

Enhancement by a Src family inhibitor of the interaction of Pyk2 and Fyn in hypothalamic neurons

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The receptor for gonadotropin-releasing hormone (GnRH) is highly expressed in hypothalamic GnRH neurons, as well as in anterior pituitary gonadotrophs. In our previous study, we found that stimulation of the GnRH receptor activated protein kinase D1 (PKD1), and PKD1 was involved in the Fyn-mediated activation of proline-rich tyrosine kinase 2 (Pyk2) in cultured GnRH neurons (GT1-7 cells). In the present study, we examined the molecular mechanisms of Pyk2 activation and the interaction of Pyk2 and Fyn in GT1-7 cells. Experiments with site-directed mutants of Pyk2 indicated that tyrosine 402 (Tyr402) was phosphorylated both by autophosphorylation and by Fyn, whereas Tyr579 was phosphorylated exclusively by Fyn. We found that dasatinib, a Src family inhibitor, enhanced the interaction of Pyk2 and Fyn. Experiments with site-directed mutants of Pyk2 and Fyn indicated that dasatinib enhanced the binding of Pyk2 autophosphorylated at Tyr402 and the Src homology 2 domain in Fyn. Our present data may suggest that fully activated Pyk2 dissociates from Fyn after Fyn-mediated phosphorylation of Pyk2 at sites other than Tyr402 and Tyr579.