DOTTORATO DI RICERCA IN BIOLOGIA CELLULARE E DELLO SVILUPPO

Proposta di assegnazione di una borsa di Dottorato ad una linea di ricerca

Titolo della ricerca: Ruolo delle Demetilasi degli istoni KDM5 nel Cancro del seno e nel melanoma. - Role of the histone demethylases KDM5 in breast cancer and melanoma

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DESCRIZIONE DELLA RICERCA

Histone lysine methylation is a post-translational modification that influences many aspects of cell biology, such as control of transcription, epigenetic inheritance, nuclear architecture and genome stability (1,2). Unlike histone acetylation, histone methylation may lead either to transcriptional repression or activation, depending on which residue is involved. Due to the importance of this epigenetic mark, a tight regulation of histone methylation has been evolved. The enzyme capable of erase methyl groups from histones are the histone lysine demethylase (KDMs) and in human these enzymes are represented by two families of proteins: the Lysine Specific histone Demethylase (LSD) family and the JmjC domain-containing family also known as the Jumonji Histone Demethylases (JHDMs) (3). These two families differ in their catalytic mechanisms. The LSD KDMs are monoamine oxidases whereas JHDMs are hydroxylases that require two cofactors for their function, Fe(II) and 2-oxoglutarate, which are bound in the JMJC catalytic domain.
Among the latter ones, particularly interesting are those using H3K4me2 and H3K4me3 as substrates, the KDM5 (or JARID1) enzymes. H3K4 methylation seems to play an important role in development and differentiation and transcriptional regulation (4,5).
Indeed, actively transcribed genes have promoters often marked with H3K4 tri-methylation (6). Therefore, at first glance, KDM5 enzymes seem to act as transcriptional repressors but, recently, it has been proposed that KDM5 demethylases could also remove tri- and di-methyl groups at enhancer regions. Since H3K4me1 modification combined with acetylated H3K27 (H3K27ac) is predictive of active enhancers, KDM5 enzymes could also have a role in activation transcription (7). KDM5 subfamily (also known as JARID1 subfamily) consists of four members, KDM5A (JARID1A), KDM5B (JARID1B), KDM5C (JARID1C) and KDM5D (JARID1D) whose deregulation in various cancers has been widely documented to contribute significantly to tumor initiation and progression (8-11).
Indeed, KDM5B was initially identified as a gene markedly over-expressed in breast cancer even before the discovery of histone lysine demethylases. Nevertheless, it was later observed that the deregulation of this protein is different among the different breast cancer subtypes thus suggesting a crucial but ambiguous role for KDM5B in breast carcinoma depending on the cell type context (12). Later on, it was also found to be over-expressed in prostate, lung and bladder carcinoma (9,10).
Beside the role in transcriptional regulation, new findings suggest that KDM5 KDMs are involved in the mechanisms of maintenance of genomic stability. In particular, KMD5B is enriched in DNA-damage sites after ionizing radiation and its demethylase activity is required
for an efficient DNA repair, in contrast with previous observations suggesting a positive role for H3K4 methylation in DNA repair. An interesting model proposed by Li and

colleagues (13) tries to define a trait d'union between the different roles of KDM5B in transcriptional regulation and in DNA repair. During transcriptional activation, PARP1 PARlylates KDM5B and prevents it from demethylating H3K4 and shutting off transcription. However, when chromatin is damaged, PARylated KDM5B can be recruited to the damaged sites by histone variant macroH2A1.1 thanks to its PAR binding domain. Local H3K4 demethylation performed by KDM5B is essential for Ku70/80 and BRCA1 recruitment, respectively in NHEJ and HR pathways. A similar mechanism has been recently proposed for KDM5A as well, even though it is still not clear whether the catalytic activity of this enzyme is necessary for DNA repair or in this case the repair is achieved by an indirect mechanism (14).
For the pivotal functions of KDM5 enzymes in different cellular processes, it is important to understand the mechanisms underlying their regulation, Moreover, given the prominent role they have in oncogenesis, they are also candidate therapeutic targets.
In this program we propose:
a - To use different chemical inhibitors of KDM5 catalytic activity and to test their effects on transcriptome, cell survival and DNA damage repair in breast cancer derived cell lines.
b - To study the functional effects of two different miRNAs which, as we previously showed, target JARID 1B mRNA and significantly modulate its abundance in breast cancer cells.
c - To extend these analyses to melanoma cell lines which over-express JARID1B.

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2 - Mocavini I, Pippa S, Licursi V, Paci P, Trisciuoglio D, Mannironi C, Presutti C, Negri R (2019). *JARID1B expression and its function in DNA damage repair are tightly regulated by miRNAs in breast cancer.* **Cancer Science**, ISSN: 1347-9032, doi: 10.1111/cas.13925
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