Atlas of Science
another view on science
http://atlasofscience.org

How does phenobarbital, a therapeutic sedative, activate a nuclear receptor through phosphorylation?

Steroid hormones activate a family of nuclear proteins called nuclear receptors to regulate every aspect of human physiology, reproduction, development, differentiation, metabolism and immunity. A nuclear receptor molecule is comprised of two major structural domains; DNA-binding (DBD) and ligand-binding (LBD). For example, estrogen directly binding the LBD to activate estrogen receptor is essential for female reproduction. Among the 46 nuclear receptors in humans, there is a unique group of nuclear receptors that are activated by therapeutic drugs and environmental pollutants, the so-called nuclear drug/xenobiotics-activated receptors. Upon drug administration, these nuclear receptors activate genes that encode enzymes and transporters in livers, increasing the hepatic capability of drug metabolism. One such nuclear receptor, constitutive active/androstane receptor (CAR), is activated by the sedative phenobarbital, which is widely used to treat epileptic patients. In response to phenobarbital, CAR binds and activates its target genes, resulting in an increase of hepatic metabolism and resistance to phenobarbital in humans and mice as well as promoting liver tumor development in mice. In stark contrast to the direct binding of estrogen to estrogen receptor, phenobarbital indirectly activates CAR. The recent discovery of this indirect activation mechanism resolved a 50 years-old mystery of how phenobarbital induces drug metabolism.

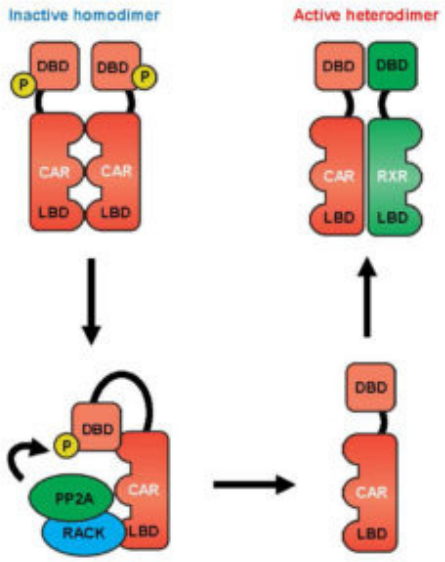

Fig. 1.

Phosphorylation modifies protein by binding a phosphoryl group $(PO_3)^-$ and regulates protein function. Nuclear receptors contain highly conserved phosphorylation motif within their DBDs.

This phosphorylation has largely been forgotten in nuclear receptor research, since it resides in the DBD. For most, the popular notion that nuclear receptors must be regulated by direct binding such as estrogen binding to the LBD of estrogen receptor has sidelined phosphorylation-related research. CAR conserves this

phosphorylation motif at threonine 38. Phenobarbital, a therapeutic sedative, utilizes it to indirectly regulate CAR activation. Phenobarbital initiates this indirect activation by directly binding to a cell membrane receptor (*e.g.* insulin receptor) to transduce its activation signal to CAR.

CAR phosphorylated at threonine 38 forms an inactive homodimer. In response to the phenobarbital-initiated signal, the phosphorylated CAR homodimer dissociates and the DBD interacts with the LBD, preventing its monomer from going back to the homodimer. Subsequently, protein phosphatase 2A binds the phosphorylated monomer to dephosphorylate it. Dephosphorylated CAR heterodimerizes with retinoid X receptor (RXR) through a surface opposite from the surface for homodimerization to activate target genes in the nucleus. Therefore, two different surfaces of the LBD and phosphorylation within the DBD integrate CAR response to phenobarbital for indirect activation. This indirect activation mechanism is novel and is a marked departure from the direct binding activation of the estrogen receptor by estrogen.

As a nuclear receptor functions in a dimer, two different surfaces are now established for both homo- and hetero-dimerization. Moreover, phosphorylation within the DBD, which is conserved in 41 out of total 46 human nuclear receptors, regulates dimerization. Imagine 41 different nuclear receptors combining multiple forms of dimerization and conserved phosphorylation to connect regulatory networks. Such combinatorial possibilities would result in tremendous functional diversification.

Masahiko Negishi
Pharmacogenetics section, Reproductive and Developmental Biology Laboratory,
National Institute of Environmental Health Sciences,
National Institutes of Health, Research Triangle Park, North Carolina, USA

Publication

Interaction of the phosphorylated DNA-binding domain in nuclear receptor CAR with its ligand-binding domain regulates CAR activation.
Shizu R, Min J, Sobhany M, Pedersen LC, Mutoh S, Negishi M
J Biol Chem. 2018 Jan 5