Implication of a Small GTPase Rac1 in the Activation of c-Jun N-terminal Kinase and Heat Shock Factor in Response to Heat Shock*

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Song Iy Han, Su Young Oh, Sang Hyekwo Woo, Kyung Hee Kim, Jae-Hong Kim, and Ho Sung Kang§

From the Department of Molecular Biology, College of Natural Sciences, Pusan National University, Pusan 609-735, and the §Department of Life Science, Kwangju Institute of Science and Technology, Kwangju 506-712, Korea

Heat shock induces c-Jun N-terminal kinase (JNK) activation as well as heat shock protein (HSP) expression through activation of the heat shock factor (HSF), but its signal pathway is not clearly understood. Since a small GTPase Rac1 has been suggested to participate in the cellular response to stresses, we examined whether Rac1 is involved in the heat shock response. Here we show that moderate heat shock (39–41 °C) induces membrane translocation of Rac1 and membrane ruffling in a Rac1-dependent manner. In addition, Rac1N17, a dominant negative mutant of Rac1, significantly inhibited JNK activation by heat shock. Since Rac1V12 was able to activate JNK via Rac1, similar inhibition by Rac1N17 of HSF activation in response to heat shock was observed. However, inhibitory effects of Rac1N17 on heat shock-induced JNK and HSF activation were reduced as the heat shock temperature increased. Rac1N17 also inhibited HSF activation by L-azetidine-2-carboxylic acid, a proline analog, and heavy metals (CdCl2), suggesting that Rac1 may be linked to HSF activation by denaturation of polypeptides in response to various proteotoxic stresses. However, Rac1N17 did not prevent phosphorylation of HSF1 in response to these proteotoxic stresses. Interestingly, a constitutively active mutant Rac1V12 did not activate the HSF. Therefore, Rac1 activation may be necessary, but not sufficient, for heat shock-inducible HSF activation and HSP expression, or otherwise a signal pathway(s) involving Rac1 may be indirectly involved in the HSF activation. In sum, we suggest that Rac1 may play a critical role(s) in several aspects of the heat shock response.

Upon exposure to elevated temperatures, cells exhibit a conserved defense mechanism, the heat shock response (also known as the stress response). The cellular response to heat shock involves an elevated expression of highly conserved proteins referred to as the heat shock proteins (HSPs)1. The HSPs are known to function as molecular chaperones during protein folding/assembly and membrane translocation and to prevent aggregation of misfolded polypeptide chains in cells (for reviews, see Refs. 1 and 2). They are also involved in protection against stress-induced apoptosis (3). The HSPs can be induced by other proteotoxic stresses such as L-azetidine-2-carboxylic acid (Azc), a proline analog, and heavy metals (1). Expression of the HSPs is mediated through the activation of the heat shock transcription factor (HSF), which binds to and activates a conserved regulatory site, the heat shock element (HSE), located in the promoters of heat-inducible HSP genes (for review, see Refs. 4 and 5). In vertebrates, HSFs 1–4 have been identified: HSFs 1, 2, and 4 are ubiquitous, whereas HSF3 has been characterized only in avian species (5). Under nonstressful conditions, HSF1 exist as a non-DNA-binding form in complex with other regulatory proteins such as HSP90 an HSP70, which are believed to function as repressors of HSF1 activation in the absence of stress (4–8). It has been suggested that nonnative polypeptides accumulated by heat shock and proteotoxic stresses function as a common proximal inducer of HSF1 activation and HSP expression (1, 9, 10). In stressed cells, misfolded proteins may compete with HSP1 for binding chaperone HSP70, and unbound HSF1 homotrimerizes, becomes transcriptionally competent and is hyperphosphorylated (4, 5, 11). Although protein kinases such as mitogen-activated protein kinase, glycogen synthase kinase 3, and JNK have been demonstrated to down-regulate HSF1 activation in response to heat shock (12, 13), the signal pathway of heat shock activation of HSF1 is as yet unclear. Heat shock also activates JNK, one of mitogen-activated protein kinase family proteins and which is implicated in a variety of cell regulation such as cell growth and apoptosis (14–18). A putative JNK phosphatase has been suggested to be responsible for the heat shock-induced JNK activation (19, 20).

A small GTPase Rac1, a member of Rho GTPase family, has been implicated in the regulation of various fundamental cellular processes, including actin cytoskeletal organization, transcriptional activation, and cell proliferation (for reviews, see Refs. 21–23). Rac1 was originally found to play essential roles in growth factor-induced membrane ruffling and cell proliferation (24–28). It also mediates activation of JNK in response to stresses such as ceramide and Fas (29–31). Therefore, the signal pathway involving Rac1 is likely involved in the stress shock factor; HSF, heat shock element; JNK, c-Jun N-terminal kinase; PAGE, polyacrylamide gel electrophoresis; ROS, reactive oxygen species; EMSA, electromobility shift assay; EMSSA, electromobility super-shift assay; Azc, L-azetidine-2-carboxylic acid; PBS, phosphate-buffered saline; DTT, dithiothreitol; EGF, epidermal growth factor; NBD, 12-(N-methyl-N-(7-nitrobenz-2-oxa-1,3-diazol-4-yl)-TBS, Tris-buffered saline.

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1 The abbreviations used are: HSP, heat shock protein; HSF, heat shock factor; HSE, heat shock element; JNK, c-Jun N-terminal kinase; PAGE, polyacrylamide gel electrophoresis; ROS, reactive oxygen species; EMSA, electromobility shift assay; EMSSA, electromobility super-shift assay; Azc, L-azetidine-2-carboxylic acid; PBS, phosphate-buffered saline; DTT, dithiothreitol; EGF, epidermal growth factor; NBD, 12-(N-methyl-N-(7-nitrobenz-2-oxa-1,3-diazol-4-yl)-TBS, Tris-buffered saline.

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§ To whom correspondence should be addressed: Dept. of Molecular Biology, College of Natural Sciences, Pusan National University, Pusan 609-735, Korea. Tel.: 82-51-510-2275; Fax: 82-51-513-9258; E-mail: hspkang@hyowon.cc.pusan.ac.kr.
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response. Here we show that a distinct signal pathway(s) involving Rac1 may be implicated in HSF and JNK activation in response to heat shock. We also demonstrate that Rac1 may be linked to HSF activation by denaturation of polypeptides by other proteotoxic stresses such as Azc and heavy metals. However, Rac1N17 did not prevent phosphorylation of HSF1 in response to these proteotoxic stresses. Although moderate heat shock induced membrane translocation and membrane ruffling in a Rac1-dependent manner, a constitutively active Rac1V12 did not activate the HSF, while it stimulated the JNK activity.

These results suggest that a signal pathway(s) involving Rac1 may be directly and indirectly linked to heat shock-induced JNK and HSF activation, respectively.

EXPERIMENTAL PROCEDURES

Cell Culture, Transfection, Western Blotting, and Immunoprecipitation—Rat-2 fibroblast cells were obtained from the American Type Culture Collection (ATCC, CRL 1764) and grown in Dulbecco's modified Eagle's medium supplemented with 10% (v/v) fetal bovine serum (Life Technologies, Inc.) and 1% penicillin-streptomycin (Life Technologies, Inc.) in a 37 °C humidified incubator with 5% CO₂. Rat-2 stable clones expressing a dominant negative Rac1N17 were prepared as described previously (32). pEXV-Myc and pEXV-Myc-Rac1V12 plasmids (gifts from Dr. Alan Hall) were expressed as N-terminally 9E10 epitope-tagged derivatives under SV40 promoter (25). Rat-2 cells were stably cotransfected with pEXV-Rac1N17 or control vector plasmid, pEXV, along with the Neor gene, and clones were selected and expanded in the presence of G418 (500 µg/ml). For preparation of constitutively active Rac1V12 cells, full-length wild type Rac1 cDNA was cloned into a pcDNA3.1 (Invitrogen), and Gly-to-Val mutagenesis at codon 12 of the Rac1 cDNA was achieved using a mutagenesis kit (Stratagene). pcDNA3.1-Rac1V12 plasmid was expressed under CMV promoter. Rat-2 cells were stably transfected with pcDNA3.1-Rac1V12 or control vector plasmid, pcDNA3.1, and clones were selected and expanded in the presence of G418. Expression of mutant Rac1 proteins was determined by Western blotting with anti-Rac1 antibody (Transduction Laboratories). Rat-2 cells expressing a dominant negative Rac1N17 or a constitutively active Rac1V12 may be directly and indirectly linked to heat shock-induced JNK and HSF activation, respectively.

Electromobility Shift Assay (EMSA) and Supershift Assay (EMSSA)—Cells (1 × 10⁶ cells) were washed three times with cold phosphate-buffered saline (PBS) and the pellet rapidly frozen at −80 °C. After thawing the pellet on ice, it was suspended in buffer A (10 mM Tris-HCl, pH 7.9, 10 mM KCl, 1.5 mM MgCl₂, 0.5 mM DTT) and heated at 100 °C for 5 min. After centrifugation, the supernatant was diluted to modified buffer D (20 mM Tris-HCl, pH 7.9, 10 mM KCl, 1.5 mM MgCl₂, 0.5 mM DTT) and heated at 80 °C. Between steps the samples were kept on ice. Double-stranded oligonucleotides containing the HSE consensus sequence (5’-GATCCTCAAGGTGTTCGAAG-3’) were labeled using Klenow polymerase (Promega) and [α-32P]dCTP (Amersham Pharmacia Biotech, 3,000 Ci/mmol, 10 mCi/ml). 20 µg of nuclear protein was preincubated for 15 min at 0 °C in 17 µl of buffer (20 mM Tris-HCl, pH 7.5, 0.2 mM EDTA, 1.5 mM MgCl₂, 0.5 mM DTT) at 80 °C. Before heating, the samples were kept on ice. Double-stranded oligonucleotides containing the HSE consensus sequence (5’-GATCCTCAAGGTGTTCGAAG-3’) were labeled using Klenow polymerase (Promega) and [α-32P]dCTP (Amersham Pharmacia Biotech, 3,000 Ci/mmol, 10 mCi/ml). 20 µg of nuclear protein was preincubated for 15 min at 0 °C in 17 µl of buffer (20 mM Tris-HCl, pH 7.5, 0.2 mM EDTA, 1.5 mM MgCl₂, 0.5 mM DTT, 100 ng/ml aprotinin, and 10 µg/ml leupeptin) and the cell extracts were incubated with anti-JNK antibody (Santa Cruz Biotechnology, Inc.) for 1 h and then protein A–agarose for an additional 1 h. After centrifugation, the precipitates were incubated in the mixture of 20 mM Heps, pH 7.4, 2 mM sodium orthovanadate, 2 mM DTT, 100 mM MgCl₂[32P]ATP, and 4 µl of glutathione S-transferase-c-Jun at 30 °C for 30 min. To stop the reaction, 4 × Laemmli sample buffer was added to the precipitates and heated at 100 °C for 5 min. After centrifugation, the supernatant was analyzed by SDS-PAGE and autoradiography.

Confocal Microscopy—Rat-2 cells were grown on round coverslips in multiwell culture plates and exposed to epidermal growth factor (EGF, 100 ng/ml) and heat shock for 10 min. The cells were fixed with 3.7% formaldehyde in PBS for 30 min on ice. The fixed cells were then permeabilized by incubating in 0.2% Triton X-100 in PBS for 15 min on ice. For actin staining, the cells were incubated with 0.165 mM NBD-phallacidin for 30 min at room temperature. The stained cells were washed three times in PBS for 15 min and mounted on slide glasses with gelovit, which was prepared by mixing 100 ml of 23% polyvinyl alcohol in PBS with 50 ml glycerol. For Rac1 staining, the permeabilized cells were blocked with 3% bovine serum albumin containing several drops of horse serum in TBS for 1 h at room temperature and then incubated with anti-Rac1 antibody (Upstate Biotechnology) overnight at 4 °C. After washing three times with TBS plus 0.1% Triton X-100 (TBS-T), the cells were stained with fluorescein isothiocyanate-conjugated secondary antibody for 1 h and washed five times in TBS-T and mounted on slide glasses with crystal mount. Then the stained cells were observed under a confocal microscope (LSM510, Carl Zeiss).

RESULTS

Preparation and Characterization of Rac1N17 and Rac1V12 Cells—To test the possible involvement of Rac1 in the heat shock response, we prepared and characterized Rat-2 fibroblast cells expressing either a dominant negative Rac1N17 or a constitutively active Rac1V12 (32). Anti-Rac1 antibody was used in Western blotting of cell extracts to detect simultaneously the amount of mutant and endogenous Rac1. Because of the tag, Rac1 mutants had a slower electrophoretic mobility than endogenous Rac1. The expression levels of mutant Rac1 proteins were similar to that of endogenous Rac1 (Fig. 1A). Expression of Rac1V12 in which amino acid 12 was substituted to Val significantly stimulated the JNK activity, while expression of Rac1N17 in which residue 17 was changed to Asn reduced the
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JNK activity (Fig. 1A). Similar results have been reported by several investigators in different cell lines (27, 28). Rac1 has been shown to play a role in mitogen signaling; when injected into quiescent cells, Rac1V12 stimulates cell cycle progression through G1 and DNA synthesis, while Rac1N17 blocks serum-induced DNA synthesis (21, 26–28). In agreement with these results, Rac1V12 cells exhibited an increased cell growth, whereas Rac1N17 cells showed a decreased cell proliferation (Fig. 1B). In addition, upon stimulation of cells with EGF, Rac1N17 cells did not show any changes in actin filaments, whereas normal cells exhibited extensive membrane ruffling within 10 min, followed by stress fiber formation (32).

Activation of Rac1 and Induction of Membrane Ruffling by Heat Shock—To test whether Rac1 is activated by heat shock, we examined its membrane translocation by confocal microscopy using anti-Rac1 antibody. As shown in Fig. 2A, exposure of Rat-2 cells to moderate heat shock (40 °C) induced membrane translocation of Rac1, similar to that in cells treated with EGF. Similar results were obtained in cells exposed to either 39 or 41 °C (data not shown). Since Rac1 mediates growth factor-induced membrane ruffling and lamellipodia (22, 23, 25), we examined alterations in the actin structure in response to heat shock (39–44 °C) by confocal microscopy using NBD-phallacidin. As exposed to heat shock for 10 min, membrane ruffling was observed (Fig. 2B). Heat shock-induced ruffling pattern was different depending on the heat shock temperature. Mild heat shock (39–41 °C)-induced ruffling was similar to that observed in growth factor-treated cells (22, 23, 25), whereas severe heat shock (43–44 °C)-induced ruffling was similar to that in cells treated with phorbol 12-myristate 13-acetate, a protein kinase C activator (34). Rac1N17 inhibited membrane ruffling in response to mild, but not to severe, heat shock (Fig. 2B). In addition, protein kinase C inhibitors, H7 and staurosporine, did not exert inhibitory effects on severe heat shock-induced ruffling, indicating that protein kinase C is not likely involved in the severe heat shock-induced actin reorganization (data not shown). These results suggest that mild heat shock-induced actin ruffling may occur via a Rac1 signal pathway, while severe heat shock-induced actin remodeling is likely regulated by a Rac1- and protein kinase C-independent mechanism.

Suppression of Heat Shock-induced HSP Expression and HSF1 Activation by Rac1N17—We also investigated whether Rac1 is involved in heat shock-induced HSF activation. Heat shock highly enhanced the synthesis of HSP70 and HSP90 as described previously (1), and Rac1N17 significantly prevented the heat shock-induced HSP expression, while unaffecting synthesis of other proteins (Fig. 4). As shown in Fig. 5A and demonstrated by others (4–6), HSF1 was involved in the heat shock-induced HSP expression. Rac1N17 significantly inhibited heat shock-induced HSF1 activation (Fig. 5B), without affecting the level of HSF1 (Fig. 5C). However, as the heat shock temperature was elevated or heat shock was imposed for prolonged period, an inhibitory effect of Rac1N17 was diminished (Figs. 4 and 5B). These results suggest that Rac1 may be required in heat shock-induced HSP expression and HSF1 activation, although the Rac1-independent mechanism also participates in severe heat shock response.

Inhibition by Rac1N17 of HSP Expression and HSF1 Activation in Response to l-Azetidine-2-Carboxylic Acid and Heavy Metals—It has been suggested that mild heat shock induces denaturation of only nascent polypeptides, while severe heat shock causes misfolding of both cellular proteins and nascent polypeptides (35, 36). Therefore, it is possible that Rac1 may be linked to HSF1
activation by denaturation of nascent polypeptides. First, we tried to confirm whether HSF1 activation in response to mild heat shock response is linked with the regulation of protein synthesis. As demonstrated by others (35, 36), preincubation with cycloheximide, an inhibitor of protein synthesis, significantly blocked HSF1 activation in response to moderate heat shock of 39–41 °C, but not to severe heat shock of 43–45 °C (Fig. 6A). We also examined the effects of cycloheximide on HSF1 activation in response to Azc, which causes denaturation of only nascent polypeptides. As shown in Fig. 6A, Azc-induced HSF1 activation was significantly inhibited by cycloheximide. With other’s previous results (35, 36), these results support that mild heat shock only affects the proteins being newly synthesized, whereas severe heat shock causes misfolding of nascent polypeptides as well as proteins preexisting in cells.

It was then examined whether Rac1 is involved in HSF activation by denaturation of nascent polypeptides. Azc-induced HSF1 activation was prevented by Rac1N17 (Fig. 6B). However, Rac1N17 also prevented HSF (possibly HSF1) activation by CdCl2 (Fig. 6B), which is thought to cause denaturation of both nascent polypeptides and proteins preexisting in cells. Furthermore, Rac1N17 also prevented HSF1 activation in response to heat shock of 43 °C, at which cycloheximide did not exert an inhibitory effect on HSF1 activation (Fig. 5B, lower panel). Taking these results together, we suggest that Rac1 may be linked to activation of HSF1 by denaturation of cellular polypeptides in response to a variety of proteotoxic stresses, including heat shock, amino acid analogs, and heavy metals.

Heat Shock-induced HSF1 Phosphorylation Is Not Prevented by Rac1N17—HSF1 has been known to be hyperphosphorylated upon exposure to proteotoxic stresses such as heat shock and heavy metals (4, 5, 11). Therefore, the effects of Rac1N17 on heat shock-induced HSF phosphorylation were also examined. As shown in Fig. 7, HSF1 was hyperphosphorylated in cells exposed to severe heat shock (43–45 °C) and cadmium chloride, while its phosphorylation slightly increased in cells treated with mild heat shock (39–41 °C) and Azc. Rac1N17 did not prevent HSF1 phosphorylation in response to these proteotoxic stresses, indicating that HSF1 phosphorylation is not likely linked to the Rac1 signal pathway.

Rac1V12 Does Not Activate HSP Expression and HSF Activation—Finally, we examined whether heat shock-induced Rac1 activation is sufficient for HSP synthesis and HSF activation. In constitutively active Rac1V12 cells, however, no significant induction of HSP synthesis and HSF activation was observed (Fig. 8). These results suggest that heat shock-induced Rac1 activation may be necessary, but not sufficient, for heat shock-induced HSP expression and HSF activation, or otherwise a signal pathway(s) involving Rac1 may be indirectly linked to the HSF activation.
A small GTPase Rac1, one of the Rho family GTPases, has been implicated in a variety of cell regulation, including actin remodeling, gene transcription, and cell proliferation (21–28). In fact, a constitutively active mutant Rac1V12 stimulated cell proliferation, whereas a dominant negative mutant Rac1N17 repressed cell growth (Fig. 1B). Rac1 has also been shown to participate in the cellular response to stresses (16, 17, 30, 31). For example, upon exposure to ceramide and Fas, cells induce apoptosis through activating the Rac1-JNK signal pathway (29). In this study, we examined whether Rac1 is implicated in the heat shock response. We show that mild heat shock (39–41 °C) induced membrane translocation and possibly activation of Rac1 (Fig. 2A). In addition, mild heat shock (39–42 °C) also induced membrane ruffling in a Rac1-dependent manner (Fig. 2B), similar to that observed in growth factor-treated cells (22, 23, 25). These results suggest that mild heat shock can activate Rac1. It has been demonstrated that PI 3-kinase and PAK, signal molecules that are involved in the Rac1 signal pathway, are activated by heat shock (37, 38), further supporting heat shock activation of Rac1. However, it is not clear how heat shock activates Rac1. Heat shock is known to increase membrane fluidity and to alter the composition of cell membrane (39–41), which may in turn lead to the activation of various membrane proteins, including receptor tyrosine kinases. In fact, heat shock was shown to activate EGF receptor tyrosine kinase in NIH-3T3 cells (37). These results allow us to speculate that multiple receptor tyrosine kinases, including EGF receptor, can be activated by heat shock, as shown by other stresses such as UV irradiation and osmotic shock (42), and their activation may result in the activation of Rac1.

Rac1N17 significantly prevented JNK activation in response to heat shock, although the inhibitory effect of Rac1N17 was reduced as the heat shock temperature increased (Fig. 3A). Since heat shock activates Rac1 (Fig. 2A), and a dominant positive mutant Rac1V12 induces the activation of JNK (Fig. 1A), mild heat shock-induced JNK activation is likely regulated via Rac1 (Fig. 9). A number of signal pathways have been reported to mediate JNK activation in response to external stimuli. It has been well demonstrated by several investigators that JNK activation by growth factors, inflammatory cytokines, and stressful agents such as ceramide is mediated through Rac1 (21, 26–28). In contrast, protein-damaging stresses such as sodium arsenite, oxidative stress, and ethanol have been shown to induce JNK activation primarily through inhibiting a putative JNK phosphatase (19, 20, 43). For example, the reactive oxygen species (ROS) is able to directly inhibit JNK phosphatase by oxidizing the SH group of the protein (19, 20). Similar to other proteotoxic stresses, heat shock has been suggested to activate JNK through inhibiting a JNK phosphatase (19, 20). Since heat shock can produce ROS (44), and the heat shock response such as HSP expression is prevented by pretreatment of anti-oxidants (45), ROS may serve as a mediator of the heat shock response. Furthermore, unlike other Rho family GTPases, Rac1 is able to produce ROS through activating NADPH oxidase (21, 46). Therefore, we examined in this study whether ROS is involved in heat shock-induced JNK activation. However, pretreatment of antioxidants such as ascorbic acid, butylated hydroxytoluene, and n-propylgallate did not prevent JNK activation by heat shock (data not shown). Thus, although Rac1 could produce ROS through activating NADPH oxidase (46), heat shock-induced JNK activation is likely regulated by the Rac1 signal pathway not involving ROS and NADPH oxidase. PAK, one of Rac1 downstream effectors and which is upstream of JNK, has been reported to be activated by heat shock (38). Therefore, the Rac1-PAK signal pathway may be responsible for heat shock-induced JNK activation. Alternatively, MKK7, which can be activated by stresses and is upstream of JNK (47), may be responsible for the heat shock-induced JNK activation (Fig. 9). We also show that HSF activation in response to heat shock is significantly prevented by Rac1N17, although the inhibitory effect of Rac1N17 on HSF activation is diminished in severe heat shock-treated cells (Figs. 4 and 5). To shed light on the specific role of Rac1 in the heat shock response, Rac1N17 also prevented HSF activation by l-azetidine-2-carboxylic acid, a proline analog, and heavy metals (CdCl2) (Fig. 6), indicating that Rac1 may be implicated in HSF activation by denaturation of polypeptides in response to proteotoxic stresses. In agreement with these findings, it has been demonstrated that mechanical stress-induced HSF activation and HSP expression in vascular smooth muscle cells are
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...heat shock response such as fever. Rac1 has been implicated in a variety of cellular functions, including growth factor-induced actin reorganization and cell proliferation, apoptosis, gene expression, and survival (21–23). Thus, fever may function as an important signal, which is required for resetting our body conditions, rather than acting as a proteotoxic stress. Further studies on functions of Rac1 in the mild heat shock response may provide a new insight into the elucidation of roles of heat shock in organism.

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REFERENCES

1. Welch, W. J., Kang, H. S., Beckmann, B. P., and Mizzen, L. A. (1991) Curr. Top. Microbiol. Immunol. 167, 31–55
2. Hoffmann, J. M., and Newbold, C. R. (1991) Curr. Opin. Microbiol. 3, 210–214
3. Masser, D. D., Carone, A. W., Bourget, L., Denis-Larose, C., and Massie, B. (1997) Mol. Cell. Biol. 17, 5317–5327
4. Wu, C. (1995) Annu. Rev. Biochem. 64, 411–469
5. Morimoto, R. I. (1998) Genes Dev. 12, 3788–3796
6. Abbravaya, K., Myers, M. P., Murphy, S. P., and Morimoto, R. I. (1992) Genes Dev. 3, 1153–1164
7. Zou, J., Gun, Y., Guettouche, T., Smith, D. F., and Voellmy, R. (1998) Cell 94, 471–480
8. Ali, A., Bhardwaj, S. O., Carroll, K., and Orsene, N. (1998) Mol. Cell. Biol. 18, 4949–4960
9. Ananthan, J., Goldberg, A. L., and Voellmy, R. (1998) Science 282, 522–524
10. Dubois, M. F., Hoanessian, A. G., and Beraudeau, O. (1991) J. Biol. Chem. 266, 7973–7971
11. Baler, R., Dahl, G., and Voellmy, R. (1993) Mol. Cell. Biol. 13, 4246–4249
12. Chu, B., Soncin, F., Price, B. D., Stevenson, M. A., and Calderwood, S. K. (1996) J. Biol. Chem. 271, 3037–3057
13. Dai, R., Frejtag, W., He, B., Zhang, Y., and Mivechi, N. F. (2000) J. Biol. Chem. 275, 18210–18218
14. Kyriakis, J. M., Banerjee, P., Nikolakaki, E., Dai, T., Rubie, E. A., Ahmad, O. F., Avruch, J., and Woodgett, J. R. (1994) Nature 369, 156–160
15. Kyriakis, J. M., and Avruch, J. (1996) Bioessays 18, 567–577
16. Xia, Z., Dickens, M., Raingeaud, J., Davis, R. J., and Greenberg, M. E. (1995) Science 267, 1286–1289
17. Verheij, M., Bose, R., Lin, X. H. Y., Yao, B., Jarvis, W. D., Grant, S., Birrer, M. J., Szabo, E., Zou, L. I., Kyriakis, J. M., Haimovitz-Friedman, A., Fuks, Z., and Kolesnick, R. N. (1996) Nature 380, 75–79
18. Adler, V., Schaffer, A., Kim, J., Dolan, L., and Ronai, Z. (1995) J. Biol. Chem. 270, 26071–26077
19. Merin, A. B., Nagel, J. A., Gabai, V. L., Zon, L., Gainiatsas, S., Mosser, D. D., Zou, L., and Shen, Y. X. (1999) Mol. Cell. Biol. 19, 2547–2555
20. Nguyen, A. N., and Shinzaki, K. (1999) Genes Dev. 13, 1653–1663
21. Van Aelst, L., and D'Souza-Schorey, C. (1997) Nature 391, 2295–2322
22. Tapon, N., and Hall, A. (1997) Curr. Opin. Cell Biol. 9, 86–92
23. Hall, A. (1998) Science 279, 529–541
24. Ridley, A. J., and Hall, A. (1992) Cell 70, 389–399
25. Ridley, A. J., Paterson, H. F., Johnston, C. L., Diekmann, D., and Hall, A. (1992) Cell 70, 401–410
26. Olsen, M. F., Ashworth, A., and Hall, A. (1995) Science 269, 1270–1272
27. Lamarche, N., Tapon, N., Stowers, L., Burbeio, P. D., Asprenxom, P., Bridges, T., Chant, J., and Hall, A. (1996) Cell 87, 519–529
28. Joneson, P., McDougel, B., Bar-Sagi, D., and Van Aelst, L. (1996) Science 274, 1374–1376
29. Brenner, B., Koppennhofer, U., Weinstock, C., Linderkamp, O., Lang, F., and Gruenbaum, E. (1997) J. Biol. Chem. 272, 22173–22181
30. Coso, O. A., Chiariello, M., Yu, Y., Teramoto, H., Crespo, P., Xu, N., Miki, T., and Gutkind, J. S. (1995) Cell 81, 1137–1146
31. Chen, A., Lin, A., Llacer, F. X., Abo, V., and Karin, M. (1995) Cell 81, 1147–1157
32. Shin, E. A., Kim, K. H., Han, S. I., Ha, K. S., Kim, J. H., Kang, K. I., Kim, H. D., and Kang, H. S. (1999) FEBS Lett. 452, 355–359
33. Kim, K. H., Kang, H. S., and Kim, H. D. (1999) J. UMBR Life 48, 429–433
34. Myat, M. M., Anderson, S., Allen, L. A., and Aderem, A. (1997) J. Biol. Chem. 272, 611–614
35. Baker, R., Welch, W. J., and Voellmy, R. (1992) J. Cell Biol. 117, 1151–1159
36. Tanabe, M., Nakai, A., Kawazoe, Y., and Nagata, K. (1997) J. Biol. Chem. 272, 15389–15395
37. Lin, R. Z., Hu, Z. W., Chiu, J. H., and Hoffman, B. B. (1997) J. Biol. Chem. 272, 12986–13002
38. Maroni, P., Bendinioli, P., Zucconorosono, C., Schiaffonati, L., and Picoletti, R. (1999) Cell Biol. Int. 24, 145–152
39. Vigh, L., Marcesa, B., and Harwood, J. L. (1998) Trends Biochem. Sci. 23, 369–374
40. Anderson, R. L., and Parker, R. (1982) Int. J. Radiat. Biol. Relat. Stud. Phys. Chem. Med. 40, 57–69
41. Murakami-Murofushi, K., Nishikawa, K., Hirakawa, H., and Murofushi, H. (1997) J. Biol. Chem. 272, 486–489
42. Rosette, C., and Karin, M. (1996) Science 274, 1194–1197
43. Kim, K. H., and Aderem, A. (1999) Cell Signal. 11, 1–14
44. Davison, J. F., Whyte, B., Bissinger, P. H., and Schiestl, R. H. (1996) Proc. Natl. Acad. Sci. U. S. A. 93, 5116–5121
45. Gorman, A. M., Heavey, B., Creagh, E., Cotter, T. G., and Samali, A. (1999)
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46. Mizuno, T., Kaibuchi, K., Ando, S., Musha, T., Hiraoka, K., Takaishi, K., Asada, M., Nunoi, H., Matsuda, I., and Takai, Y. (1992) J. Biol. Chem. 267, 10215–10218
47. Moriguchi, T., Toyoshima, F., Masuyama, N., Hanafusa, H., Gotoh, Y., and Nishida, E. (1997) EMBO J. 16, 7045–7053
48. Xu, Q., Schett, G., Li, C., Hu, Y., and Wick, G., (2000) Circ. Res. 86, 1122–1128
49. Ozaki, M., Deshpande, S. S., Angelow, P., Suzuki, S., and Irani, K., (2000) J. Biol. Chem. 275, 35377–35383
50. Dewey, W. C. (1989) Radiat. Res. 120, 191–204
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