THE KINASE SUPPRESSOR OF RAS (KSR-1) IS REQUIRED FOR DIFFERENTIATION OF HL60 CELLS INDUCED BY NEAR-PHYSIOLOGICAL LEVELS OF 1,25 DIHYDROXYVITAMIN D₃

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The activity of KSR-1, a kinase or a molecular scaffold upstream from Raf-1, is involved in the MEK/ERK MAP kinase cascade, which signals cell growth, survival or differentiation, depending on the cellular context. This laboratory has previously shown that in human leukemia HL60 cells, both the phosphorylation of the Raf-1 protein by KSR-associated kinase activity, and the monocytic differentiation induced by 1,25-dihydroxyvitamin D₃ (1,25D₃), are inhibited by the pharmacological inhibitors PD98059 and U0126 [Wang and Studzinski, Exp. Cell Res. 268 (2001) 294]. Here, we provide evidence that KSR is upregulated in HL60 cells undergoing 1,25D₃-induced differentiation, and an antisense oligo, but not a sense oligo, to KSR inhibits this differentiation. The inhibition of differentiation is paralleled by the reduced phosphorylation of RAF-1 Ser 259. Conversely, ectopic expression of FLAG-wild type KSR potentiates the differentiation-inducing effects of low concentrations of 1,25D₃. Additional preliminary data indicate that the kinase activity of KSR is required for these effects, as a transfection of a kinase inactive (ki) construct of KSR did not significantly increase the 1,25D₃-induced differentiation. Also, the 1,25D₃-induced G1 cell cycle block was potentiated by the wild type, but not the ki-KSR construct. Taken together, these data suggest that KSR-1 participates in the signaling of monocytic differentiation through the phosphorylation of Raf-1, and that agents that enhance KSR expression may potentiate the effects of anti-cancer drugs by reducing the proliferative rate of malignant cells. Supported by grant R01 CA-44722 from the National Cancer Institute.