RESEARCH HIGHLIGHT

Cell Research (2014) 24:1284-1285.

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Expanding the functional role of long noncoding RNAs

Cell Research (2014) **24**:1284-1285. doi:10.1038/cr.2014.104; published online 8 August 2014

New findings bring to light a previously unappreciated mechanism involved in the regulation of the oncoprotein MYC. Interesting observations find that the long noncoding RNA (lncRNA) *PVT1* is active in controlling levels of MYC through regulation of MYC protein stability.

The oncogene *MYC* is localized in the chromosomal region 8q24.21, a region frequently amplified in many human tumors. In addition to *MYC*, this locus also hosts the *GSDMC* and *CCDC26* genes as well as the lncRNA *PVT1*. A gain of function for all of these genes is observed in many tumors along with *MYC*.

In the paper by Tseng *et al*. [1], the authors investigate whether low copy number gain of one, or more, of the genes on 8q24.21 promotes cancer. In human breast and ovarian cancers, gain of 8q24.21 is often accompanied by *ERBB2/HER2* amplification [2, 3]. Therefore, by using the *MMTVneu* transgenic mice, which harbor amplified *Erbb2/Her2*, the authors generated a series of mouse models: *gain(Myc)*, *gain(Pvt1,Ccdc26,Gsdmc)* and *gain(Myc,Pvt1,Ccdc26,Gsdmc)*. The authors observed that single supernumerary of *gain(Myc)* and *gain(Pvt1,Ccdc26,Gsdmc)* was not sufficient to promote cancer in the *MMTVneu* mouse model. In contrast, *gain(Myc,Pvt1,Ccdc26,Gsdmc)* showed shorter mammary tumor latency and increased penetrance compared to the other genotypes, suggesting that other genetic elements, not only *Myc*, within the 8q24.21 region, are involved in cancer progression. The authors excluded *Gsdmc* and *Ccdc26* from this process and focused on the functional role of the lncRNA *PVT1*.

To elaborate their findings, the studies were expanded to human breast cancer cell lines, which harbor high copy gains of 8q24.21. Transcriptional activity of the two genes did not appear linked since knockdown of either *PVT1* or *MYC* did not affect the mRNA expression of one another. However, cellular proliferation was decreased upon knockdown of *MYC* as well as *PVT1* and knockdown of both *MYC* and *PVT1* did not further reduce cellular proliferation, thus suggesting that *MYC* and *PVT1* may act in the same pathway.

Although changes in expression of *PVT1* had no effect on *MYC* mRNA expression, it was noted that high levels of *PVT1* correlated with increased levels MYC protein and this effect appeared to be a consequence of increased stability of the MYC protein. It is well established that MYC protein degradation is promoted by phosphorylation of threonine 58 (Thr58) (Figure 1A) [4]. The authors speculated and observed that *PVT1* protects the MYC protein from phosphorylation-mediated degradation (Figure 1B).

In order to further dissect the mechanism of action, Tseng *et al*. [1] showed that MYC and *PVT1* co-localize in the nucleus. Moreover, RNA immunoprecipitation of MYC supported the notion that MYC and *PVT1* are indeed part of the same complex. In addition, the authors applied the CRISPR method to the MYC-driven colon cancer cell line HCT116 and generated *PVT1*-null cells (ΔPVT1-HCT116). In support of their findings, the ΔPVT1-HCT116 cells

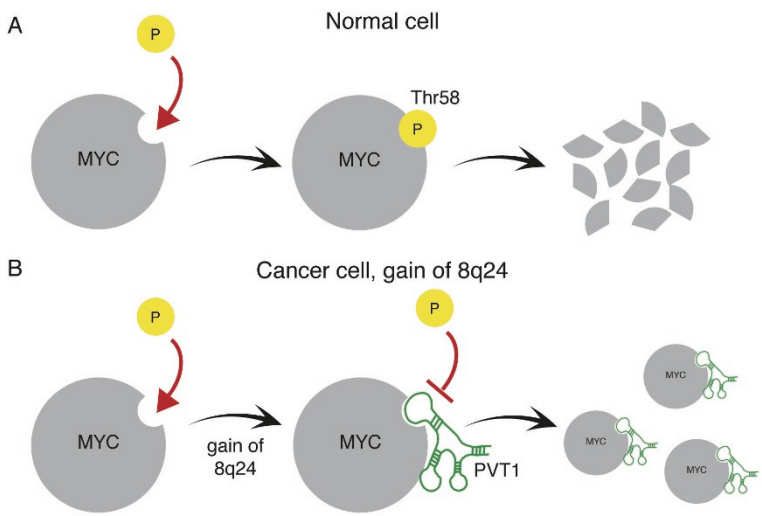

Figure 1 (A) In normal cells, MYC is targeted by phosphorylation on threonine 58 (Thr58) and becomes destabilized and degraded. **(B)** In cancer cells, gain of 8q24 promotes the expression of MYC and *PVT1*. *PVT1* interferes with the phosphorylation of Thr58 on MYC, which stabilizes the protein and increases its level.

demonstrated reduced proliferation and impaired colony formation in soft agar, and xenograft studies showed either failed tumor formation or reduced volume compared to the parental HCT116 cells. Finally, using publically available databases, the authors found that > 97% of tumors with increased 8q24 copy number had increased copy number of both *MYC* and *PVT1* genes.

In summary, this exciting study provides convincing data that *PVT1* is an important regulatory lncRNA, which is involved in modulating the phosphorylation of MYC and cancer progression. The findings presented within this study are in line with previous reports suggesting that phosphorylation of STAT3 is regulated by a lncRNA, lnc-DC [5].

While the functional investigation of lncRNAs has long focused on chromatin remodeling [6], a role beyond chromatin is now emerging. The intriguing studies carried out by Tseng *et al*. [1] suggest that several questions remain to be addressed. It would indeed be of great interest to precisely investigate how *PVT1* is interfering with the phosphorylation of MYC. For instance, 1) Is there a specific domain of *PVT1*, which is responsible for the action [7]? 2) Does *PVT1* form direct interactions with MYC thereby blocking the phosphorylation of Thr58? And 3) Does *PVT1* interact with other proteins except MYC? By addressing these questions in future studies, it could be possible to identify regulatory sequences within *PVT1*. This would not only allow for better understanding of the interplay between *PVT1* and MYC, but could also lead to the identification of conserved regions and RNA structures. Such structures might also be present in other lncRNAs with similar functions and serve as the basis of potentially new therapeutics to disrupt such interactions and affect cancer progression.

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References

1 Tseng YY, Moriarity BS, Gong W, *et al*. *Nature* 2014; **512**:82-86.
2 Park K, Kwak K, Kim J, *et al*. *Hum Pathol* 2005; **36**:634-639.
3 Al-Kuraya K, Schraml P, Torhorst J, *et al*. *Cancer Res* 2004; **64**:8534-8540.
4 Yeh E, Cunningham M, Arnold H, *et al*. *Nat Cell Biol* 2004; **6**:308-318.
5 Wang P, Xue Y, Han Y, *et al*. *Science* 2014; **344**:310-313.
6 Johnsson P, Ackley A, Vidarsdottir L, *et al*. *Nat Struct Mol Biol* 2013; **20**:440-446.
7 Johnsson P, Lipovich L, Grandér D, *et al*. *Biochim Biophys Acta* 2014; **1840**:1063-1071.