Amino Acids (2002) 23: 95–101
DOI 10.1007/s00726-001-0114-6

Amino
Acids
Printed in Austria

Submolecular adventures of brain tyrosine: what are we searching for now?

A. Kochman[1], **Cz. Kośka**[2], and **D. Metodiewa**[2]

[1] Department of Pathological Anatomy, Medical University of Wrocław, Wrocław, Poland
[2] Institute of Applied Radiation Chemistry, Technical University of Łódź, Łódź, Poland

Received June 28, 2001
Accepted August 6, 2001
Published online June 17, 2002

Summary. This overview summarizes recent findings on the role of tyrosyl radical (TyrO$^\bullet$) in the multitudinous neurochemical systems of brain, and theorizes on the putative role of TyrO$^\bullet$ in neurological disorders [Parkinson's disease (PD), Alzheimer's disease (AD) and amyotrophic lateral sclerosis (ALS)]. TyrO$^\bullet$ and tyrosine per se can interact with reactive oxygen species (ROS) and reactive nitrogen species (RNS) via radical mechanisms and chain propagating reactions. The concentration of TyrO$^\bullet$, ROS and RNS can increase dramatically under conditions of generalized stress: oxidative, nitrative or reductive as well, and this can induce damage directly (by lipid peroxidation) or indirectly (by proteins oxidation and/or nitration), potentially causing apoptotic neuronal cell death or autoschizis.

Evidence of lesion-induced neuronal oxidative stress includes the presence of protein peroxides (TyrOOH), DT (o,o'-dityrosine) and 3-NT (3-nitrotyrosine). Mechanistic details of protein- and enzymatic oxidation/nitration *in vivo* remain unresolved, although recent *in vitro* data strongly implicate free radical pathways via TyrO$^\bullet$. Nitration/denitration processes can be pathological, but they also may play: 1) a signal transduction role, because nitration of tyrosine residues through TyrO$^\bullet$ formation can modulate, as well the phosphorylation (tyrosine kinases activity) and/or tyrosine hydroxylation (tyrosine hydroxylase inactivation), leading to consequent dopamine synthesis failure and increased degradation of target proteins, respectively; 2) a role of "blocker" for radical-radical reactions (scavenging of NO$^\bullet$, $NO_2^\bullet$ and $CO_3^{\bullet-}$ by TyrO$^\bullet$); 3) a role of limiting factors for peroxynitrite formation, by lowering $O_2^{\bullet-}$ formation, which is strongly linked to the pathogenesis of neural diseases.

It is still not known if tyrosine oxidation/nitration via TyrO$^\bullet$ formation is 1) a footprint of generalized stress and neuronal disorders, or 2) an important part of $O_2^{\bullet-}$ and NO$^\bullet$ metabolism, or 3) merely a part of integral processes for maintaining of neuronal homeostasis. The full answer to these questions should be of top research priority, as the problem of increased free radical formation in brain and/or imbalance of the ratios ROS/RNS/TyrO$^\bullet$ may be all important in defining whether oxidative stress is the critical determinant of tissue and neural cell injury that leads to pathological end-points.

Keywords: Tyrosine – Oxidation – Nitration – Mechanisms – Neural disorders – Generalized stress

Generalized cellular stress (oxidative, nitrative and reductive) is regarded as a key element underlying the onset of neurodegenerative disorders: Parkinson's disease (PD), Alzheimer disease (AD) and amyotrophic lateral sclerosis (ALS) (Clement et al., 1998; Torreilles et al., 1999; de Gray, 2000; Metodiewa and Kośka, 2000). Active neurons produce and release reactive oxygen species (ROS) and reactive nitrogen species (RNS), both of which create oxidative stress. Excess production of ROS and RNS can overwhelm neuronal antioxidant capacity and thus inactivate vital cellular processes and ultimately lead to cell death. Oxidative species that might mediate this damage include $^\bullet$OH (hydroxyl radicals), tyrosyl radicals (TyrO$^\bullet$) and RNS such as peroxynitrite (PN), NO$^\bullet$ (nitric oxide) and $NO_2^\bullet$ (nitric dioxide). The aim of this review is to focus on the role(s) of tyrosine (Tyr) and its radical TyrO$^\bullet$ in the sequence of events arising after neural cell injury has been initiated (generalized stress conditions). We hypothesize on the role of these radical species in the aetiology and progression of neurologic disorders. The role of Tyr (and its radical derivatives) in oxidative/nitrative events (incl. reactions with ROS and RNS) in the initiation of neurodegenerative disorders (e.g., PD, AD, ALS) has not been adequately addressed.

As shown in Scheme 1, one-electron oxidation of Tyr by heme ferryl ions (classical peroxidatic mechanism) (Metodiewa and Dunford, 1993) results in the highly reactive TyrO$^\bullet$ which by itself can cause lipid peroxidation (Savenkova et al., 1994), crosslinking of protein(s) tyrosyl residues *in situ* (Pichorner et al., 1995; Winterbourn et al., 1997; Tien, 1999; van Dalen et al., 2000), or coupling to give DT (o,o'-dityrosine)

and formation of a peroxide adduct TyrOOH (tyrosine peroxide).

The reaction of TyrO$^\bullet$ with superoxide ($O_2^{\bullet-}$) is a scavenging reaction, which proceeds rather by addition than by electron transfer (Pichorner et al., 1995):

$$2\text{Tyr} + H_2H_2 \xrightarrow{\text{Px}} 2\text{TyrO}^\bullet + 2H_2O \quad (1)$$

$$2\text{TyrO}^\bullet \rightarrow \text{Tyr-Tyr (DT)} \quad (2)$$

$$\text{TyrO}^\bullet + O_2^{\bullet-} + H^+ \rightarrow \text{Tyr} + O_2 \quad (3)$$

$$\text{TyrO}^\bullet + O_2^{\bullet-} + H^+ \rightarrow \text{TyrOOH} \quad (4)$$

It is worth nothing that a decrease in the $O_2^{\bullet-}$ concentration (reactions 3 and 4) and H_2O_2 formation by SOD (cellular antioxidant), reduction and acidification of the intracellular milieu can constitute a "signal" for H_2O_2-mediated apoptosis, thereby inducing a reductive as opposed to an oxidative stress (Clement et al., 1998). This mechanism remains to be explored, because of the new-found existence of peroxidases in human *substantia nigra* (Calzigna et al., 2000).

Evidence of generalized stress within neuronal lessions includes the presence of DT and 3-NT (3-nitrotyrosine) which, in turn, indicate that TyrO$^\bullet$, ROS and RNS can attack Tyr residues of neural proteins (Metodiewa, 1998; Pennathur et al., 1999; Metodiewa and Kośka, 2000). Most of the recently proposed mechanisms of Tyr nitration *in vivo* rely on free radical reactions, demonstrated in model solutions:

(i) peroxidative (peroxidase-catalyzed) oxidation of Tyr to TyrO$^\bullet$ (reaction 1) and peroxidative oxidation of nitrite to $NO_2^\bullet$ (van Dalen et al., 2000). Notably, this recently proposed mechanism is consistent with TyrO$^\bullet$ that exchanges with tyrosyl residues and couples with $NO_2^\bullet$ to form 3-NT. The nonenzymatic oxidation of Tyr by $^\bullet$OH can also result in TyrO$^\bullet$ and Tyr(OH)$^\bullet$ (Santos et al., 2000). Thus, nitration of phenolic Tyr requires an obligatory one-electron step (oxidation), and formed TyrO$^\bullet$ is the precursor for both DT (reaction 2) and 3-NT, formed by the following addition of $NO_2^\bullet$ or NO$^\bullet$ (Prütz et al., 1985; Eiserich et al., 1995; Lymar et al., 1996; Daiber et al., 1998);

(ii) oxidation of the formed product (adduct) of recombination reaction as follows (Goldstein et al., 2000):

$$\text{TyrO}^\bullet + \text{NO}^\bullet \Leftrightarrow \text{TyrONO} \rightarrow \text{3-NT} \quad (5)$$

This adduct could be oxidized to 3-NT in the presence of appropriate oxidants (e.g., GSH). Catalysis of $O_2^{\bullet-}$ dismutation by SOD will not

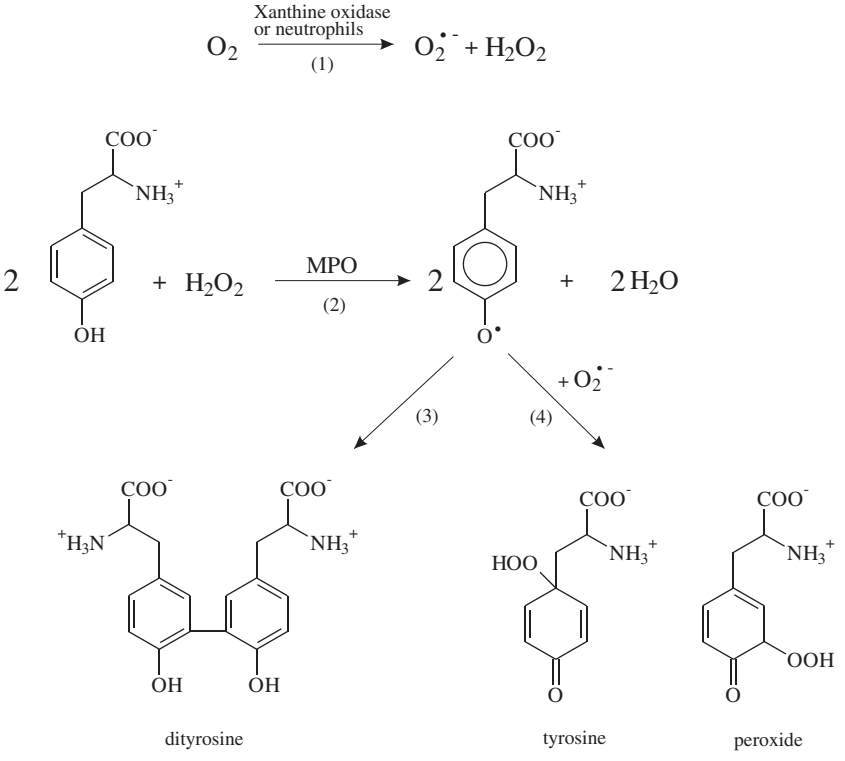

Fig. 1. Reaction scheme for formation of DT (o,o'-dityrosine and TyrOOH (tyrosine peroxide) after peroxidatic oxidation of tyrosine by MPO (myeloperoxidase) (Winterbourn et al., 1997)

prevent nitration (reaction 5). This reversible reaction of $TyrO^{\bullet}$ with $NO^{\bullet}$ is one of its important properties: once formed, 3-NT may act as a promoter of repetitive redox cycling by its reduction to the corresponding nitroanion radical, which can be oxidized by molecular O_2 to yield $O_2^{\bullet -}$, and regenerate to parent 3-NT (Krainev et al., 1998);

(iii) auto-oxidation of $NO^{\bullet}$ to $NO_2^{\bullet}$, carbonate radical oxidation of Tyr to $TyrO^{\bullet}$, or oxidation by $NO_2^{\bullet}$ as follows (Liu et al., 1998; Santos et al., 2000):

$$4NO^{\bullet} + O_2 \rightarrow 2NO_2^{\bullet} \rightarrow N_2O_4 \quad (6)$$

$$NO_2^{\bullet} + Tyr \rightarrow TyrO^{\bullet} \quad (7)$$

$$CO_3^{\bullet -} + Tyr \rightarrow TyrO^{\bullet} + HCO_3^- \quad (8)$$

$$NO_2^{\bullet} + TyrO^{\bullet} \rightarrow [O_2N\text{-}Tyr] \rightarrow \text{3-NT} + \text{products} \quad (9)$$

(iv) peroxynitrite (PN) decomposition in reaction with CO_2 to $CO_3^{\bullet -}$ and $NO_2^{\bullet}$, where $TyrO^{\bullet}$ is the expected precursor of 3-NT, nitrodityrosine (NO_2-DT) and DT as well (Zhang et al., 2000). Bicarbonate stimulates 3-NT formation in solutions containing nitrite anion (NO_2^-), SOD1 and H_2O_2, where the fast electron transfer reaction between $CO_3^{\bullet -}$ and NO_2^- results in $NO_2^{\bullet}$ formation. $NO_2^{\bullet}$ can abstract the phenolic H atom of Tyr (reaction 7) (Santos et al., 2000; Zhan et al., 2000) to $TyrO^{\bullet}$ which recombines with $NO_2^{\bullet}$ to form 3-NT. The formation of 3-NDT (3-nitro dityrosine) was explained as follows (Zhang et al., 2000):

$$CO_3^{\bullet -} + NO_2 - Tyr \rightarrow HCO_3^- + NO_2 - Tyr^{\bullet} \quad (10)$$

$$NO_2 - TyrO^{\bullet} + TyrO^{\bullet} \rightarrow NO_2 - \text{DiTyr (3-NDT)} \quad (11)$$

Notably, 3-NDT (reactions 7, 10, 11) may be used as new markers of both oxidation/nitration reactions of Tyr *in vivo* (Zhang et al., 2000). Bicarbonate-mediated Tyr oxidation/nitration, catalyzed peroxidatively by SOD1 via formation of a putative $CO_3^{\bullet -}$, is shown in Fig. 2.

Currently, the exact and complete mechanism by which Tyr is nitrated, either *in vivo* or in model solutions *in vitro*, remains controversial. Notably, the relationship between 3-NT in proteins and free 3-NT is still unknown, as is the biological/pathological relevance.

Tyr modification to 3-NT via $TyrO^{\bullet}$ by PN reportedly occurs in the presence and/or in the absence of

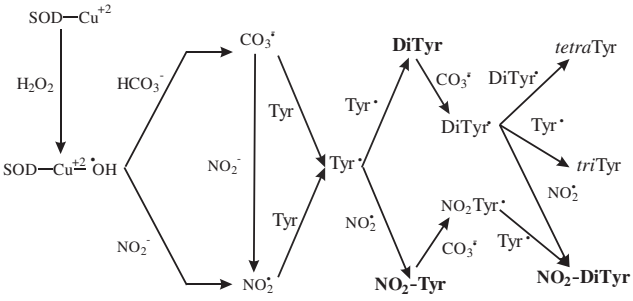

Fig. 2. Reaction scheme of the major reaction pathways leading to formation of DT, 3-NT, NO_2-DiTyr and other higher oxidation products of tyrosine formed during SOD1/H_2O_2/HCO_3^--induced oxidation of tyrosine, in the presence and absence of nitrite (Zhang et al., 2000)

the biologically important bicarbonate-CO_2 pair. At physiological pH, PN can be a significant source of 3-NT (Reiter et al., 2000; Sawa et al., 2000). This recent finding conflicts with the earlier proposal that PN, formed from $NO^{\bullet}$ and $O_2^{\bullet -}$, does not nitrate Tyr at physiological pH (Pfeiffer and Mayer, 1998). However, it is now known that DT formation (reaction 2) outcompetes Tyr nitration at a low steady-state concentration of PN (Pfeiffer and Mayer, 2000). The recent kinetic model from radiation chemical experiments (Goldstein et al., 2000) focuses less on the minor role of PN as a Tyr nitrating agent, and more on DT formation as the major product of $TyrO^{\bullet}$ reaction with $NO^{\bullet}$. This clearly demonstrates that nitration of Tyr via $TyrO^{\bullet}$, in the presence of bicarbonate, depends on the rates of generation of reactants ($NO^{\bullet}$ and $O_2^{\bullet -}$) as shown in the scheme (Sawa et al., 2000):

$$NO^{\bullet} + O_2^{\bullet -} \rightarrow ONOO^- \text{ (PN)} \quad (12)$$

It is noteworthy that a kinetic model of the system $TyrO^{\bullet}/NO^{\bullet}/O_2^{\bullet -}$ was also constructed (Stanbro, 1999), to include the chemical and kinetic "nature" of these reactants. It was proposed that the reversible reaction of $NO^{\bullet}$ and $TyrO^{\bullet}$ (reaction 5) acts to "buffer" the concentration of $TyrO^{\bullet}$ in the system, while the reaction of $TyrO^{\bullet}$ and $O_2^{\bullet -}$ (reaction 3) scavenges it. Quantitatively, the reaction of $NO^{\bullet}$ and $O_2^{\bullet -}$ (reaction 12) is a more important process, but the pathological significance would appear to be related to $TyrO^{\bullet}$ formation and decay as well, which can not be neglected in any considerations of generalized stress. Some alternative pathways for Tyr oxidation/nitration via $TyrO^{\bullet}$ are illustrated in Fig. 3.

The local protein environment and location of Tyr on a loop structure of the molecule, along with its

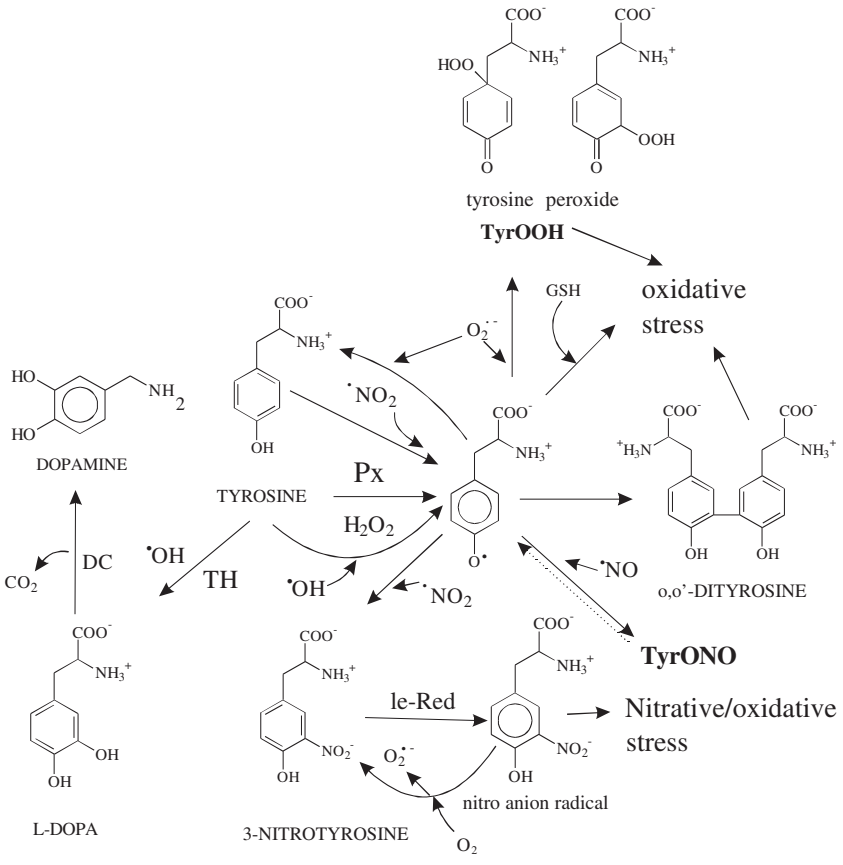

Fig. 3. Diagram of some alternate pathways for tyrosine metabolism. Principal enzymatic pathways include TH-catalyzed DOPA formation, followed by DC-catalyzed dopamine formation

attraction to neighbouring negative charges, are just some of the important factors determining selectivity and efficiency of Tyr oxidation/nitration *in vivo* (Eiserich et al., 1998; Krainev et al., 1998; Souza et al., 1999; Santos et al., 2000).

Nitration of protein-bound and free Tyr, via TyrO•, may modulate cellular (neural) functions through inactivation of enzymes such as tyrosine hydroxylase (TH), the rate-limiting step in catecholamine synthesis. If this leads to consequent dopamine synthesis failure, the severity of parkinsonian symptoms can be exacerbated (Ara et al., 1998). Addition of the neurotoxins 1-methyl-4-phenylpyridinium or 6-hydroxydopamine (MPP^+, 6-OHDA) to dopaminergic neurons (primary culture) leads to nitration of Tyr residues in TH (Pong et al., 2000). TyrO• have been detected during turnover of PGH synthase (prostaglandin endoperoxide H synthase), with the modified residue Tyr 385 being a source of catalytically active TyrO• (Goodwin et al., 1999), which in turn can recombine with NO• in a radical-radical reaction (Guittet et al., 1999; Shi et al., 2000). NO•-trapping of the TyrO• of PGHS can also lead to Tyr imminoxyl radical and 3-NT formation (Gunther et al., 1997). PN-mediated inactivation of Mn-SOD involves nitration and oxidation of its critical Tyr residues (Crow et al., 1997; MacMillan-Crow et al., 1998). The proposed mechanism (Minetti et al., 2000) for nitration of oxyhemoglobin residues involves TyrO• formation via long-rate electron transfer to ferryl hemoglobin, followed by its reaction with $NO_2^{\bullet}$.

Tyrosine nitration via TyrO• is a covalent post-translational protein modification derived from the reaction of proteins with nitrating agents (Souza et al., 1999), which may regulate the posttranslational modification of Tyr residues in pre- and postsynaptic proteins (Di Stasi et al., 1999).

Oxidation of protein-Tyr, via TyrO• and RNS, is dependent on the presence of CO_2 and metal ions, as well as the concentration of reagents – according to kinetic laws (Ducrocq et al., 1999; Stanbro, 1999; Goldstein et al., 2000; van Dalen et al., 2000; Zhan et al., 2000). Basically, nitration of only one or two Tyr residues per protein molecule would still result in loss

of catalytic activity (Ara et al., 1999; Souza et al., 2000). Also, proteasome-degradation of the modified proteins would be accelerated. Elimination of nitrated proteins may represent mechanisms of cellular defense from oxidative/nitrative stress (Souza et al., 2000).

New findings add weight to the concept that TyrO• plays a pivotal role in the pathogenesis of PD. Good et al. (1998) demonstrated the presence of NT-immunoreactivity in Lewy bodies within melanized neurons and in amorphous deposits of degenerating neurons. Tyr residues of neurofilaments, the major structural neuronal proteins, are susceptible to nitration via TyrO• by mutated SOD *in vitro*, and the attendant disruption of their assembly may play a significant role in the development of ALS (Crow et al., 1997). Elevated free 3-NT level, but not protein-bound 3-NT or •OH, implicates TyrO• formation in ALS-like diseases (Brujin et al., 1997). Increased protein nitration in neurons containing neurofibrillary tangles (NFTs) of AD brains is either an indicator of TyrO• formation as an intermediate and involvement of PN in the pathology of AD (Torreilles et al., 1999; Smith et al., 2000). Tyr nitration via TyrO• interferes with phosphorylation: this PN-mediated process can inhibit the ability of protein tyrosine kinases to phosphorylate peptides (Gow et al., 1996), thereby disturbing the kinases(s) mechanisms underlying memory formation (Riedel and Micheau, 1999). Evidence for the existence of oxidative and nitrative stress within inflammatory demyelinating disorders includes the presence of 3-NT, protein peroxides, and lipid peroxides (Smith et al., 1999).

Tyr residues play a crucial role in monoamine oxidase (MAO) substrate and inhibitor specificities (Geha et al., 2000), raising the question of how MAO oxidation/nitration affects dopamine catabolism and melanin formation. Also, cytochrome C nitration may represent both oxidative and signalling events occurring during protein-bound TyrO• formation and RNS-mediated injury of neuronal cells (Cassina et al., 2000). Abundant evidence now supports the concept that TyrO• may promote oxidative reactions at sites of inflammation and in oxidative pathways, in the absence of metal ions.

Notably, the striatum and ventral midbrain of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine- (MPTP)-treated mice exhibit elevated levels of both DT and 3-NT, either free or protein-bound (Pennathur et al., 1999). Nitration of Tyr residues of TH, presumably by TyrO•, parallels the decrease of dopamine level in striatum (Ara et al., 1999). These data lend additional support to the concept that TyrO• is involved in the pathogenesis of PD (Pennathur et al., 1999).

Many studies now indicate that TyrO•-initiated nitration of Tyr produces alterations in either function or activity of neuronal proteins, neurotransmitters, and enzymes (van der Vliet et al., 1995; Gow et al., 1996; Crow et al., 1997; Gunther et al., 1997; Squadrito and Pryor, 1998; Ara et al., 1999; Di Stasi et al., 1999; Souza et al., 2000; Mihm et al., 2001). This can lead to neuropathological states by (i) incorporating a negative charge in hydrophobic Tyr, thereby enhancing proteolysis (Souza et al., 2000); (ii) modifying protein conformation (Prütz et al., 1985; Eiserich et al., 1998; Guittet et al., 1999; Torreilles et al., 1999; Cassina et al., 2000; Mihm et al., 2001); (iii) "blocking" Tyr phosphorylation and altering cell signalling (Gow et al., 1996; Squadrito and Pryor, 1998; Di Stasi et al., 1999).

Currently, it is still unclear as to whether oxidation/nitration of Tyr and associated DT and 3-NT formation, may be a part of those processes required for maintaining brain homeostasis at the cellular level (Nakaki and Fuji, 1999). Thus, increases in DT and 3-NT levels in neuropathological states clearly point toward TyrO• and RNS reactions as being physiologically relevant (Pennathur et al., 1999). If Tyr nitration by TyrO• and RNS reactions is considered to be a neuronal damaging process, it is evident that TyrO•, once formed, has the potential to both initiate and sustain the cascade of reactions ultimately leading to neuronal death.

The above problems relating to TyrO• and its role in intraneuronal protein oxidation and nitration (see Fig. 3) should be a top research priority. Increased free radical formation in brain and/or imbalance in the ratios RNS/ROS/TyrO• may be all important in neuropathological disorders resulting from generalized (oxidative) stress. For now, it is not possible to give an exact or complete description of all possible reactions and interconversions of TyrO•, RNS and ROS within neural cell(s), merely by projecting data derived from model systems *in vitro* and *in vivo*.

In this review we have highlighted the pivotal role for TyrO• in initiating neuronal damage and promoting onset and progression of neurodegenerative disorders. An enormous amount of additional study in needed to determine all possible neurotoxic actions and consequences of TyrO• in human brain.

Acknowledgements

The support of KBN (Poland) Grant 6PO4 A 086 19 is gratefully acknowledged. We thank Prof. R.M. Kostrzewa (East Tennessee State University) for valuable suggestions and helpful discussions.

References

Ara J, Przedborski S, Naini AB, Jackson-Lewis V, Trifiletti RR, Horwitz J, Ischiropoulos H (1998) Inactivation of tyrosine hydroxylase by nitration following exposure to peroxynitrite and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). Proc Natl Acad Sci USA 95: 7659–7663

Bruijn LJ, Beal MF, Becher MW, Schulz JB, Wong PC, Price DL, Cleveland DW (1997) Elevated free nitrotyrosine levels, but not protein-bound nitrotyrosine or hydroxyl radicals, throughout ALS – like disease implicate tyrosine nitration as an aberrant in vivo property of one familiae ALS – linked superoxide dismutase 1 mutant. Proc Natl Acad Sci USA 94: 7606–7611

Calzigna L, De Juliis A, Zanatta L (2000) Enzymatic dopamine peroxidation in substantia nigra of human brain. Clin Chim Acta 300: 131–138

Cassina AM, Hodara R, Souza JM, Thomson L, Castro L, Ischiropoulos H, Freeman BA, Radi R (2000) Cytochrome c nitration by peroxynitrite. J Biol Chem 275: 21409–21415

Clement MV, Ponton A, Pervaiz S (1998) Apoptosis induced by hydrogen peroxide is mediated by decreased superoxide anion concentration and reduction of intracellular milieu. FEBS Lett 440: 13–18

Crow JPP, Ye YZ, Strong M, Kirk M, Barnes S, Beckman JS (1997) Superoxide dismutase catalyzes nitration of tyrosines by peroxynitrite in the rod and head domains of neurofilaments. J Neurochem 69: 1945–1953

Daiber A, Moehl M, Ulrich V (1998) New aspects in the reaction mechanism of phenol with peroxynitrite: the role of phenoxy radicals. Nitric Oxide 2: 259–269

de Grey AD (2000) The reductive hotspot hypothesis: and update. Arch Biochem Biophys 373: 295–301

Di Stasi AM, Mallozzi C, Macchia G, Petrucci TC, Minetti M (1999) Peroxynitrite induces tyrosine nitration and modulates tyrosine phosphorylation of synaptic proteins. J Neurochem 73: 727–735

Ducrocq C, Blanchard B, Pignatelli B, Ohshima H (1999) Peroxynitrite: an endogenous oxidizing and nitrating agent. Cell Mol Life 55: 1068–1077

Eiserich JP, Butler J, van der Vliet A, Cross CE, Halliwell B (1995) Nitric oxide rapidly scavenges tyrosine and tryptophan radicals. Biochem J 310: 745–749

Eiserich JP, Patel RP, O'Donnell VB (1998) Pathophysiology of nitric oxide and related species: free radical reactions and modification of biomolecules. Mol Aspects Med 19: 221–357

Geha RM, Rebrin I, Chen K, Shih JC (2001) Substrate and inhibitor specificities for human monoamine oxidase A and B are influenced by a single amino acid. J Biol Chem 276: 9877–9882

Goldstein S, Czapski G, Lind J, Merényi G (2000) Tyrosine nitration by simultaneous generation of $NO^{\bullet}$ and $O_2^{\bullet-}$ under physiological conditions. How the radicals do the job. J Biol Chem 275: 3031–3036

Good PF, Hsu A, Werner P, Peri DP, Hanow CW (1998) Protein nitration in Parkinson's disease. J Neuropathol Exp Neurol 57: 338–342

Goodwin DC, Gunther MR, Hsi LC, et al (1998) Nitric oxide trapping of tyrosyl radicals generated during PGHS turnover. Detection of the radical derivative of tyrosine 385. J Biol Chem 273: 8903–8909

Gow AJ, Duran D, Malcolm S, Ischiropoulos H (1996) Effect of peroxynitrite – induced protein modifications on tyrosine phosphorylation and degradation. FEBS Lett 385: 63–66

Gunther MR, Hsi LC, Curtis IF, et al (1997) Nitric oxide trapping of the tyrosyl radical by PGHS-22 leads to tyrosine iminoxyl radical and nitrotyrosine formation. J Biol Chem 272: 17086–17090

Guittet O, Roy B, Lepoivre M (1999) Nitric oxide: a radical molecule in quest of free radicals in proteins. Cell Mol Life Sci 55: 1054–1067

Krainev AG, Williams TD, Bigelow D (1998) Enzymatic reduction of 3-nitrotyrosine generates superoxide. Chem Res Toxicol 11: 495–502

Liu XP, Miller MJS, Joshi MS, Thomas DD, Lancaster JR (1998) Accelerated reaction of nitric oxide with $O_2^{\bullet-}$ within the hydrophobic interior of biological membranes. Proc Natl Acad Sci USA 95: 2175–2179

Lymar S, Jiang Q, Hurst JK (1996) Mechanism of carbon dioxide-catalyzed oxidation of Tyr by peroxynitrite. Biochemistry 24: 7855–7861

Mac Millan-Crow LA, Crow IP, Thompson IA (1998) Peroxynitrite mediated inactivation of manganese superoxide dismutase involves nitration and oxidation of critical tyrosine residues. Biochemistry 37: 1613–1622

Metodiewa D (1998) Molecular mechanism of cellular injury produced by neurotoxic amino acids that generate reactive oxygen species. Amino Acids 14: 181–187

Metodiewa D, Dunford HB (1993) Medical aspects and techniques for peroxidases and catalases. In: Scott G (ed) Atmospheric oxidation and antioxidants vol 3. Elsevier, Amsterdam, pp 287–332

Metodiewa D, Kośka Cz (2000) Reactive oxygen species and reactive nitrogen species: relevance to cyto(neuro) toxic events and neurologic disorders. An Overview. Neurotoxicity Research 1: 197–233

Mihm MJ, Schanbacher BL, Wallace BL, Wallace LJ, Uretsky NJ, Bauer IA (2001) Free 3-nitrotyrosine causes striatal neurodegeneration in vivo. J Neurosci 21: RC 149

Minetti M, Pietraforte D, Carbone V, Salzano AM, Scorza G, Marino G (2000) Scavenging of peroxynitrite by oxyhemoglobin and identification of modified globin residues. Biochemistry 39: 6689–6697

Nakaki T, Fujii T (1999) Nitration modifying function of proteins, hormones and neurotransmitters. Jpn J Pharmacol 79: 125–129

Pennathur S, Jackson-Lewis V, Przedborski S, Heinecke JW (1999) Mass spectrometric quantification of 3-nitrotyrosine, ortho-tyrosine, and o,o'-dityrosine in brain tissue of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-treated mice, a model of oxidative stress in Parkinson's disease. J Biol Chem 274: 34621–34628

Pfeiffer S, Mayer B (1998) Lack of tyrosine nitration by peroxynitrite generated at physiological pH. J Biol Chem 273: 27280–27285

Pfeiffer S, Schmidt K, Mayer B (2000) Dityrosine formation outcompetes tyrosine nitration at low steady-state concentrations of peroxynitrite. Implications for tyrosine modification by nitric oxide/superoxide in vivo. J Biol Chem 275: 6346–6352

Pichorner H, Metodiewa D, Winterbourn CC (1995) Generation of superoxide and tyrosine peroxide as a result of tyrosyl radical scavenging by glutathione. Arch Bioch Biophys 323: 429–437

Pong K, Doctorow SR, Baudry M (2000) Prevention of 1-methyl-4-phenylpyridinium and 6-hydroksydopamine –induced nitration of tyrosine hydroxylase and neurotoxicity by EUK-134, a superoxide dismutase and catalase mimetic, in cultured dopaminergic neurons. Brain Res 881: 182–189

Prütz WA, Monig H, Butler J, Land EJ (1985) Reactions of nitrogen dioxide in aqueous model systems: oxidation of tyrosine units in peptides and proteins. Arch Biochem Biophys 243: 125–134

Reiter CD, Teng RJ, Beckman JS (2000) Superoxide reacts with nitric oxide to nitrate tyrosine at physiological pH via peroxynitrite. J Biol Chem 275: 32460–32466

Riedel G, Micheau J (1999) Introduction: molecular mechanisms of memory formation – from receptor activation to synaptic changes. Cell Mol Life Sci 55: 521–524

Santos CXC, Bonini MG, Augusto O (2000) Role of the carbonate radical anion in tyrosine nitration and hydroxylation by peroxynitrite. Arch Biochem Biophys 377: 146–152

Savenkova MJ, Mueller DM, Heinecke JW (1994) Tyrosyl radical generated by myeloperoxidase is a physiological catalyst for the initiator of lipid peroxidation of low density lipoprotein. J Biol Chem 269: 20394–20400

Sawa T, Akaike T, Maeda H (2000) Tyrosine nitration by peroxynitrite formed from nitric oxide and superoxide generated by xanthine oxidase. J Biol Chem 275: 32467–32474

Smith KJ, Kapoor R, Felts PA (1999) Demyelination: the role of reactive oxygen and nitrogen species. Brain Pathol 9: 69–92

Smith MA, Ninomura A, Zhu X, Takeda A, Perry G (2000) Metabolic, metallic and mitotic sources of oxidative stress in Alzheimer disease. Antioxid Redox Signal 2: 413–420

Souza JM, Daikhin E, Yudkoff M, Raman CS, Ischiropoulos H (1999) Factors determining the selectivity of protein tyrosine nitration. Arch Biochem Biophys 371: 169–178

Souza JM, Choi I, Chen Q, et al (2000) Proteolytic degradation of tyrosine nitrated proteins. Arch Biochem Biophys 380: 360–366

Squadrito GL, Pryor WA (1998) Oxidative chemistry of nitric oxide: the roles of superoxide, peroxynitrite and carbon dioxide. Free Rad Biol Med 25: 392–403

Stanbro WD (1999) A kinetic model of the system: tyrosyl radical-nitrogen oxide and superoxide ion. J Theor Biol 197: 557–567

Tien M (1999) Myeloperoxidase-catalyzed oxidation of tyrosine. Arch Biochem Biophys 367: 61–66

Torreilles F, Salman-Tabcheh S, Guerin M-C, Torreilles J (1999) Neurodegenerative disorders: the role of peroxynitrite. Brain Res Rev 30: 153–163

van Dalen CJ, Winterbourn CC, Senthilmohan R, Kettle AJ (2000) Nitrite as a substrate and inhibitor of myeloperoxidase. Implications for nitration and hypochlorous acid production sites of inflammation. J Biol Chem 275: 11638–11644

van der Vliet A, Eiserich JP, O'Neill CA, Halliwell B, Cross CE (1995) Tyrosine modification by reactive nitrogen species: a closer look. Arch Biochem Biophys 319: 341–349

Winterbourn CC, Pichorner H, Kettle AJ (1997) Myeloperoxidase-dependent generation of a tyrosine peroxide by neutrophils. Arch Biochem Biophys 338: 15–21

Zhang H, Joseph J, Felix C, Kalyanaraman B (2000) Bicarbonate enhances the hydroxylation, nitration and peroxidation reactions catalyzed by copper, zinc superoxide dismutase. Intermediacy of carbonate anion radical. J Biol Chem 275: 14038–14045

Authors' address: Dr. Diana Metodiewa, Institute of Applied Radiation Chemistry, Technical University of Łodż, Wróblewskiego 15, 93-590 Łodż, Poland, Fax (48-42) 636-02-46,
E-mail: media@mitr.p.lodz.pl