The correlation between *p*53 and *mdm*2 proteins are $r = 0.9$

Conculusion

We conclude that if $x(t)$ increases then $y(t)$ also increases, and the correlation between two set of values is positive. When the *p*53 molecules are increased then automatically the *mdm*2 molecules are also increased. In normal cells the level of *p*53 is very low. When the *p*53 protein is bind to *mdm*2 gene in the network, *p*53 gene will decide the fate of the cell. From this network the level of *p*53 is raised , *p*53 gene will gives signal to the cell and then the cell shifts to apoptosis. Further , the function of *p*53 gene will play an important role in the genetheraphy treatment for cancer.

Reference

1. Horhat, F.R., Neamtu, M., Mircea, G., "Mathematical models and numerical simulations for the *P*53-*Mdm*2 Network", *Applied Sciences*, Vol. **10**, pp. 94-106 (2008).
2. Bose, Indrani and Ghosh, Bhaswar, "The *p*53-*Mdm*2 network : from oscillations to apoptosis", *J. Biosci.,* **32(5)**, 000–000, © Indian Academy of Sciences August (2007).
3. Tiana, G., Jensen, M.H. and Sneppen, K., "Time Delay as a key to apoptosis induction in the *p*53 Network", *European Physical Journal*, **B29**, 135-1-40 (2002)
4. Kevin, M. Ryan, Andrew, C. Phillips and Karen, H. Vousden, "Regulation and function of the *p*53 tumor suppressor protein", *Current Opinion in Cell Biology*, **13**, 332-337 (2001).
5. Chonga, K.H., Samarasinghea, S. and Kulasiria, D., "Mathematical modelling of *p*53 basal dynamics and DNA damage response", *20th international Congress on Modelling and Simulation*, Adelaide, Australia, 1-6 December (2013).
6. Zhiyuan, Li, Ming, Ni, Jikun, Li, Yuping, Zhang, Qi, Ouyang, Chao, Tang, "Decision making of the *p*53 network : Death by integration", *Journal of Theoretical Biology*, **271**, 205–211 (2011).
7. Steele, R. J. C., Thompson, A. M., Hall, P. A. and Lane, D. P., "The *p*53 tumour suppressor protein", *Bri'N-sh Joumal of Surgery*, **85**, 1460-1467 (1998).

8. Wang, Hua and Peng, Guang, "Mathematical Model of Dynamic Protein Interactions Regulating *p*53 Protein Stability for Tumor Suppression", *Computational and Mathematical Methods in Medicine*, Article ID 358980, 6 pages (2013).
9. Tsygvintsev, Alexei, Marino, Simeone and Kirschner, Denise E., "*A Mathematical Model of Gene Therapy for the Treatment of Cancer*".
10. Ballesta, Annabelle, Lopez, Jonathan, Popgeorgiev, Nikolay, Gonzalo, Philippe, Doumic, Marie, Gillet, Germain, "Data-driven modeling of SRC control on the mitochondrial pathway of apoptosis : implication for anticancer therapy optimization", *Public Library of Science*, **9** (4) (2013).
11. Giorgio, Gagliaa, Yinghua, Guana, Jagesh, V. Shaha and Galit, Lahava, "Activation and control of *p*53 tetramerization in individual living cells", Medical Research Council Laboratory of Molecular Biology, Cambridge, United Kingdom (2013).

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