Absence of WDR13 in p53-Null Mice Increases Longevity

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Short Report

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Abstract

Absence of WDR13 is known to be involved in pancreatic, colon and uterine hyperproliferation. However, a recent study showed its antiproliferative role liver regeneration in response to hepatotoxins. These findings intrigued to study the role of WDR13-null condition in Trp53 knockout mouse to study the tumour predisposition and survival. We report absence of *Wdr13* in *Trp53*-null background alleviates the tumour load, in turn increasing total survival in mouse model.

Background

Cancer is a grave disease that arises because of abnormal cell growth and can invade or spread to other body parts (1). The rate of occurrence of cancer worldwide is increasing at an alarming rate, 12.7 million cases in 2008 (2). The p53 is a key protein for tumour suppression and found to be mutated in over half of all human cancers (3). Individuals with Li–Fraumeni Syndrome, a classic cancer predisposition disorder, inherit mutant p53 allele and are highly cancer-prone (4). The null mutation of p53 does not display embryonic lethality but have decreased survival as compared to wild-type mice; the majority of *p53* mice succumbed to tumours between 2–9 months of age (5, 6).

Earlier studies on WDR13 suggest its involvement in cell cycle regulation in different tissues like pancreas (7–9), liver (10) adipose (11), colon (12) and uterus (13). *Wdr13* expression has also been reported in many human cancers like breast cancer, glioma, Ewing's sarcoma, lung cancer, melanoma, testes cancer, ovary teratocarcinoma, and urothelial cancer (Human protein atlas <http://www.proteinatlas.org/ENSG00000101940-WDR13/cancer>). High expression of WDR13 in above mentioned cancers further strengthens the postulate that WDR13 has a vital role in regulating cell cycle. Therefore, it was interesting to study the cancerous phenotype after the introgression of *Wdr13*-null mutation in a cancer mouse model. To test above proposition, *Wdr13*^{-/-} mice were mated with *p53*^{-/-} mice and *Wdr13*^{-/-}|*p53*^{-/-} double knockout mouse was generated.

Methods/experimental

Animals-

Ethics approval-

The animals were approved by the Institutional Animal Ethics Committee (IAEC) of CSIRCCMB, Hyderabad, India (trial registration number- 20/1999/CPCSEA dated 10/3/99). C57Bl/6J *Wdr13*^{-/-} male mice were mated with *p53*^{-/-} females to obtain double heterozygous animals. Then the littermates were crossed to obtain double knockout (DKO) animals. Mice were housed at 22–25°C temperature with 12 hr light/dark cycle. All the mice were fed standard chow for entire experimental duration ad libitum. 20 animals per group were used, DKO were 21. Severely ailing mice were sacrificed according to the IAEC rules.

Genotyping- *Wdr13* genotyping was done as explained in Singh, et al, 2012 (11) and *p53* genotyping was performed as described by Jacks, et al, 1994 (6).

Results And Discussion

The absence of WDR13 in *p53*^{-/-} mice enhanced their longevity: while all the *p53*^{-/-} mice died by 36 weeks of age the double knockout mice survived more than a year, as determined by Kaplan-Meier estimate (Fig. 1).

The double knockout mice were comparatively healthy and had lower tumour load at 6 months of age as compared to *p53*^{-/-} mice. Further analyses of the tissues revealed significant difference in the *Wdr13*^{-/0}*p53*^{-/-} double knockout mice and the *p53*^{-/-} mice at 6 months.

The results establish that the absence of WDR13 in *p53*^{-/-} mice is beneficial for survival of *p53*^{-/-} mice as the double knockout mice had better longevity. The double knockout mice also had lesser tumour load as compared to *p53*^{-/-} mice. Role of WDR13 in cell cycle regulation in different tissues has been attributed to PPAR γ . *Wdr13*^{-/0} mice is known to have a higher expression levels of PPAR γ in adipose (8) and in liver (10). Activation of PPAR γ slows the regeneration of liver in CCl₄ administered *Wdr13*^{-/0} mice (10). The promoter of *Ppar γ* has an AP1 site and WDR13 is known to bind AP1 sites with c-Jun (8). PPAR γ is known to stimulate apoptosis in cancer cells thereby ameliorating cancer phenotype (14). PPAR γ agonists have been used to study apoptosis in cell lines (15). In thyroid cancer cells lines, troglitazone demonstrated increased c-Myc expression without changing the expression of Bcl-2 or Bax (16).

Conclusions- In the light of these observations, dysregulation of PPAR γ in absence of WDR can be attributed to be involved in amelioration of cancerous phenotype of *p53*^{-/-} mice. Studying the expression levels of PPAR γ and its target proteins in different tissues of *Wdr13*^{-/0}*p53*^{-/-} mice is an interesting venue for future research.

Declarations

Ethics approval- The animals were approved by the Institutional Animal Ethics Committee (IAEC) of CSIRCCMB, Hyderabad, India (trial registration number- 20/1999/CPCSEA dated 10/3/99).

Consent for publication- Authors provide consent of publication.

Availability of data and material- Data available on request

Competing interest- Authors declare no conflicting interest.

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Authors' contributions- APM conceived the idea and performed all the experiments. APM wrote the manuscript. BJL helped in maintenance and breeding of mice.

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References

1. de Martel C, Ferlay J, Franceschi S, Vignat J, Bray F, Forman D et al (2012) Global burden of cancers attributable to infections in 2008: a review and synthetic analysis. *Lancet Oncol* 13(6):607–615
2. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D (2011) Global cancer statistics. *CA Cancer J Clin* 61(2):69–90
3. Brady CA, Attardi LD. p53 at a glance. *J Cell Sci.* 2010 Jul 21;123(15):2527 LP – 2532
4. Malkin D. Li-Fraumeni Syndrome. Levine AJ (eds) (2011) vol 2, *Genes & Cancer*. Los Angeles, CA, Sage, pp 475–484
5. Donehower L, Harvey M, Slagle BL, McArthur MJ, Montgomery C, Butel JS et al (1992) Mice deficient for p53 are developmentally normal but susceptible to spontaneous tumours. *Nature* 356:215–221
6. Jacks T, Remington L, Williams BO, Schmitt EM, Halachmi S, Bronson RT et al (1994) Tumor spectrum analysis in p53-mutant mice. *Curr Biol* 4(1):1–7
7. Singh VP, Lakshmi BJ, Singh S, Shah V, Goel S, Sarathi DP et al. Lack of Wdr13 gene in mice leads to enhanced pancreatic beta cell proliferation, hyperinsulinemia and mild obesity. *PLoS One*. 2012;7
8. Singh VP, Gurunathan C, Singh S, Singh B, Lakshmi BJ, Mishra AP et al. Genetic deletion of Wdr13 improves the metabolic phenotype of Lepr (db/db) mice by modulating AP1 and PPARgamma target genes. *Diabetologia*. 2015;58
9. Mishra AP, Yedella K, Lakshmi JB, Siva AB (2018) Wdr13 and streptozotocin-induced diabetes. *Nutr Diabetes* 8(1):57
10. Mishra AP, Siva AB, Gurunathan C, Komala Y, Lakshmi BJ (2020) Impaired liver regeneration and lipid homeostasis in CCl4 treated WDR13 deficient mice. *Lab Anim Res* 36(1):41
11. Singh VP, Lakshmi BJ, Singh S, Shah V, Goel S, Sarathi DP et al (2012) Lack of Wdr13 Gene in Mice Leads to Enhanced Pancreatic Beta Cell Proliferation, Hyperinsulinemia and Mild Obesity. *PLoS One* 7(6):1–10
12. Singh VP, Katta S, Kumar S (2017) WD-repeat protein WDR13 is a novel transcriptional regulator of c-Jun and modulates intestinal homeostasis in mice. *BMC Cancer* 17(1):148
13. Singh S, Pavuluri S, Jyothi Lakshmi B, Biswa BB, Venkatachalam B, Tripura C et al (2020) Molecular characterization of Wdr13 knockout female mice uteri: a model for human endometrial hyperplasia. *Sci Rep* 10(1):14621
14. Nagamine M, Okumura T, Tanno S, Sawamukai M, Motomura W, Takahashi N et al (2003) PPAR gamma ligand-induced apoptosis through a p53-dependent mechanism in human gastric cancer cells. *Cancer Sci* 94(4):338–343

15. Prost S, Relouzat F, Spentchian M, Ouzegdouh Y, Saliba J, Massonnet G et al (2015) Erosion of the chronic myeloid leukaemia stem cell pool by PPAR γ agonists. *Nature* 525(7569):380–383
16. Elrod H, Sun SY. PPAR γ and apoptosis in cancer. *PPAR Res.* 2008;2008

Figures

Figure 1

Kaplan Miere survival graph of all genotypes. Survival of Double Knockout (DKO) mice is significantly ($p<0.0001$, Log rank Mantel-Cox Test) higher than the p53^{-/-} mice.