A Coiled-coil Tetramerization Domain of BCR-ABL Is Essential for the Interactions of SH2-containing Signal Transduction Molecules*

(Received for publication, January 29, 1996, and in revised form, July 23, 1996)

Tetsu Tsuchi†‡, Keisuke Miyazawa‡, Gen-Sheng Feng†, Hal E. Broxmeyer*†‡¶, and Keisuke Toyama†

From the †First Department of Internal Medicine, Tokyo Medical College, Tokyo, Japan, the Departments of *Biochemistry and Molecular Biology, **Medicine (Hematology/Oncology), ‡Microbiology and Immunology, and the †Walther Oncology Center, Indiana University School of Medicine, Indianapolis, Indiana 46202

BCR-ABL is a chimeric oncoprotein that exhibits deregulated tyrosine kinase activity and is implicated in the pathogenesis of Philadelphia chromosome (Ph1)-positive leukemia. We have previously shown SH2-containing phosphotyrosine phosphatase SHP-2 forms stable complexes with BCR-ABL and Grb2 in BCR-ABL-transformed cells (Tsuchi, T., Feng, G. S., Shen, R., Song, H. Y., Donner, D., Pawson, T., and Broxmeyer, H. E. (1994) J. Biol. Chem. 269, 15381–15387). To elucidate the structural requirement of BCR-ABL for the interactions with SH2-containing signaling molecules, we examined a series of BCR-ABL mutants which include the Grb2 binding site-deleted BCR-ABL (1–63 BCR/ABL), the tetramerization domain-deleted BCR-ABL (64–509 BCR/ABL), and the SH2 domain-deleted BCR-ABL (BCR/ABL ΔSH2). These BCR-ABL mutants were previously shown to reduce the transforming activity in fibroblasts. We found that the tetramerization domain-deleted BCR-ABL did not induce the tyrosine phosphorylation of SHP-2 and the interactions of BCR-ABL, SHP-2, and Grb2. In vitro kinase assays have also shown that the tetramerization domain-deleted BCR-ABL mutant did not phosphorylate GST-SHP-2 in vitro. SHP-2 was co-immunoprecipitated with phosphatidylinositol 3-kinase in BCR/ABL p210-transformed cells; however, this interaction was not observed in the tetramerization domain-deleted BCR-ABL mutant. Therefore the tetramerization domain of BCR-ABL is essential for interactions of these downstream molecules.

The Philadelphia chromosome (Ph1) is one of the best known cytogenetic abnormalities in human hematologic malignancy (1, 2). It is fundamentally characteristic of chronic myelogenous leukemia, but it also is seen in many cases of acute lymphoblastic leukemia and occasionally in acute myelogenous leukemia. The molecular basis of the Ph1 chromosome involves the reciprocal translocation between the BCR gene on chromosome 22 and c-ABL on chromosome 9 (3). The normal cellular function of the BCR gene is unknown, but c-ABL is the cellular homologue of the transforming gene of Abelson leukemia virus and belongs to the broad family of nonreceptor protein kinases (4). The chimeric fusion gene (BCR-ABL) results in truncation of c-ABL at its amino-terminal exon by BCR sequences, leading to a fusion protein (BCR-ABL) with deregulated tyrosine kinase activities (5, 6). BCR-ABL has been shown to transform fibroblast and hematopoietic cells in vitro (7, 8) and to cause a chronic myelogenous leukemia-like disease in mice (9). Thus, BCR-ABL has been linked as a decisive inciting factor in the pathogenesis of chronic myelogenous leukemia.

The connection between the BCR-ABL tyrosine kinase and downstream signal transduction pathways is provided by multiple functional domains (10–18). The first 63 amino acids of BCR contain a coiled-coil tetramerization domain which activates the tyrosine kinase as well as the F-actin binding function of BCR-ABL (10, 11). The second BCR region, between amino acids 176 and 242, contains a tyrosine residue at position 177, which becomes phosphorylated on tyrosine and binds to the SH2 domain of the adapter protein Grb2 (12, 13). Mutation of tyrosine 177 to phenylalanine abolishes Grb2 binding and blocks hematopoietic transformation (12). Three regions in ABL, the SH2 domain, the tyrosine kinase domain, and the F-actin binding domain, are required for transformation of fibroblasts (14).

Previously, we have shown that SHP-2, a phosphotyrosine phosphatase widely expressed in all the tissues in mammals, complexes with BCR-ABL and Grb2 in BCR-ABL-transformed cells (19). In the present study we examined the structural requirement of BCR-ABL for interaction with SH2-containing signal transduction molecules. It was found that the tetramerization-deleted BCR-ABL lacked both tyrosine phosphorylation of SHP-2 and the association with SHP-2 and Grb2. SHP-2 co-immunoprecipitates with the p85 subunit of PI 3-kinase in BCR/ABL p210-expressing cells; however, this interaction was not observed in the tetramerization domain-deleted BCR-ABL transfected cells. These results suggest that the tetramerization domain of BCR-ABL is essential for interactions of these downstream signaling molecules and that these molecular interactions are involved in the transformation of BCR-ABL.

MATERIALS AND METHODS

Antibodies—Polyclonal anti-SHP-2 antibody (raised against a GST fusion protein containing the SHP-2 residues 2–216) was as described previously (19). Anti-phosphotyrosine mAb (PY20), anti-Grb2 mAb (GR81), anti-SHP-2 mAb, and affinity-purified anti-PI 3-kinase (p85) antibodies were obtained from Transduction Laboratories (Lexington, KY). Affinity-purified anti-Grb2 (raised against a peptide corresponding to residues 195–217 mapping at the carboxyl terminus of human Grb2) and GST-Grb2 fusion protein (corresponding to amino acids 1–217 of Grb2 of murine origin) and anti-GST mAb were obtained from

*This work was supported by United States Public Health Service Grants RO1 HL54037, R01 CA36464, R01 HL48549, and R01 HL49202, by a grant-in-aid from the Ministry of Education, Science and Culture of Japan (to T. T.). The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

† To whom correspondence should be addressed: First Dept. of Internal Medicine, Tokyo Medical College, Tokyo Medical College, Tokyo, Japan, the Departments of Biochemistry and Molecular Biology, Medicine (Hematology/Oncology), Microbiology and Immunology.

The abbreviations used are: PI 3-kinase, phosphatidylinositol 3-kinase; GST, glutathione S-transferase; mAb, monoclonal antibody.
BCR-ABL in SH2-containing Signal Transduction Molecules

Santa Cruz Biotechnology, Inc. (Santa Cruz, CA). Anti-phosphotyrosine mAb (4G10) was obtained from Upstate Biotechnology Inc. (Lake Placid, NY). Anti-ABL mAb (24–21) was purchased from Oncogene Science Inc. (Cambridge, MA).

Cells and Culture—The NSF/N1.17 cell line was obtained from Dr. H. Scott Bovill (Indiana University School of Medicine, Indianapolis, IN) and has been described elsewhere (20). This cell line was cultured in McCoy’s 5A modified medium (Life Technologies, Inc.) supplemented with 15% fetal calf serum (Hyclone Laboratories, Logan, UT) and 15% WEHI-3 cell line conditioned media.

Plasmid Constructs—Expression plasmids used in this study were as previously described (11, 14, 18). The p210 coding fragment was cleaved from pSp65BCR-ABLp210 (21) and cloned into the retroviral vector pSLXCMV (11).

DNA Transfection—NSF/N1.17 cells were transfected by electroporation as described previously (22). Beginning 48 h after electroporation, cells were selected with G418 (Life Technologies, Inc.). Within 10 days of plating in the selection medium, G418-resistant sublines were obtained.

Immunoblotting and Immunoprecipitation—Immunoblotting and immunoprecipitation were performed as described previously (22).

In Vitro Binding Assays—GST-SHP-2 SH2 (amino acids 2–216) fusion protein was freshly prepared for the in vitro binding experiments (19). To dissociate and denature the preexisting protein complexes, cells were lysed in a small volume (105 cells in 100 μl) of lysis buffer (20 mM Tris-HCl, pH 7.4, 150 mM NaCl, 10% glycerol, 0.2% Triton X-100 (Bio-Rad), 1% SDS, 1 mM phenylmethylsulfonyl fluoride, 0.15 unit/ml aprotinin, 10 mg/ml leupeptin, 100 mM sodium fluoride, and 2 mM sodium orthovanadate). After centrifugation, the supernatants were heated to 95 °C for 5 min, cooled on ice, and then diluted with Triton X-100 lysin buffer to 1 ml so that the final concentration of SDS was 0.1%. In vitro binding assays, glutathione-Sepharose beads (Pharmacia Biotech Inc.) with bound fusion proteins (approximately 5 μg of fusion protein per binding reaction) were incubated in denatured proteins at 4 °C for 90 min. The beads were washed four times with TNGN washing buffer (20 mM Tris-HCl, pH 7.4, 150 mM NaCl, 10% glycerol, 0.1% Triton X-100 (Bio-Rad), 100 mM sodium fluoride, and 2 mM sodium orthovanadate) before analysis.

In Vitro Kinase Assays—For immune complex kinase assays, immunoprecipitated proteins were wash three times with TNGN washing buffer and one time with kinase buffer (50 mM Tris-HCl, pH 7.4, 10 mM MnCl2, 1 mM dithiothreitol). The resulting complexes were re-suspended in 30 μl of kinase buffer supplemented with 2 μg of GST-SHP-2 fusion protein and 10 μCi of [32P] (Amer sham Corp.) and incubated for 5 min at room temperature. Samples were boiled for 5 min after addition of 30 μl of SDS-sample buffer, separated by SDS-polyacrylamide gel electrophoresis, and transferred to membranes for autoradiography.

RESULTS AND DISCUSSION

A Coiled-coil Teramerization Domain of BCR Is Required for the Interactions of SHP-2 and BCR-ABL—The structures of the different BCR-ABL fusion constructs are shown in Fig. 1. Deletions of the coiled-coil tetramerization domain (64–509 BCR/ABL), the second BCR region including the Grb2 binding site (1–63 BCR/ABL), and the SH2 domain (BCR/ΔSH2 ABL) have all been shown to reduce BCR-ABL transformation in rodent fibroblast cells (14). Furthermore, these BCR-ABL mutant transfected cells, except 64–509 BCR/ABL-transfected cells, were growth factor-independent for proliferation (data not shown). These mutants were chosen to study the structural requirements for the interaction between the SH2-containing signaling molecules and BCR-ABL. These BCR-ABL transfected cell lines were shown to express similar amounts of SHP-2 protein (Fig. 3A). Cell lysates were incubated with anti-SHP-2 antibodies and precipitated proteins were analyzed by immunoblotting with anti-phosphotyrosine mAbs (Fig. 3B). SHP-2 was phosphorylated on tyrosine in all of the BCR-ABL transfected cells except for the 64–509 BCR/ABL mutant (Fig. 3B).

To determine the functional domains of BCR-ABL that are responsible for the interaction with SHP-2, cells were lysed and heat-denatured at 95 °C in the presence of SDS. These lysates were incubated with GST-SHP-2 SH2 fusion proteins immobilized on glutathione beads. The unbound proteins were removed and immunoblotted with anti-ABL mAb (Fig. 3C). SHP-2 was able to bind to all of the BCR-ABL proteins except for the 64–509 BCR/ABL mutant. These results demonstrate that the coiled-coil tetramerization domain of BCR-ABL is required for the interaction between SHP-2 and BCR-ABL.

Multiple Signaling Pathways from BCR-ABL to Ras—Potential mediators of BCR-ABL-induced Ras activation in hematopoietic cells include the Shc adapter proteins and SHP-2 phosphatase (13, 17–19, 22). Both Shc-Grb2 and SHP-2-Grb2 complexes have been implicated in epidermal growth factor- or platelet-derived growth factor-stimulated Ras activation (23–25). We have previously shown that BCR-ABL co-immunoprecipitated with Shc, Grb2, and SHP-2 in BCR-ABL-transformed...
cells (19, 22). To examine the interactions of BCR-ABL, Grb2, and SHP-2, we utilized co-immunoprecipitation and in vitro binding assays. Cell lysates were immunoprecipitated with anti-Grb2 and then immunoblotted with anti-ABL. The background band around 50 kDa is the immunoglobulin heavy chain from the immunoprecipitation. B, equal amounts of total proteins from the indicated cell lines were immunoprecipitated (IP) with anti-ABL, and these immune complexes were subjected to immunoblot analysis with anti-phosphotyrosine mAb (4G10 + PY20). Similar results were obtained in two independent experiments.

Fig. 2. Autophosphorylation of BCR-ABL proteins in vivo. A, cell lysates from the indicated cell lines were immunoprecipitated (IP) with anti-ABL or mouse immunoglobulins as a control (C, as noted for immunoprecipitation). Immunoprecipitates were separated by 7.5% SDS-polyacrylamide gel and immunoblotted with anti-ABL. The background band around 50 kDa is the immunoglobulin heavy chain from the immunoprecipitation. B, equal amounts of total proteins from the indicated cell lines were immunoprecipitated (IP) with anti-ABL, and these immune complexes were subjected to immunoblot analysis with anti-phosphotyrosine mAb (4G10 + PY20). Similar results were obtained in two independent experiments.

BCR-ABL in SH2-containing Signal Transduction Molecules

Fig. 3. A coiled-coil tetramerization domain of BCR is required for the interaction between SHP-2 and BCR-ABL. A, cell lysates from each cell lines were immunoprecipitated (IP) with polyclonal anti-SHP-2 antibodies or rabbit immunoglobulin (IgG), and immunoblotted (BLOT) with anti-SHP-2 mAb. B, cell lysates from indicated cell lines were immunoprecipitated with polyclonal anti-SHP-2 and immunoblotted with anti-phosphotyrosine mAbs (4G10 + PY20). C, cell lysates from indicated cell lines were denatured by heating at 95 °C for 5 min in the presence of 1% SDS prior to incubation with immobilized GST-SHP-2 SH2 fusion proteins or GST alone. Samples were analyzed by 7.5% SDS-polyacrylamide gel and immunoblotted with anti-ABL. Similar results were obtained in each of two separate experiments.
similar experiment, Grb2 was detected in anti-SHP-2 immunoprecipitates from 1–63 BCR/ABL, 1–509 BCR/ABL, and BCR/ABL p210-expressing cells but not from 64–509 BCR/ABL-expressing cells. To obtain an overview of complexes formed in 1–63 BCR/ABL-expressing cells or in BCR/ABL p210-expressing cells, cell lysates were immunoprecipitated with anti-Grb2, and these immune complexes were analyzed by immunoblotting with anti-phosphotyrosine (Fig. 4D, left). Grb2 formed a specific complex with BCR/ABL and 70-kDa tyrosine-phosphorylated protein in 1–63 BCR/ABL and BCR/ABL p210-expressing cells (Fig. 4D, left). To test whether Grb2 can directly bind to SHP-2 and whether the 70-kDa Grb2-bound tyrosine-phosphorylated protein corresponds to SHP-2, protein complexes in the cell lysates were dissociated and denatured by 95°C heat and SDS treatment prior to GST-Grb2 binding assays. The bound proteins were analyzed by immunoblotting with anti-SHP-2 (Fig. 4D, right). Grb2 was still able to bind to SHP-2, which corresponds to the 70-kDa tyrosine-phosphorylated protein, in 1–63 BCR/ABL and BCR/ABL p210-expressing cells (Fig. 4D, right). In addition to direct interaction of Grb2 and BCR-ABL shown in Fig. 4, A and B, these results suggest that Grb2 indirectly associates with BCR-ABL through SHP-2.

**Co-immunoprecipitation of SHP-2 and PI 3-kinase in BCR-ABL-transformed Cells**—Recent studies have shown that the SH3 domains of Grb2 directly bind to the proline-rich motifs of PI 3-kinase p85 subunit (26). Since Grb2 also associates with SHP-2 in BCR-ABL-transformed cells (Fig. 4C), we wished to evaluate possible interactions of SHP-2, Grb2, and PI 3-kinase. Cell lysates were immunoprecipitated with anti-PI 3-kinase (p85) antibody, and these immune complexes were analyzed by immunoblotting with anti-SHP-2 (Fig. 5A). SHP-2 could be detected in anti-PI 3-kinase (p85) immunoprecipitates from 1–63 BCR/ABL, 1–509 BCR/ABL, and BCR/ABL p210-expressing cells but not from 64–509 BCR/ABL-expressing cells. In a similar experiment, PI 3-kinase (p85) could be detected in anti-SHP-2 immunoprecipitates from 1–63 BCR/ABL, 1–509 BCR/ABL, and BCR/ABL p210-expressing cells but not from 64–509 BCR/ABL-expressing cells. These results demonstrate that SHP-2 and PI 3-kinase (p85) form physiological complexes in 1–63 BCR/ABL, 1–509 BCR/ABL, and BCR/ABL p210-expressing cells; however, this interaction was not observed in 64–509 BCR/ABL-expressing cells.

Cell lysates were immunoprecipitated with anti-PI 3-kinase (p85) and then immunoblotted with anti-Grb2 (Fig. 5C). Grb2 was detected in anti-PI 3-kinase (p85) immunoprecipitates
from all cell lines (Fig. 5C). To test whether Grb2 directly binds to PI 3-kinase (p85) in BCR-ABL-expressing cells, cell lysates were dissociated and denatured by 95 °C heat and SDS treatment prior to GST-Grb2 binding assays, and these complexes were analyzed by immunoblotting with anti-PI 3-kinase (p85) (Fig. 5D). Grb2 was still able to bind to PI 3-kinase (p85) in these conditions (Fig. 5D). These data suggest that Grb2 mediates the complex formations of SHP-2 and PI 3-kinase.

In Vitro Phosphorylation of GST-SHP-2 by BCR-ABL Tyrosine Kinase—SHP-2 is constitutively phosphorylated on tyrosine in BCR-ABL-expressing cells except in the 64–509 BCR/ABL mutant-expressing cells (Fig. 3B). It is possible that the association of SHP-2 with BCR-ABL would result in an increased tyrosine phosphorylation of SHP-2. To test this hypothesis, cell lysates were immunoprecipitated with anti-ABL, and these immunoprecipitated proteins were incubated with GST-SHP-2 fusion proteins and [γ-32P]ATP. GST-SHP-2 fusion proteins were collected by glutathione-Sepharose beads and separated by SDS-polyacrylamide gel electrophoresis, and 32P-labeled proteins were detected by autoradiography (Fig. 6A). As a control for equal recovery of GST-SHP-2 fusion proteins in this reaction, the same blot was immunoblotted with anti-GST mAb (Fig. 6B). GST-SHP-2 fusion proteins were phosphorylated in vitro in the cell lysates from 1–63 BCR/ABL, 1–509 BCR/ABL, and BCR/ABL p210-expressing cells but not from 64–509 BCR/ABL-expressing cells (Fig. 6A). These results demonstrate that BCR-ABL is the tyrosine kinase that phosphorylates SHP-2 in vitro.

Activation of the tyrosine kinase and transforming functions of BCR-ABL is dependent upon the fused BCR sequences (27, 28). The BCR sequences alter the subcellular localization of ABL. In BCR-ABL fusion proteins, the fused BCR sequences block nuclear translocation and activate the F-actin-binding function (14, 27). The F-actin-binding function is required for BCR-ABL to efficiently transform cells (14). A tetramerization domain has been identified in the BCR protein (11). This domain coincides within BCR domain 1 (amino acids 1–63) that is essential for transforming activity of BCR-ABL (11). Tetramerization of BCR-ABL correlates with activation of the tyrosine kinase and F-actin binding function of ABL (11). Deletion of the tetramerization domain inactivates the oligomerization function as well as the ability of BCR-ABL to transform rodent fibroblast or to

**Fig. 5. Co-immunoprecipitation of SHP-2 and PI 3-kinase in BCR-ABL-transformed cells.** A, cell lysates from indicated cell lines were immunoprecipitated with affinity-purified anti-PI 3-kinase p85 subunit antibodies and immunoblotted with anti-SHP-2 mAb. B, cell lysates were immunoprecipitated with polyclonal anti-SHP-2 and immunoblotted with affinity-purified anti-PI 3-kinase (p85). C, cell lysates were incubated with anti-PI 3-kinase (p85) and immunoblotted with anti-Grb2 mAb. D, cell lysates were denatured and incubated with GST-Grb2 or GST. These protein complexes were analyzed by anti-PI 3-kinase (p85) immunoblotting. Similar results were obtained in each of two separate experiments.
abrogate interleukin-3 dependence in hematopoietic cells (11).

In the present study, we demonstrate that in the BCR-ABL mutants deletion of the tetramerization domain of BCR did not allow induction of the tyrosine phosphorylation of SHP-2 and the interactions of BCR-ABL, SHP-2, and Grb2. In vitro kinase assays have also shown that the tetramerization domain-deleted BCR-ABL mutant did not phosphorylate GST-SHP-2 in vitro. The tetramerization domain of BCR-ABL activates ABL by inducing intermolecular cross-phosphorylation of ABL tyrosine kinase (11). The resulting phosphorylation of tyrosine residues on BCR-ABL trigger the interactions of these SH2-containing signal transduction molecules. Our identification of the specific amino acid residues involved in the biologically relevant association of BCR-ABL with SH2-containing signaling molecules suggests that this interaction may be a prime candidate for testing the therapeutic utility of peptides to disrupt the transformation capacity of BCR-ABL.

Grb2 has been shown to bind tyrosine 177 within the first exon of BCR-ABL, and mutation of this residue abrogates the ability of BCR-ABL to transform fibroblasts and hematopoietic cells (12, 13). In contrast to a report by others (12), we established a growth factor-independent cell line which expressed a Grb2 binding site-deleted BCR-ABL mutant (1–64 BCR/ABL). Grb2 binding to BCR-ABL via tyrosine 177 is not required for the transformation of the interleukin-3-dependent cell line to growth factor-independent proliferation. We have also shown that Grb2 forms physiological complexes with the Grb2 binding site-deleted BCR-ABL (1–63 BCR/ABL) in vitro. These results allowed us to examine alternative pathways to Ras. SHP-2-Grb2 complexes are present in hematopoietic cells which express the Grb2 binding site-deleted BCR-ABL mutant (1–63 BCR/ABL). SHP-2 is able to bind directly to the 1–63 BCR/ABL mutant. These results suggest that Grb2 indirectly binds to BCR-ABL through SHP-2 and that there are multiple signaling pathways from BCR-ABL to Ras. SHP-2 may act as an alternative pathway from BCR-ABL to Ras.

Acknowledgments—We thank Dr. J. Y. J. Wang (University of California at San Diego, La Jolla, CA) for the generous gifts of reagents.

REFERENCES

1. Bowley, J. D. (1973) Nature 243, 290–293
2. Tauchi, T., and Broxmeyer H. E. (1995) Int. J. Hematol. 61, 105–112
3. Heiterkamp, N., Stamm, R., and Groffen, J. (1985) Nature 315, 758–761
4. Goff, S. P., Gilbea, E., Witte, O. N., and Baltimore, D. (1980) Cell 22, 777–785
5. Konopka, J. B., Watanabe, S. M., and Witte, O. N. (1984) Cell 37, 1035–1042
6. Kloetzner, W., Kurzrock, R., Smith, L., Talpaz, M., Speller, M., Gutterman, J., and Arlinghaus, R. (1985) Virology 140, 230–238
7. Young, J. C., and Witte, O. N. (1988) Mol. Cell. Biol. 8, 4079–4087
8. Lugo, T. G., Pendergast, A. M., and Witte, O. N. (1990) Science 247, 1079–1082
9. Daley, G. Q., Van Etten, R. A., and Baltimore, D. (1990) Science 247, 824–830
10. McWhirter, J. R., and Wang, J. Y. J. (1991) Mol. Cell. Biol. 11, 1553–1565
11. McWhirter, J. R., Galasso, D. L., and Wang, J. Y. J. (1993) Mol. Cell. Biol. 13, 7587–7595
12. Pendergast, A. M., Quilliam, L. A., Cripe, L. D., Bassing, C. H., Dai, Z., Li, N., Batzer, A., Rabun, K. M., Der, C. J., Schlessinger, J., and Gishizky, M. L. (1993) Cell 75, 175–185
13. Pui, L., Liu, J., Gish, G., Mhamalui, G., Bowtell, D., Peiseci, P. G., Arlinghaus, R., and Pawson, T. (1994) EMBO J. 13, 764–773
14. McWirtter, J. R., and Wang, Y. J. J. (1995) EMBO J. 12, 1533–1546
15. Afar, D. E. H., Goga, A., McLaughlin, J., Witte, O. N., and Sawyer, C. L. (1994) Science 264, 424–426
16. Pleshaw, M. W., McWhirter, J. R., and Wang, Y. J. J. (1995) Mol. Cell. Biol. 15, 1296–1299
17. Goga, A., McLaughlin, J., Afar, D. E. H., Saffran, D. C., and Witte, O. N. (1995) Cell 82, 981–988
18. Cortez, D., Kadlec, L., and Pendergast, A. M. (1995) Mol. Cell. Biol. 15, 5531–5541
19. Tauchi, T., Feng, G.-S., Shen, R., Song, H. Y., Donner, D., Pawson, T., and Broxmeyer, H. E. (1994) J. Biol. Chem. 269, 15381–15387
20. Mandaenas, R. A., Leibowitz, D. S., Gharanefaghki, B., Tauchi, T., Burgess, G. S., Miyazawa, R., Jayaram, H. N., and Bowell, H. S. (1993) Blood 82, 1838–1847
21. Feinstein, E., Cimino, G., Gale, R. P., Alнима, M., Berthier, R., Kishi, K., Goldman, J., Zaccaria, A., Berrebi, A., and Canaani, E. (1991) Oncogene 4, 1477–1481
22. Tauchi, T., Bowswell, H. S., Leibowitz, D., and Broxmyeyer, H. E. (1994) J. Exp. Med. 176, 167–175
23. Rosakis-Abeck, M., Pernely, R., Wade, J., Pawson, T., and Bowell, D. (1993) Nature 365, 83–85
24. Li, W., Nishimura, R., Khishibian, A., Batzer, A. G., Kim, W. J. H., Cooper, J. A., and Schlessinger, J. (1994) Mol. Cell. Biol. 14, 509–517
25. Pronk, G. J., DeVries-Smits, A. M. M., Buday, L., Downward, J., Maaassen, J. A., Medema, R. H., and Bos, J. L. (1994) Mol. Cell. Biol. 14, 1575–1581
26. Wang, J., Auger, K. R., Jarvis, L., Shi, Y., and Roberts, T. M. (1995) J. Biol. Chem. 270, 12774–12780
27. McWirtter, J. R., and Wang, Y. J. J. (1991) Mol. Cell. Biol. 11, 1553–1565
28. Muller, A. J., Young, J. C., Pendergast, A. M., Pondel, M., Landau, R. N., Littman, D. R., and Witte, O. N. (1991) Mol. Cell. Biol. 11, 1785–1792