FEBS 20622 FEBS Letters 433 (1998) 9–14

Minireview

# Photoaffinity labeling as an approach to study supramolecular nucleoprotein complexes

D.G. Knorre*, T.S. Godovikova

*Institute of Bioorganic Chemistry, Siberian Division of Russian Academy of Sciences, pr. Academika Lavrentyeva 8, Novosibirsk 630090, Russia*

Received 21 May 1998; revised version received 3 July 1998

**Abstract The modern approaches for studying the detailed structure of nucleoprotein complexes involved in replication and transcription, based on the use of nucleic acids with photoreactive groups incorporated into definite positions of polynucleotide chain, are considered. Methods of preparation of photoreactive nucleic acids of this type are presented. Their use for positioning of RNA polymerase III and transcription factors as well as of the main participants of the replication machinery at the respective templates is described. A survey of the data concerning the amino acid residues modified in the course of photoaffinity labeling of proteins is also presented and some complications are discussed.**


*Key words:* Photoaffinity modification; Nucleoprotein; Photoreactive nucleic acid; Arylazide; Photomodified amino acid residue

Photoaffinity labeling has become one of the most efficient approaches for studying the structure of active sites of biopolymers and supramolecular structures of nucleoprotein complexes. It makes it possible to perform modification in the millisecond range, thus opening up new opportunities for the study of dynamic events in such systems. The first experiment of this sort was carried out to study the T7 RNA polymerase binding to the DNA template. The mixture of the components was directly irradiated with UV-light after definite short intervals following their mixing. It was demonstrated that the initial regions of the photoattachment of the enzyme are located rather far from the promoter. The results of more postponed irradiation indicate that RNA polymerase moves along the DNA template towards the promoter region, probably by linear diffusion [1]. Already this rather simple example demonstrates the great significance of photocrosslinking studies of the dynamics of protein-nucleic acid interactions in supramolecular systems participating in the most essential genetic processes such as replication and transcription. Therefore, it seems quite reasonable that photoaffinity labeling was most intensively elaborated for the investigation of nucleoprotein structures and dynamics of the processes of multiplication and expression of genetic information.

Certainly the final goal of such experiments is elucidation of the residues which are in mutual contact at distinct stages of the process. Direct photocrosslinking by UV-irradiation at 260 nm seems to be rather unfavorable in this case since a number of bases may participate in the process. This requires further elucidation of the position of bases involved in the photocrosslinking. A much more precise approach has become available due to elaboration of methods permitting the introduction of photoreactive groups into definite positions of the nucleic acid chain. The first goal of this review is to describe these methods and to present the most impressive examples of the application of such derivatives to the study of the nucleoprotein systems.

Several types of photoreactive groups have been proposed for incorporation into definite positions of nucleic acids including 4-thiouridine, 5-bromouridine and various arylazido derivatives [2]. These nucleic acid analogs make it possible to align definite components of the replication and transcription apparatus: polymerases themselves and various auxiliary proteins (e.g. transcription factors) at respective sites of nucleic acid. We shall restrict our consideration to arylazido derivatives since these have been used most intensively and have led to definite results for such complicated systems as DNA templates interacting with eukaryotic transcription factors and its interaction with a number of accessory proteins participating in DNA replication. One of the essential advantages of arylazido derivatives of nucleic acids and oligodeoxyribonucleotides is the possibility of varying rather easily the types of reactive groups as well as the length and rigidity of spacers connecting these groups with the respective heterocycles.

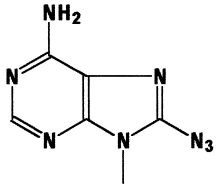
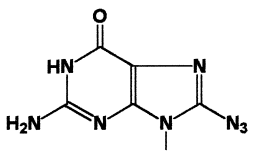
As far as protein components are concerned our knowledge of the sites and chemistry of the protein photoaffinity labeling within nucleoproteins remains rather poor [2]. Nevertheless, significant amounts of data have accumulated showing which amino acids are modified in the course of photoaffinity labeling of individual proteins, especially with arylazides. Therefore, our second goal is to present a survey of these data and to consider the main tendencies in photomodification of amino acid residues with this group of reagents under specific conditions when photoreactive groups are fixed in the vicinity of these residues. This consideration seems to be important for further movement towards a better understanding of the mechanisms of protein-nucleic acid and protein-protein interactions in supramolecular structures.

One way of preparation of nucleic acids modified with photoreactive group at definite positions is based on pulse elongation with photoreactive deoxynucleoside-5′-triphosphates (dNTP) of a synthetic primer annealed with a predetermined region of the template [3]. Usually these reagents contain photoreactive groups tethered either to the C5 of uracil ring or to the 4-amino group of cytosine. Another approach is based on the chemical synthesis of primers with introduction of the photoreactive monomer at a definite stage of the synthesis [4]. For investigation of the replication system it is desirable to prepare photoreactive primers with various defi-

*Corresponding author. Fax: (7) (3832) 333677.
E-mail: knorre@modul.bioch.nsk.su


PII: S0014-5793(98)00860-6

Table 1
Amino acid residues of proteins subjected to photoaffinity labeling with the presented arylazido reagents

| Photoreactive groups | Photoaffinits reagents | Amino acid residue modified | Protein modified | Ref. |
|---|---|---|---|---|
|  | 8-azido-ADP | Tyr-1041<br>Tyr-398 | Hamster P-glycoprotein | [14] |
| | 8-azido-ATP | Lys-199 | Interferon inducible 2',5'-oligoadenylate synthetase | [15] |
| | (2'(3')-0-(2,4,6-trinitrophenyl)-8-azido-AMP/ATP | Lys-186<br>Lys-72<br>Lys-87 | Cytochrome C | [16] |
| | 8-azido-2'-deoxyadenosine containing DNA | Lys-244 | Rel protein (transcription factor) | [17] |
| | 8-azido-AMP | Tyr-1006 | Transhydrogenase from beef heart | [18] |
| | 8-azido-cAMP | Tyr-371<br>R(E200/D)<br>Tyr-371<br>R(F247/Y)<br>Trp-371<br>R(Y371/W) | cAMP-dependent protein kinase I | [19] |
| | 8-azido-AMP | Tyr-244 | cAMP-dependent protein kinase | [20] |
| | 8-azido-cAMP | Tyr-371<br>Trp-260 | cAMP-dependent protein kinase | [21] |
| | 8-azido-ATP | Tyr-766 | E.coli DNA polymerase I Klenow fragment | [22] |
| | 8-azido-A-DNA | Tyr-766 | E.coli DNA polymerase I | [23] |
| | 8-azido-adenosine 3',5'-bisphosphate | Thr-82 | Bovine pancreatic ribonuclease A | [24] |
| | 8-azido-adenosine | Ala-177<br>Ile-321 | Human placental S-adenosylhomocysteine hydrolase | [25] |
| | 8-azido-2'-O-dansyl-ATP | Leu-115<br>Cys-25<br>His-36 | Adenylate kinase | [26] |
|  | 8-azido-GTP | Lys-119 | Human recombinant cap binding protein eIF-4E | [27] |
| | 8-azido-GTP | Lys | Glutamate dehydrogenase isoenzymes | [28] |
| | 8-azido-GTP | Lys-445 | Glutamate dehydrogenase (regulatory site) | [29] |

nite distances of photoreactive residues from their 3′-termini. This may be done by further restricted elongation with natural dNTPs [4]. To study interactions of RNA genes with RNA polymerase and transcription factors elongation of the primer containing photoreactive residues at definite positions should be performed to convert the probe to double stranded DNA with functional regions essential for the transcription machinery.

The first method was used to prepare probes containing 5-[*N*-(*p*-azidobenzoyl)-3-aminoallyl]dU residues for the study of the contact points of the suppressor $tRNA^{Tyr}$ gene with transcription factor TFIIIC of the yeast RNA polymerase III [5].

Table 1 (*Continued*).

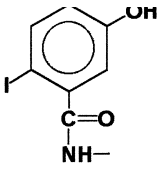
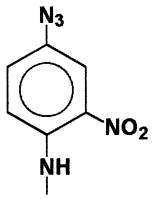
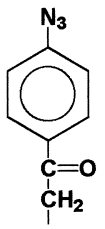
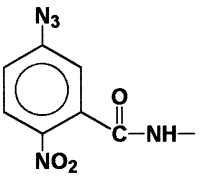
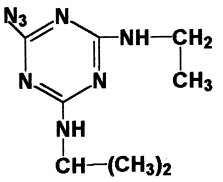
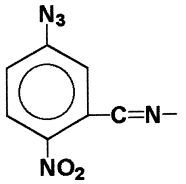
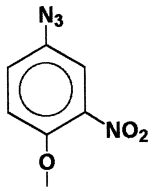
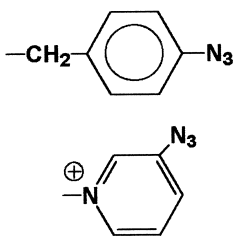
| | | | | |
|---|---|---|---|---|
|  | biotin-YIPSAEKI linked via K to the iodo-4-azido-salicylic acid | Trp | T cell receptor | [44] |
| | [2'3'-spin labeled]-2-azido-ATP | Tyr-345<br>Tyr-368 | Mitochondrial F1-ATPase | [33] |
| | 2-azido-adenosine-ATP | Pro-13 | Cpn (yeast chaperonin) | [34] |
|  | 2[(4-azido-2-nitrophenyl)-amino] ethyl diphosphate | Trp-130 | Skeletal myosin | [35] |
| | 2[(4-azido-2-nitrophenyl)-amino] ethyl diphosphate | Arg-128<br>Cys-198 | Scallop myosin | [36] |
| | 2[(4-azido-2-nitrophenyl)-amino] ethyl diphosphate | Agr-128 | Scallop myosin | [37] |
| | Gly-Pro-Arg-N-(4-azido-2-nitrophenyl)-Lys amide | Tyr-363 | Primary fibrin polymerization site | [38] |
|  | 3-(p-azidobenzyl)-4-hydroxycoumarin | Thr-127<br>Tyr-128 | NAD(P)H: quinone reductase | [39] |
|  | 2-[(4-azidophenacyl)-thio]-2'-dATP | Asp-732 | E.coli DNA polymerase I Klenow fragment | [40] |
| | 8-(p-azidophenacylthio)-cGMP | Val-524<br>Val-525<br>Ala-526 | Retinal rod cGMP-activated channels | [41] |
| | p-azidophenacyl linked to the thiophosphate of Tp(s)CpGpA (p(s) – phosphorthioate) | Tyr-161 | Taq I restriction endonuclease | [42] |
|  | 6-α(β)-(5-azido-2-nitrobenzoyl)amido[17-α)-$^{3}$H]estradiol | Tyr-49 | Mouse monoclonal antiestradiol antibody | [43] |
|  | | | | |

Table 1 (*Continued.*

| | | | | |
|---|---|---|---|---|
|  | 2-azido-4-ethylamino-6-isopropylamino-S-triazin | Met-214 | Photosystem-2 reaction centers from thylakoid membranes | [45] |
|  | 5-[(3-azido-6-nitrobenzylidene)amino]-2,6-diamino-4-pyrimidinone | Lys-198<br>Lys-194 | Rat liver phenylalanine hydroxylase | [46] |
|  | 4-azido-2-nitrophenyl phosphate | Asn-398<br>Pro-668 | $Na^+/K^+$ - ATPase | [47] |
|  | p'-$N^{6-}$-(4-azido-phenylethyl) adenosine-$p^2$-4-(3-azido-pyrimidinio)butyl diphosphate | Thr-86 | Lactate dehydrogenase from pig heart | [48] |

Further, it was successfully used to study the contact points of the 5S RNA gene with transcription factors [3]. A large body of probes were prepared with positions of photoreactive residues from −49 to +160 either in the transcribed or in the non-transcribed strand (positions are numbered relative to the transcription start residues). Detailed information concerning contact points was obtained in this way using photocrosslinking of respective DNA to a mixture of the three main transcription factors TFIIIA, TFIIIB and TFIIIC. For example, several residues of the DNA probe in the range +20–+32 were crosslinked to the 95 kDa subunit of TFIIIC, whereas the points in the ranges −10–+3 and +12–+21 were crosslinked to the 135 kDa subunit of the same factor. In this work crosslinked positions of TFIIIA, the 90 and 70 kDa subunits of TFIIIB and the 145, 135, 95 and 90 kDa subunits of TFIIIC were localized [3]. Somewhat earlier a similar approach resulted in the elucidation of the contact points of the SUP $tRNA^{Tyr}$ gene with TFIIIB and TFIIIC [6]. Similarly the alignment of the RNA polymerase III subunits at the same template was investigated [7]. To obtain more exhaustive information the partly fractionated S-100 extract was recently [8] used to study photocrosslinking with immobilized photoreactive DNA containing the SUP $tRNA^{Tyr}$ gene. A new photoreactive derivative 4-[*N*-(*p*-azidobenzoyl)-2-aminoethyl]-dCTP was used for pulse primer extension. The results were consistent with those obtained with purified factors and enzyme in [6,7]. However, a novel 40 kDa protein was found to be photocrosslinked additionally to the gene.

The second method was applied to establish the positioning of the *Escherichia coli* DNA polymerase III subunits at the template-primer complex. Oligodeoxyribonucleotides supplied with a $(CH_2)_3$-$NH_2$ spacer at C5 of the deoxyuridine at one of the positions −3, −9, −13, −18, −22, −27 or −46 (relative to the 3′ end) were prepared [4]. They were converted to photoreactive derivatives by treatment with *N*-hydroxysuccinimide ester of 5-azido-2-nitrobenzoic acid (NAB-dU fragments). Irradiation of the template-primer complexes with the enzyme resulted in preferential photocrosslinking of the primer residues −3, −9 and −13 to the α-subunit (this subunit contains the catalytic site for DNA polymerization) of the core enzyme

complex α-ε-θ; residues −13, −18 and −22 to the γ-subunit of the γ-complex (γ-δ-δ′-χ-ω); residue −22 to the β-subunit. The approach makes it possible to follow the events preceding elongation. It was demonstrated that the γ-subunit covalently binds to the −3 residue of the primer in the preinitiation complex (γ-complex+β) and its displacement to the −13 to −22 region takes place after addition of the core enzyme [4].

Interesting information based on photocrosslinking data was obtained for the T4 phage replication system [9]. The minimal essential set of proteins involved consists of gp43 (polymerase itself), gp32 (single strand binding protein) and accessory proteins gp44, gp62 and gp45 which form a clamp providing the processivity of replication. To study this system two primers with a NAB-dU residue at position −4 or −20 were synthesized and annealed with the complementary region of single stranded ΦX174 DNA served as a template. Due to the specific structure of the template following the part annealed with primer d(...GAAGACGCAGT...) both primers could be enzymatically extended by 5 or 10 nucleotide residues using appropriate combination of dNTPs. Thus primers with a photoreactive group at positions −4, −9, −14, −20, −25 or −30 were prepared and subjected to photocrosslinking to the components of the replication system. In the presence of the non-hydrolyzable ATP analog with one S atom at γ-phosphate (ATPγS) the polymerase photocrosslinking was very weak and a rather high level of crosslinking of gp44 and gp62 to residues −4 and −14 was recorded. The −14 and −20 residues were found to photocrosslink most intensively to gp45.

In this system it was demonstrated that photocrosslinking makes it possible to follow the nucleoprotein complex rearrangements accompanying the complex assembly. When ATP was substituted for ATPγS, a severe change of the crosslinking pattern was observed. DNA polymerase was crosslinked to the −4 residue whereas no crosslinking of gp44 occurred and only weak crosslinking of gp62 remained. This suggests that in the absence of ATP these proteins occupy the region of location of polymerase and are displaced by the latter due to ATP hydrolysis. The crosslinking of gp45 to residue −14 completely disappears in this system and that to residue −20 is much weaker than in the ATP containing system. Therefore, it may be concluded that gp45 moves downstream in the presence of ATP.

So far, only first attempts that make use of proteins as the photoreactive group carrier in supramolecular structures have been described. As an example the elucidation of the binding of transcription factors with the C-terminal domain (CTD) of the large subunit of RNA polymerase II may be presented [10]. This domain consists of a number of repeating oligopeptide sequences Tyr-Ser-Pro-Thr-Ser-Pro-Ser which may be phosphorylated at their OH groups. CTD is involved in a number of functions of RNA polymerase II. Recombinant CTD was enzymatically thiophosphorylated with [$^{35}$S]ATPγS at a number of positions. Alkylation with $BrCH_2C(O)C_6H_4N_3$ converted CTD-thiophosphate to photoreactive protein. Irradiation of this CTD derivative with a HeLa transcription extract resulted in photocrosslinking to several extract components. Crosslinked proteins containing a $^{35}$S label were separated from CTD by treatment with phenylmercury acetate. According to the molecular masses in SDS-gel electrophoresis, proteins of 34 kDa and 74 kDa were labeled predominantly and comigrated with the β-subunit of TFIIE and one of the TFIIF subunits [10].

Recently photoreactive oligoribodeoxynucleotide derivatives have been tried as a tool for investigation of chromatin which is a significantly more complicated system than that described above [11,12]. The $(pdT)_{16}$ derivatives carrying photoreactive X-C(O)-NH-$(CH_2)_2$-NH residues at the 5′-end, where X = 2-$NO_2$, 5-$N_3$-$C_6H_3$-, 4-$N_3$-$C_6H_4$-, or 4-$N_3$-$C_6F_4$-, were found to modify specifically a definite set of proteins. This set did not differ from those obtained with alkylating derivatives, but the modification extent of proteins was higher [11]. The most efficient modification of the chromatin proteins in the vicinity of $(pdA)_n$ repeats was achieved with 4-$N_3C_6H_4NH(CH_2)_4$NH-$(pdT)_{16}$ [12]. An essential advantage of this type of photoreactive group is its specificity towards proteins without any noticeable reactivity to nucleic acids [13].

A survey of recent data [14–48] concerning modification points of the photoaffinity labeled proteins by arylazide derivatives is presented in Table 1. Unfortunately, so far, very poor data are available about the photoaffinity modification points of the nucleic acid polymerizing enzymes. These data are insufficient to draw definite conclusions concerning the main tendencies of the photoaffinity labeling of various amino acid residues in proteins. Therefore, it seemed reasonable to present the whole set of existing data dealing with the elucidation of photomodified amino acid residues in the course of affinity labeling of different proteins with arylazido derivatives. This permitted us to attempt to draw some general conclusions concerning the probability of interaction of definite amino acid residues with various types of irradiated arylazides.

The data presented in Table 1 were mainly obtained by Edman degradation of the photomodified peptides after their chromatographic isolation from the respective hydrolysates. It seems that the new approach based on electrospray and MALDI mass spectrometry will substitute for the more laborious stepwise Edman procedure in the near future. For example, electrospray mass spectrometry was successfully used to identify modified peptides obtained from T7 RNA polymerase affinity labeled with a psoralen derivative of oligonucleotide [49].

As is seen in Table 1, photoadducts are predominantly formed with nucleophilic groups, which is typical of singlet nitrenes (mainly with lysine, tyrosine and tryptophan residues). Direct one-step insertion of singlet nitrenes into the aliphatic C-H bond is found in a small number of cases. Reactions of aliphatic residues via triplet nitrenes should proceed via a set of free radical reactions. However, it requires the formation of rather stable intermediate radicals, which seems rather improbable for aliphatic amino acids found to be modified (Ala, Ile, Leu, Val, Pro).

An interesting observation was made by Wower et al. [24] for the 8-azidoadenosine derivative used for the photoaffinity labeling of bovine pancreatic RNase. It was found that preirradiated 8-$N_3$-pAp retained the ability to modify the enzyme in the dark at the same point as under irradiation of the mixture of the enzyme with the photoreagent. Unfortunately, similar control experiments are absent for all other cases of photoaffinity labeling with 8-azidoadenosine derivatives. As a result, it remains unclear whether this is an exception or a general property of these types of photoreactive groups. Earlier a similar phenomenon was found for some *p*-azidoaniline

derivatives. The nitrene formed under irradiation isomerizes rapidly to a *p*-quinonediimine residue capable of electrophilic attack in the dark on the nucleophilic targets [50].

It should be emphasized that treatment of biopolymers with photoreactive compounds under irradiation does not necessarily result in photocrosslinking. Thus irradiation of phosphoenolpyruvate carboxy kinase with the substrate analog 8-azido-GTP inactivates the enzyme without incorporation of the photoanalog moiety into the enzyme. It was found that under irradiation the photoanalog induces formation of a disulfide bridge between two adjacent cysteines [51]. A lack of correlation between the extents of modification and inactivation was observed also in the study of photoaffinity modification of DNA polymerase I (*E. coli*) with *p*-$N_3C_6H_4NH$-[α-$^{32}$P]dTTP. Inactivation of the enzyme was not accompanied by the incorporation of the $^{32}$P label in the enzyme [52]. The authors explain this result by the cleavage of the bond linking the protein modified by the *p*-quinonediimine residue and γ-phosphate of dTTP. These data clearly demonstrate that the absence of photocrosslinking does not necessarily mean the absence of any photoreaction with the target, and consequently, of a direct contact of a photoreactive group of some reagent with an appropriate part of a partner. In particular, the experimental results dealing with the positioning of proteins at specific DNA described in [3–8] should be considered with some caution. Some proteins that are in contact with the DNA reactive residues could escape crosslinking due to a reaction that does not result in the introduction of a label into a protein. Both results as well as the dark reactions of the preirradiated aromatic azides described above demonstrate clearly the importance of a deeper knowledge of chemical mechanisms of photoaffinity labeling.

## References


[1] Park, C.S., Wu, F.Y.-H. and Wu, C.-W. (1982) J. Biol. Chem. 257, 6590–6596.
[2] Meisenheimer, K.M. and Koch, T.H. (1997) Crit. Rev. Biochem. Mol. Biol. 32, 101–140.
[3] Braun, B.R., Bartholomew, B., Kassavetis, G.A. and Geiduschek, E.P. (1992) J. Mol. Biol. 228, 1063–1077.
[4] Reems, J.A., Wood, S. and McHenry, S. (1995) J. Biol. Chem. 270, 5606–5613.
[5] Bartholomew, B., Kassavetis, G.A., Brown, B.R. and Geiduschek, E.P. (1990) EMBO J. 9, 2197–2205.
[6] Bartholomew, B., Kassavetis, G.A. and Geiduschek, E.P. (1991) Mol. Cell Biol. 11, 5181–5189.
[7] Bartholomew, B., Durkovich, D., Kassavetis, G.A. and Geiduschek, E.P. (1993) Mol. Cell Biol. 13, 942–952.
[8] Lannutti, B.J., Persinger, J. and Bartholomew, B. (1996) Biochemistry 35, 9821–9831.
[9] Capson, T.L., Benkovic, S.J. and Nossal, N.G. (1991) Cell 65, 249–258.
[10] Kang, M.E. and Dahmus, M.E. (1995) J. Biol. Chem. 270, 23390–23397.
[11] Chernolovskaya, E.L., Cherepanov, P.P., Gorozhankin, A.V., Dobrikov, M.I., Vlassov, V.V. and Kobets, N.D. (1993) Bioorg. Khim. 19, 889–893.
[12] Kobets, N.D., Gorozhankin, A.V., Godovikova, T.S., Silnikov, V.N. and Knorre, D.G. (1996) Dokl. Akad. Nauk 349, 822–825.
[13] Godovikova, T.S., Berezovski, M.V. and Knorre, D.G. (1995) Bioorg. Khim. 21, 857–867.
[14] Sankaran, B., Bhagat, S. and Senior, A.E. (1997) FEBS Lett. 417, 119–122.
[15] Kon, N. and Suhadolnik, R.J. (1996) J. Biol. Chem. 271, 19983–19990.
[16] McIntosh, D.B., Parrish, J.C. and Wallace, C.J.A. (1996) J. Biol. Chem. 271, 18379–18386.
[17] Liu, J., Fan, Q.R., Sodeoka, M., Lane, W.S. and Verdine, G.L. (1994) Chem. Biol. 1, 47–55.
[18] Hu, P.S., Persson, B., Hoog, J.O., Jornvall, H., Hartog, A.F., Berden, J.A., Holmberg, E. and Rydstrom, J. (1992) Biochim. Biophys. Acta 1102, 19–29.
[19] Ringheim, G.E. and Taylor, S.S. (1990) J. Biol. Chem. 265, 19472–19478.
[20] Ringheim, G.E., Saraswat, L.D., Bubis, J. and Taylor, S.S. (1988) J. Biol. Chem. 263, 18247–18252.
[21] Bubis, J., Saraswat, L.D. and Taylor, S.S. (1988) Biochemistry 27, 1570–1576.
[22] Rush, J. and Konigsberg, W.H. (1990) J. Biol. Chem. 265, 4821–4827.
[23] Catalano, C.E., Allen, D.J. and Benkovic, S.J. (1990) Biochemistry 29, 3612–3621.
[24] Wower, J., Aymie, M., Hixson, S.S. and Zimmermann, R.A. (1989) Biochemistry 28, 1563–1567.
[25] Yuan, C.S. and Borchardt, R.T. (1995) J. Biol. Chem. 270, 16140–16146.
[26] Chuan, H., Lin, J. and Wang, J.H. (1989) J. Biol. Chem. 264, 7981–7988.
[27] Friedland, D.E., Shoemaker, M.T., Xie, Y., Wang, Y., Hagedorn, C.H. and Goss, D.J. (1997) Protein Sci. 6, 125–131.
[28] Cho, S.W., Ahn, J.Y., Lee, J. and Choi, S.Y. (1996) Biochemistry 35, 13907–13913.
[29] Shoemaker, M.T. and Haley, B.E. (1993) Biochemistry 32, 1883–1890.
[30] Kim, H., Jacobson, M.K., Rolli, V., Menissier-de Murcia, J., Reinbolt, J., Simonin, F., Ruf, A., Schulz, G. and de Murcia, G. (1997) Biochem. J. 322, (Pt. 2) 469–475.
[31] Grammer, J.C., Kuwayama, H. and Yount, R.G. (1993) Biochemistry 32, 5725–5732.
[32] Mayinger, P. and Klingenberg, M. (1992) Biochemistry 31, 10536–10543.
[33] Vogel, P.D., Nett, J.H., Sauer, H.E., Schmadel, K., Cross, R.L. and Trommer, W.E. (1992) J. Biol. Chem. 267, 11982–11986.
[34] Bramhall, E.A., Cross, R.L., Rospert, S., Steede, N.K. and Landry, S.J. (1997) Eur. J. Biochem. 244, 627–634.
[35] Okamoto, Y. and Yount, R.G. (1985) Proc. Natl. Acad. Sci. USA 82, 1575–1579.
[36] Kerwin, B.A. and Yount, R.G. (1993) Proc. Natl. Acad. Sci. USA 90, 35–39.
[37] Kerwin, B.A. and Yount, R.G. (1992) Bioconjugate Chem. 3, 328–336.
[38] Yamazumi, K. and Doolittle, R.F. (1992) Proc. Natl. Acad. Sci. USA 89, 2893–2896.
[39] Myszka, D.G. and Swenson, R.P. (1991) J. Biol. Chem. 266, 4789–47897.
[40] Moore II, B.M., Jalluri, R.K. and Doughty, M.B. (1996) Biochemistry 35, 11642–11651.
[41] Brown, R.L., Gramling, R., Bert, R.J. and Karpen, J.W. (1995) Biochemistry 34, 8365–8370.
[42] Mayer, A.N. and Barany, F. (1995) Gene 153, 1–8.
[43] Rousselot, P., Mappus, E., Blachere, T., de Ravel, M.R., Grenot, C., Tonnelle, C. and Cuilleron, C.Y. (1997) Biochemistry 36, 7860–7868.
[44] Romero, P., Casanova, J.L., Cerottini, J.C., Maryanski, J.L. and Luescher, I.F. (1993) J. Exp. Med. 177, 1247–1256.
[45] Whitelegge, J.P., Jewess, P., Pickering, M.G., Gerrish, C., Camilleri, P. and Bowyer, J.R. (1992) Eur. J. Biochem. 207, 1077–1084.
[46] Gibbs, B.S. and Benkovic, S.J. (1991) Biochemistry 30, 6795–6802.
[47] Tran, C.M. and Farley, R.A. (1996) Biochemistry 35, 47–55.
[48] Becker, S., Bergman, T., Hjelmqvist, L., Jeck, R., Jornvall, H., Leibrock, H. and Woenckhaus, C. (1996) Biochim. Biophys. Acta 1293, 277–283.
[49] Sastry, S.S. (1996) Biochemistry 35, 13519–13530.
[50] Badashkeyeva, A.G., Gall, T.S., Efimova, E.V., Knorre, D.G., Lebedev, A.V. and Mysina, S.D. (1983) FEBS Lett. 155, 263–266.
[51] Lewis, C.T., Haley, B.E. and Carlson, G.M. (1989) Biochemistry 28, 9248–9255.
[52] Kudryashova, N.V., Shamanina, M.Yu., Godovikova, T.S., Ananko, E.A., Akhmadieva, F.F. and Romashchenko, N.A. (1993) Biokhimiya 58, 224–230.