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Review

Tryptophan metabolism and brain function: focus on kynurenine and other indole metabolites

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Abstract

The synthesis of NAD (or NADP) from tryptophan involves a series of enzymes and the formation of a number of intermediates which are collectively called 'kynurenines.' In the late 1970s and early 1980s, it became clear that intraventricular administration of several 'kynurenines' could cause convulsions and that one of the 'kynurenines,' quinolinic acid, was an agonist of a sub-population of NMDA receptors and caused excitotoxic neuronal death. A related metabolite, kynurenic acid, could, on the other hand, reduce excitotoxin-induced neuronal death by antagonising ionotropic glutamate receptors. Since then, modifications in quinolinic and kynurenic acid synthesis have been proposed as a pathogenetic mechanism in Huntington's chorea and epilepsy. It was subsequently shown that a robust activation of the kynurenine pathway and a large accumulation of quinolinic acid in the central nervous system occurred in several inflammatory neurological disorders. More recently, it has been shown that 3OH-kynurenine or 3OH-anthranilic acid, two other kynurenine metabolites, may cause either apoptotic or necrotic neuronal death in cultures and that inhibitors of kynurenine hydroxylase may reduce neuronal death in in vitro and in vivo models of brain ischaemia or excitotoxicity. Finally, it has been reported that indole metabolites, indirectly linked to the kynurenine pathway, are able to modify neuronal function and animal behaviour by interacting with voltage-dependent Na^+ channels. Oxindole, one of these metabolites, has sedative and anticonvulsant properties and accumulates in the blood and brain when liver function is impaired. In conclusion, a number of metabolites affecting brain function originate from tryptophan metabolism. Selective inhibitors of their forming enzymes may be useful to understand their role in physiology or as therapeutic agents in pathology.

Keywords: Tryptophan; Quinolinic acid; Kynurenic acid; Kynurenine; NMDA receptor; Oxindole; Ischaemia brain; Encephalopathy hepatic

1. Introduction

The kynurenine metabolic pathway of tryptophan metabolism was investigated in depth in the 1950s and 1960s in order to understand its contribution to the synthesis of NAD and NADP, two ubiquitary coenzymes involved in basic cellular processes (Wolf, 1974). In the late 1970s, reports on the convulsant and other behavioural actions of systemic administration of kynurenine derivatives opened the way to studies on their role in brain function (Lapin, 1980). Quinolinic acid is probably the most studied kynurenine metabolite because, by interacting with glutamate receptors of the NMDA type, it may cause convulsion and excitotoxicity (Stone and Perkins, 1981; Foster et al., 1983; Schwarcz et al., 1983). Kynurenic acid is another widely studied neuroactive tryptophan metabolite formed along the kynurenine pathway. It is present in the mammalian brain and is able to antagonise the excitatory amino acid receptors (Perkins and Stone, 1982b; Foster et al., 1983; Moroni et al., 1984a). Other neuroactive kynurenine metabolites are: 3OH-kynurenine, a compound able to cause neuronal damage (necrosis and apoptosis) in cell cultures (Eastman and Guilarte, 1989; Okuda et al., 1998), and picolinic acid, which is able to induce the expression of nitric oxide (NO) synthase in macrophages (Varesio et al., 1990; Melillo et al., 1994). Finally, kynurenine itself seems to be able to increase the expression and synthesis of growth factors (Dong-Ruyl et al., 1998).

Recently, it has been shown that tryptophan is also metabolised in the gut by bacterial tryptophanases and indole is the resulting metabolite (Vederas et al., 1978; Kawata et al., 1991). Indole is then absorbed and further metabolised into oxindole, a strongly sedative compound

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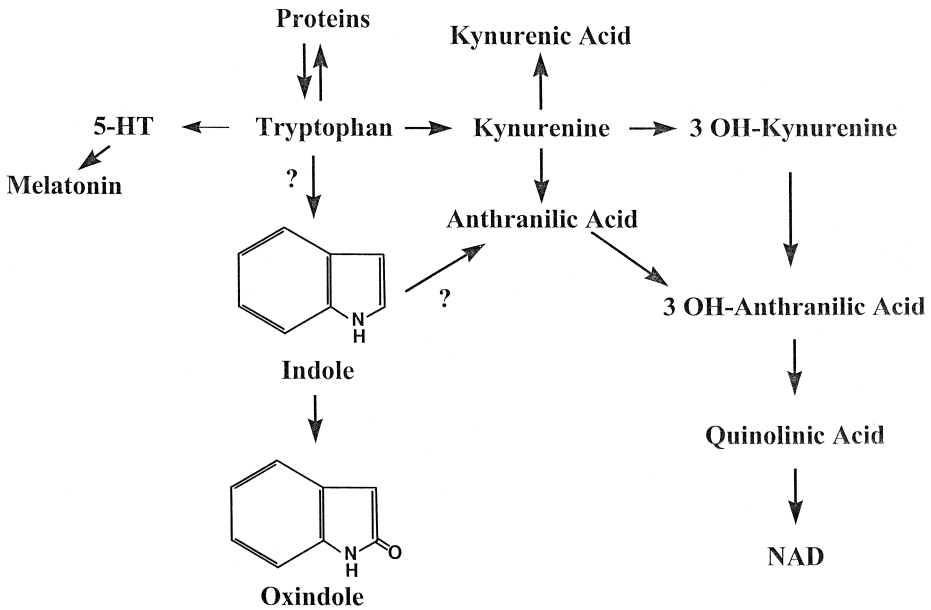

Fig. 1. Possible metabolic pathways of tryptophan metabolism.

which accumulates in the blood and brain when liver function is impaired (see Fig. 1) (Carpenedo et al., 1997; Moroni et al., 1998a).

Excellent reviews covering the neuropharmacology of kynurenine derivatives (Stone and Connick, 1985; Stone, 1993; Reinhard et al., 1994) or the role of kynurenine forming enzymes (Carlin et al., 1989; Taylor and Feng, 1991) are available. Here we will focus mainly on the brain actions of kynurenine metabolites and the regulation of their synthesis and metabolism. We will also discuss the pharmacological tools which are available to study the pathway and which may be proposed for therapeutic strategies in pathological conditions.

2. Formyl-kynurenine and kynurenine

At least two different enzymes, tryptophan-dioxygenase (tryptophan pyrrolase; E.C. 1.13.1.2) and indoleamine 2,3-dioxygenase (E.C. 1.13.11.42) catalyse the irreversible cleavage of the indole ring of L-tryptophan leading to the formation of formyl-kynurenine, which is then rapidly metabolised into kynurenine. The first of these, tryptophan-dioxygenase, was described in 1937 (Kotake and Masayama, 1937) and the regulation of its activity was studied in depth in the 1950s with the aim of understanding the control of tryptophan availability for 5-hydroxytryptamine (5-HT) and NAD synthesis. It is a heme-containing protein mostly expressed in liver-derived cells and it is able to metabolise L- but not D-tryptophan (Schutz et al., 1972; Bender, 1989). A cDNA encoding for tryptophan-dioxygenase has been isolated and characterised and the primary structure of the protein has been deduced (Schmid et al., 1982; Maezono et al., 1990). It has also been shown that the gene contains glucocorticoid-responsive elements which are probably responsible for the control of its synthesis (Danesh et al., 1983, 1987). In fact, glucocorticoid administrations increase tryptophan-dioxygenase formation in the liver (Knox and Mehler, 1951) and cause a decrease of blood tryptophan levels associated with kynurenine accumulation. Since kynurenine and tryptophan compete for the same carrier to be transported from blood to brain, glucocorticoids may cause a depletion in the brain tryptophan pool and a significant decrease in brain 5-HT synthesis (Green et al., 1975; Young, 1981). In view of the importance of 5-HT in brain function, it has been proposed that the induction of tryptophan-dioxygenase is responsible for the mood changes (depression) found in patients chronically treated with glucocorticoids or suffering from Cushing's disease. With the aim of reducing tryptophan catabolism in liver and increasing its availability in brain, a number of tryptophan-dioxygenase inhibitors have been studied in rodents (Salter et al., 1995; Reinhard et al., 1996). Indeed, the administration of (*E*)-6-fluoro-3[-2-(pyridyl)vinyl]-1*H*-indole (680C91) (see Fig. 3) or (*E*)-3-[2-(4′-pyridyl)vinyl]-1*H*-Indole (540C91), two tryptophan-dioxygenase inhibitors, causes a robust increase in the rate of brain 5-HT synthesis (Salter et al., 1995; Reinhard et al., 1996). It could be interesting to evaluate whether or not these agents are effective in the treatment of glucocorticoid-induced mood disorders.

Indoleamine 2,3-dioxygenase is the second enzyme able to open the indole ring of tryptophan. Its existence was suspected for a long time because a large percentage of patients with rheumatoid arthritis, tuberculosis, leukaemia, Hodgkin's disease and bladder tumours excreted exceptionally large amount of kynurenine into the urine without having an induced tryptophan-dioxygenase in the liver.

The enzyme was firstly described by Hayaishi et al. in the early 1960s in the rabbit intestine as a heme-containing protein (42.000 M.W.) able to catalyse the incorporation of superoxide anion into the pyrrole moiety of indole (see Hayaishi et al., 1984 for a review). In order to evaluate its activity in vitro, methylene blue and ascorbate are required to maintain an active ferrous form during the catalysis (Hirata and Hayaishi, 1975). The name indoleamine 2,3-dioxygenase originates from the proposed importance of the enzyme in the metabolism of the indoleamines 5-HT, tryptamine and melatonin (Hirata and Hayaishi, 1975; Yoshida and Hayaishi, 1987). In fact, while tryptophan-dioxygenase specifically acts on L-tryptophan, indoleamine 2,3-dioxygenase may open the indole ring of the amines, D-tryptophan, L- or D-5OH-tryptophan, but it is not active on scatole, indole or indole acetic acid. Furthermore, while tryptophan-dioxygenase is predominantly located in the liver, indoleamine 2,3-dioxygenase is present in most mammalian organs, including intestine, placenta, lung, blood mononuclear phagocytes, epididymis, endocrine and central nervous systems (Watanabe et al., 1981). Indoleamine 2,3-dioxygenase is strongly induced in mouse lung after systemic administration of lipopolysaccharides, pokeweed mitogen, or during influenza virus infections (Yoshida et al., 1979, 1986). Interferons and other pro-inflammatory cytokines also induce indoleamine 2,3-dioxygenase expression when systemically administered to rodents or locally applied in vitro to several cell types, (Carlin et al., 1989; Taylor and Feng, 1991). The cDNA encoding for indoleamine 2,3-dioxygenase has been identified and expressed (Dai and Gupta, 1990a; Tone et al., 1990); two interferon-stimulated response elements (ISRE) and at least one γ-interferon-activated sequence (GAS) are present in the gene to regulate its expression (Dai and Gupta, 1990b; Konan and Taylor, 1996). While interferons and pro-inflammatory cytokines stimulate, other cytokines such as interleukin IV or growth factors such as transforming growth factor β (TGF-β), antagonise indoleamine 2,3-dioxygenase expression and activity, thus suggesting that the enzyme is under tight immunological control (Musso et al., 1994; Yuan et al., 1998).

NO is another important mediator able to inhibit indoleamine 2,3-dioxygenase activity in γ-interferon primed mononuclear phagocytes in vitro (Thomas et al., 1994). The mechanism for this inhibition is not clear and may involve either direct interaction of NO with the heme iron or with superoxide anion. Additional support for the notion that indoleamine 2,3-dioxygenase activity and NO formation may be functionally linked comes from the observation that picolinic acid, one of the products of kynurenine metabolism, induces (Melillo et al., 1994), while 3-hydroxyanthranilic acid, another kynurenine metabolite, inhibits NO synthase expression in murine macrophages (Sekkai et al., 1997). Why do interferons, cytokines, growth factors and NO control indoleamine 2,3-dioxygenase expression and activity? Tryptophan is the least abundant of the essential amino acids and it is required for the maintenance of a number of cellular functions. Changes in indoleamine 2,3-dioxygenase activity may regulate local tryptophan concentrations and protein synthesis. It has been proposed that a reduction in tryptophan availability in the host cells is the mechanism through which γ-interferon inhibits the growth of pathogens such as *Toxoplasma gondi*, *Clamidia psittaci*, *C. trachomatis*, group B *Streptococci* and *Enterococci* (Pfefferkorn, 1984; Shemer et al., 1987; Carlin et al., 1989). Tryptophan deprivation, however, does not explain why γ-interferon inhibits the growth of *Escherichia coli* or *Listeria monocytogens*, suggesting that γ-interferon may also reduce the growth of pathogens through tryptophan-independent mechanisms (MacKenzie et al., 1998). In a similar manner, a tryptophan deprivation mechanism has been proposed to explain the anti-proliferative effects of γ-interferon against several types of neoplastic cells. The experimental basis of this proposal is the demonstration that γ-interferon is not able to reduce the reproductive growth of neoplastic cells unable to express indoleamine 2,3-dioxygenase and that a relationship exists between γ-interferon-mediated indoleamine 2,3-dioxygenase induction and inhibition of neoplastic growth in vitro (Feng and Taylor, 1989; Takikawa et al., 1991). It has, however, been difficult to reproduce this correlation in vivo and it has been suggested that γ-interferon may also reduce neoplastic cell proliferation without inducing indoleamine 2,3-dioxygenase and depleting the cells of tryptophan (Burke et al., 1995).

Another field of research in which the induction of indoleamine 2,3-dioxygenase seems to play a key role is the immunological tolerance between the mother and the foetus. It has been recently shown that administration of an indoleamine 2,3-dioxygenase inhibitor (1-methyl-tryptophan, see Fig. 3; Cady and Sono, 1991) to pregnant mothers causes rejection of allogenic, but not syngeneic concepti (Bonney and Matzinger, 1998; Munn et al., 1998) and proposed that indoleamine 2,3-dioxygenase induction in trophoblast and then in placenta is sufficient to inhibit the maternal immune system and prevent foetal expulsion. It is difficult to understand how a decrease in tryptophan concentration in a single district (the placenta) may affect the whole maternal immune system. We feel that this simple interpretation of the above-mentioned experimental results does not consider that a number of compounds able to affect the immune system of the mother and the foetus, may originate from tryptophan metabolism. In our opinion, the biological meaning of the careful immunological control of indoleamine 2,3-dioxygenase activity is an interesting field of research which is still relatively unexplored.

The opening of the indole ring of tryptophan by either tryptophan-dioxygenase or indoleamine 2,3-dioxygenase results in the formation of formyl-kynurenine which is rapidly metabolised into kynurenine. Unfortunately, relatively little work has been performed on formyl-kynurenine biological activity. Its conversion into kynurenine is rapid

and almost complete because formamidase, the enzyme responsible for formyl-kynurenine metabolism, is quite abundant in most mammalian organs (Mehler and Knox, 1950).

Kynurenine is the key compound of the pathway (see Fig. 2) and is present in blood, brain and peripheral organs in low micromolar concentrations. Peripheral kynurenine is transported through the blood–brain barrier by the large neutral amino acid carrier and may easily reach the central nervous system. Systemic administration of kynurenine can therefore easily reach the brain where it is taken up by glial cells which further metabolise it (Speciale et al., 1989; Fukui et al., 1995). In the late 1970s and early 1980s, a number of studies reported that direct kynurenine injection into the rodent brain caused convulsions (Lapin, 1981, 1982; Pinelli et al., 1984), while systemic injection of very small doses of kynurenine decreased blood pressure in anaesthetised rats (Lapin, 1976). The electrophysiological or neurochemical basis of these effects remains to be discovered, but it has been shown that kynurenine does not directly modify neuronal electrical activity in brain slices or in isolated neurones and has no direct depressant effects on cardiac or smooth muscle tissues (Stone and Connick, 1985).

In the course of screening studies aimed to identify compounds able to increase nerve growth factor (NGF) formation, it has been recently observed that kynurenine application to glial cell cultures increases NGF synthesis and content (Dong-Ruyl et al., 1997, 1998). In view of the importance of NGF on the trophism of different types of neurones, these data could open a new avenue on the interpretation of kynurenine's physiological role and on why its neosynthesis is so carefully controlled.

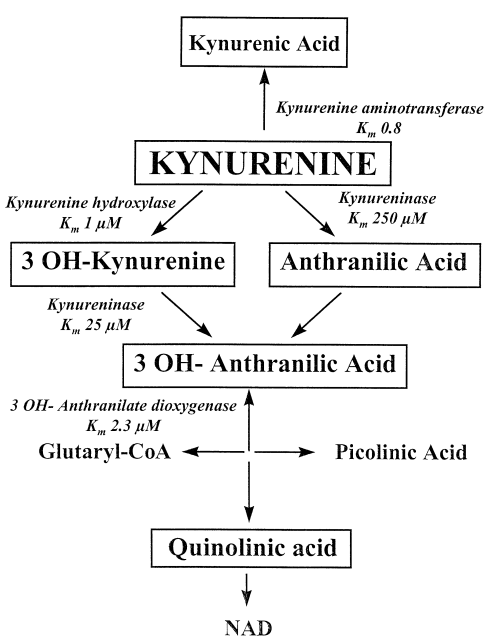

Fig. 2. Kynurenine metabolism: the affinity of the different enzymes for their substrates is also reported.

3. OH-kynurenine and kynurenic acid

At least three different enzymatic activities may metabolise kynurenine in mammalian tissues: (1) kynurenine hydroxylase (E.C. 1.14.13.9.) with the formation of 3OH-kynurenine; (2) kynurenine aminotransferase with the formation of kynurenic acid; (3) kynureninase with the formation of anthranilic acid (see Fig. 2).

Kynurenine hydroxylase is a flavin-containing monooxygenase requiring NADPH as an electron donor (DeCastro et al., 1956) and it is localised in the outer mitochondrial membranes of liver, placenta, spleen, kidney, and brain (Erickson et al., 1992). The enzyme has been recently purified from the pig liver (Uemura and Hirai, 1998) and cloned by hybridisation screening a human liver cDNA library (Alberati-Giani et al., 1997). It has a high affinity for the substrate (K_m in the low micromolar range), thus suggesting that, under physiological conditions, it metabolises most of the available kynurenine (Bender and McCreanor, 1982).

Notably, it has been reported that kynurenine hydroxylase expression increases in inflammatory conditions or after immune stimulation (Saito et al., 1993b). 3OH-kynurenine, the product of its activity, accumulates in the brain of vitamin B-6 deficient neonatal rats (Guilarte and Wagner, 1987) and, at relatively elevated concentrations, it causes cytotoxicity when added to neuronal cells in primary cultures (Eastman and Guilarte, 1989) or when locally injected into the brain (Nakagami et al., 1996). Recently, it has been reported that relatively low concentrations (nanomolar) of 3OH-kynurenine may cause apoptotic cell death of neurones in primary cultures. Interestingly, this neurotoxicity has the features of regional selectivity because the compound is taken up with different affinities by different neuronal populations and it causes accumulation of H_2O_2 inside the cells (Okuda et al., 1998).

Kynurenine amino-transferase is the second enzymatic activity able to metabolise kynurenine and kynurenic acid is the product of this metabolism (see Fig. 2). This activity was first detected in homogenates of mammalian kidney or liver in the presence of pyridoxal phosphate and α-chetoglutarate (Mason, 1954; Okamoto and Hayaishi, 1970). In mammalian peripheral organs, several pyridoxal phosphate-dependent aminotransferases are able to catalyse the transamination of kynurenine (Okuno and Kido, 1991) and two distinct enzymes have been described in rat and human brains (Okuno et al., 1991; Guidetti et al., 1997). The first of them (kynurenine amino-transferase 1) prefers pyruvate as a co-substrate and it is competitively inhibited by glutamine, phenylalanine and tryptophan. Antibodies against this enzyme have shown its preferential location in forebrain astrocytes (Du et al., 1992), but some immunoreactivity also seems to be present in a small percentage of neurones, especially in rat medulla and spinal cord (Kapoor et al., 1997). Interestingly, a cDNA

clone encoding for a soluble protein with kynurenine amino-transferase 1 activity has been isolated from a rat brain library (Alberati-Giani et al., 1995) and surprisingly, its sequence is identical to that of the previously cloned enzyme glutamine transaminase K (E.C. 2.6.1.14) (Perry et al., 1993; Mosca et al., 1994; Alberati-Giani et al., 1995). Another rat brain cDNA clone, encoding for an isoform of the above-mentioned kynurenine amino-transferase/glutamine transaminase K from which it differs because of an additional oligonucleotide stretch coding for 32 amino acids at the NH_2-terminus, has been isolated. It has been proposed that this isoform possesses a peptide necessary for targeting the protein toward the mitochondrial matrix (Malherbe et al., 1995; Cesura et al., 1996). The existence of a mitochondrial protein with kynurenine amino-transferase/glutamine transaminase K activity has also been reported (Stevens et al., 1988) and it has been suggested that the 'mitochondrial' form of kynurenine amino-transferase 1 *iso*-enzyme is predominant in brain, whereas the 'soluble' form is more abundant in kidney (Cooper et al., 1993).

The second enzyme (kynurenine amino-transferase II; E.C. 2.6.1.7) has a preference for oxoglutarate as a co-substrate, displays L-aminoadipate aminotransferase activity and is localised in the soluble cytoplasm (Okuno et al., 1991). Recently, the enzyme was cloned from rat kidney (Buchli et al., 1995) and its presence in the brain has been confirmed with Northern blot hybridisation (Gramsbergen et al., 1997). Lesion and pharmacological studies suggest that a large portion of kynurenic acid present in brain extracellular spaces may indeed originate from kynurenine amino-transferase II activity (Guidetti et al., 1997). Both kynurenine amino-transferase I and II enzymes have K_m in the millimolar range, suggesting that kynurenine availability is the rate limiting step for kynurenic acid biosynthesis. In brain slices and in microdialysis experiments, indeed, the levels of extracellular kynurenate increase linearly with kynurenine availability and it appears that, once formed in glial cells, kynurenic acid is readily liberated into the extracellular space (Swartz et al., 1990; Gramsbergen et al., 1997). The regulation of kynurenate concentration in the brain may have neuropharmacological relevance because at relatively low concentrations (micromolar), kynurenic acid is a glycine$_b$ receptor antagonist (Stone, 1993), while at larger concentrations, it also interacts as an antagonist with glutamate recognition sites of NMDA and α-amino-3-hydroxy-5-methyl-4-isoxazolepropionate (AMPA) receptors. Kynurenic acid is physiologically present in the nervous tissue at nanomolar concentrations (Moroni et al., 1988) and, only after a significant increase, it may be expected to affect excitatory amino acid-mediated neurotransmission. The administration of large doses of kynurenine to neonatal rats has been shown to attenuate brain damage induced by ischaemia or by local injection of NMDA (Nozaki and Beal, 1992). When kynurenine was added to the superfusion fluid of brain slices, neoformed kynurenate was sufficient to abolish convulsions induced by the lack of Mg^{2+} (Scharfman and Ofer, 1997). In order to obtain a larger increase in the brain extracellular concentration of kynurenic acid, a compound which is disposed by the brain through a probenecid-sensitive carrier (Moroni et al., 1988), the administration of kynurenine has been combined with that of probenecid. This association seems to be able to reduce seizures induced by NMDA or pentylenetetrazol (Vécsei et al., 1991) and to attenuate quinolinic acid neurotoxicity in rodent brains (Santamaria et al., 1994; Miranda et al., 1997). On the other hand, experimental manipulations, such as hypoglycaemia or interference with cellular energy metabolism, may reduce kynurenic acid formation in different animal models (Gramsbergen et al., 1997; Hodgkins and Schwarcz, 1998). Reduction in extracellular kynurenic acid concentrations has also been shown in the rat basal ganglia after amphetamine treatment and has been associated to increased vulnerability to excitotoxic insults (Poeggeler et al., 1998). These observations suggest the possibility that, by modulating the brain extracellular kynurenic acid concentrations, it is possible to reduce or increase excitotoxic neuronal damage.

4. Neuropharmacological effects of kynurenine hydroxylase inhibitors: decrease of ischaemic brain damage

The administration of kynurenine hydroxylase inhibitors to rats, mice or gerbils is one of the approaches which may be used to increase kynurenine availability and to cause a significant increase in brain kynurenate content (Moroni et al., 1991; Connick et al., 1992; Russi et al., 1992; Speciale et al., 1996). Systemic administration of kynurenine hydroxylase inhibitors reduces spontaneous locomotor activity and increases the threshold level of electroshock or audiogenic-induced seizures (Carpenedo et al., 1994; Chiarugi et al., 1995). Since these effects are qualitatively similar to what occurs when glycine receptor antagonists are systemically administered (Sing et al., 1990; Vartanian and Taylor, 1991; Dennison et al., 1992; Rogawski, 1992), it is reasonable to assume that by inhibiting kynurenine hydroxylase it is possible to reduce NMDA receptor function in vivo (Moroni et al., 1991; Russi et al., 1992). Activation of NMDA receptors plays a key role in the induction of post-ischaemic neuronal damage and antagonists of this receptor complex reduce the dramatic consequences of focal or global brain ischaemia (Simon et al., 1984; Meldrum, 1985; Gill et al., 1987; Choi, 1988; Olney, 1990; Pellegrini-Giampietro et al., 1994). However, the administration of competitive or non-competitive NMDA receptor antagonists may cause neuronal toxicity (Olney et al., 1989) and a long series of other side effects resulting in a benefit-to-risk ratio that is not acceptable for clinical studies (Small and Buchan, 1997). Kynurenine hydroxylase inhibitors have therefore been tested as neuro-

protectant agents in models of brain ischaemia primarily because of their properties of increasing kynurenate concentration in the brain. It should also be mentioned that their systemic administration is also expected to decrease brain concentrations of 3OH-kynurenine, a metabolite provided with excitotoxic actions in neuronal cell lines (Eastman and Guilarte, 1989) and able to cause, in a concentration-dependent manner, either necrotic or apoptotic types of neuronal death (Okuda et al., 1998). Structure–activity studies have in fact shown that 3OH-kynurenine, and other *o*-aminophenols, may be subject to oxidative reactions initiated by their conversion to quinonimines, a process associated with concomitant production of oxygen-derived free radicals (Hiraku et al., 1995). The involvement of these reactive species in the pathogenesis of ischaemic neuronal death has been widely studied in the last several years and it has been shown that oxygen-derived free radicals and glutamate-mediated neurotransmission co-operate in the development of ischaemic neuronal death (Pellegrini-Giampietro et al., 1990). Finally, another reason whereby kynurenine hydroxylase inhibitors have been studied in models of brain ischaemia is the commonly held concept that 3OH-kynurenine formation is one of the steps required for the synthesis of quinolinic acid, a compound provided with NMDA agonist and excitotoxic properties. It is now accepted that, under basal conditions, the administration of large doses of inhibitors of kynurenine hydroxylase do not modify basal blood or brain quinolinic acid content, while a significant decrease in blood or brain concentrations of this excitotoxin was observed when the inhibitors were administered to immune-stimulated animals (Moroni et al., 1991; Chiarugi and Moroni, 1999b). Until a few years ago, the only compound with kynurenine-hydroxylase inhibiting properties was nicotinyl-alanine, an analogue of kynurenine originally investigated as a metabolic intermediate for NAD biosynthesis (Decker et al., 1963). The specificity of this compound toward kynurenine hydroxylase is relatively poor (Moroni et al., 1991; Russi et al., 1992; Moroni et al., 1996). Several more specific inhibitors have been synthesised and tested in the last several years. The first of them was (*m*-nitrobenzoyl)-alanine (Carpenedo et al., 1994; Pellicciari et al., 1994; Natalini et al., 1995), a relatively potent competitive and selective inhibitor (IC_{50} 1μM; see Fig. 3). New, more potent and more selective compounds have been recently described by two pharmaceutical companies (Speciale et al., 1996; Roever et al., 1997) and their biochemical effects have been studied in vivo. Appropriate doses and

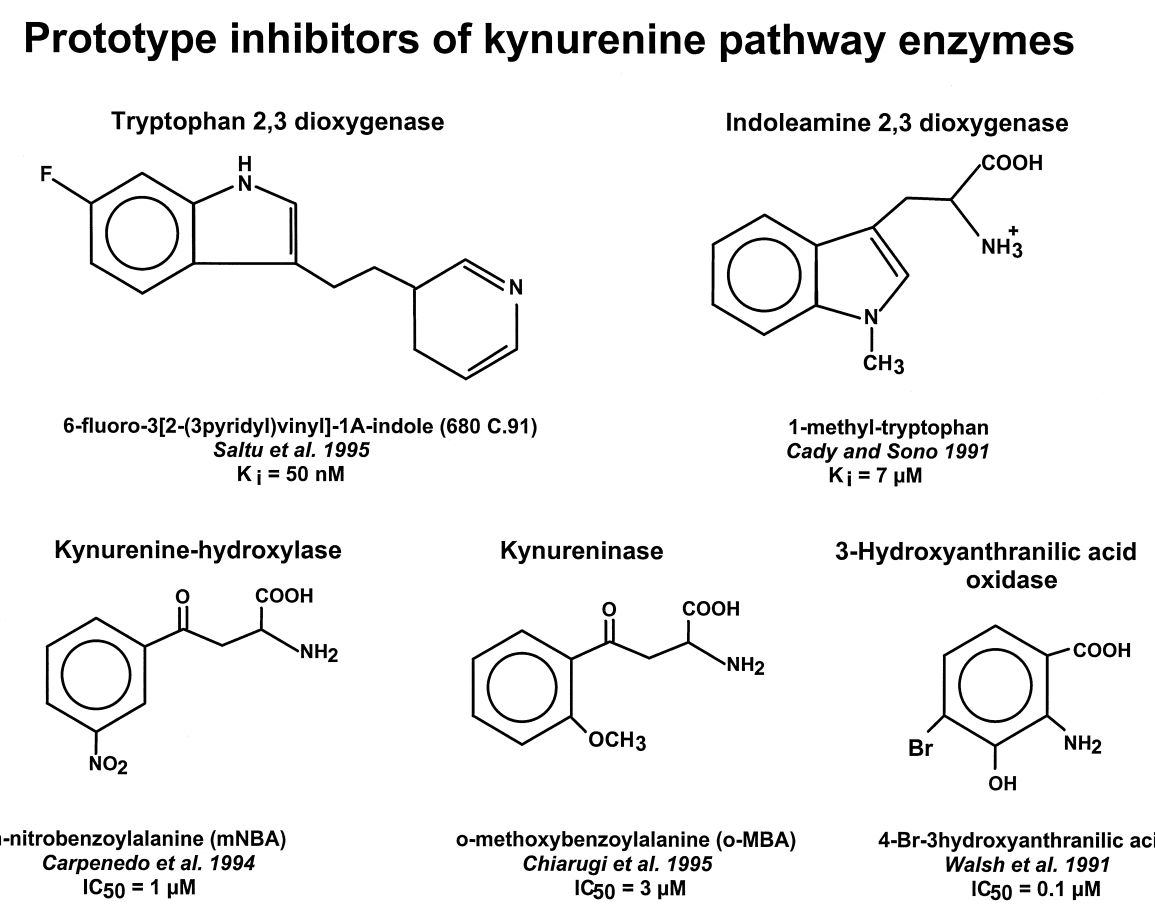

Fig. 3. Molecular structures of prototype inhibitors of kynurenine pathway enzymes. The potencies of the different inhibitors and the appropriate references are reported.

administration schedules of either (*m*-nitrobenzoyl)-alanine or 3,4-dimethoxy-[-*N*-4-(nitrophenyl)thiazol-2yl]-benzenesulfonamide (Ro 61-8048), one of the newly synthesised compounds, causes neuroprotective properties on brain lesions induced in gerbil hippocampus by transient bilateral carotid occlusion or in the rat cortex by middle cerebral artery occlusion (Cozzi et al., 1999). Neuroprotective effects of kynurenine hydroxylase inhibitors have been recently reported in organotypic hippocampal slice cultures exposed to oxygen and glucose deprivation (Moroni et al., 1999) and against excitotoxic striatal lesions in rats pretreated with amphetamine in order to reduce local kynurenic acid concentrations (Schwarcz et al., 1999). The above-mentioned results are therefore in line with the hypothesis that by shifting kynurenine metabolism toward kynurenic acid formation, it is possible to reduce glutamate receptor activation and excitotoxic or ischaemic neuronal damage.

5. 3OH-anthranilic and quinolinic acids

3OH-anthranilic acid is present in most mammalian organs, including brain, at relatively low concentrations (nanomolar) (Baran and Schwarcz, 1999). Its synthesis may occur either through the hydrolysis of 3OH-kynurenine or through direct oxidation of anthranilic acid (see Fig. 2). Both pathways operate in rat brain slices (Baran and Schwarcz, 1999), but while the enzymatic activity catalysing the hydrolysis of 3OH-kynurenine has been carefully characterised, probably several oxidases are able to metabolise anthranilic acid into its 3-hydroxy metabolite. Interestingly, it has been recently reported that low micromolar concentrations of 3OH-anthranilic acid, when added to primary cultures of striatal neurones, cause apoptotic features similar to those induced by 3OH-kynurenine. This type of neurotoxicity seems to be ascribed to the *o*-aminophenol structure which is shared by both 3OH-kynurenine and 3OH-anthranilic acid. The compounds are taken up by the neurones where they generate reactive oxygen species able to trigger the apoptotic process (Okuda et al., 1998). At relatively large concentrations, 3OH-anthranilic acid has been shown to inhibit the expression of NO synthase in a macrophage cell line probably because it prevents nuclear factor-kB activation similar to other agents which may modify the cell redox state (Sekkai et al., 1997). Obviously, in physiological conditions and in most of the organs, the compound is rapidly metabolised and its concentration remains well below those able to induce toxicity.

As previously mentioned, kynureninase [E.C. 3.7.1.3.] is the main enzyme responsible for 3OH-anthranilic acid formation. In mammals, a single protein catalyses the cleavage of either 3OH-kynurenine or kynurenine, while in bacteria, two different enzymes, one active on 3OH-kynurenine and the second on kynurenine, have been described (McDermott et al., 1973). Mammalian kynureninase is a pyridoxal phosphate-dependent enzyme predominantly located in the cytosol, and it has been purified and characterised mostly from rodent liver, kidney or spleen (Kawai et al., 1988). The purified enzyme, besides acting on the kynurenines, also has a cysteine conjugate β-lyase enzymatic activity and cleaves the *S*-cysteine conjugates of certain halogenated xenobiotics into possibly nephrotoxic thiols (Stevens, 1985). The rat and human enzymes have been cloned and expressed and it has been confirmed that the affinity of both native or expressed enzymes is approximately 10-fold higher for 3OH-kynurenine than for kynurenine (K_m for kynurenine 250 and K_m for 3OH-kynurenine 25 μM) (Alberati-Giani et al., 1996a; Toma et al., 1997). Interestingly, γ-interferon induces kynureninase activity in rat macrophages but not in rat microglial cells (Alberati-Giani et al., 1996b). Several inhibitors of bacterial kynureninase have been designed and studied with the aim of understanding the molecular basis of the enzymatic activity (Dua et al., 1993). In rodents, the inhibition of the enzyme obtained with systemic administration of *o*-methoxybenzoylalanine (*o*MBA), a prototype inhibitor of the mammalian enzyme (see Fig. 3), leads to a modest accumulation of kynurenic acid in the brain and prevents audiogenic convulsion in DBA2 mice (Carpenedo et al., 1994; Chiarugi et al., 1995, 1996). It has been difficult to further characterise the metabolic effects of the kynureninase inhibitors in vivo because *o*MBA, an extremely selective inhibitor in vitro, inhibits not only kynureninase but also 3OH-anthranilic acid 3,4-dioxygenase (E.C. 1.13.11.6) in complex systems and in vivo (Chiarugi and Moroni, 1999a).

3OH-anthranilate-3,4-dioxygenase is the enzyme able to cleave the benzene ring of 3OH-anthranilate into α-amino-β-carboxymuconate-ε-semialdeide, a relatively unstable compound which largely undergoes spontaneous cyclisation with the formation of quinolinic acid. A portion of α-amino-β-carboxymuconate-ε-semialdeide is metabolised by α-amino-β-carboxymuconate-ε-semialdeide decarboxylase (E.C. 4.1.1.45) with the formation of picolinic or aminomuconic acids (total oxidative pathway, see Fig. 2).

3OH-anthranilate-3,4-dioxygenase has been purified from mammalian liver or kidney and it has been shown that it requires both ferrous ions (Fe^{2+}) and sulfidryl groups for its activity (Long et al., 1954; Decker et al., 1961; Koontz and Shiman, 1976). The enzyme is also present, although in small amounts, in mammalian brains (Foster et al., 1986) and immunohistochemical studies, using selective antibodies have shown that it is contained in astrocytes. In the striatum, the enzyme is present in astrocytic processes which surround glutamate-containing axon terminals (Koehler et al., 1988; Roberts et al., 1995). An increase in cerebral 3OH-anthranilate-3,4-dioxygenase activity has been reported in autoptic material of Huntington's disease patients (Schwarcz et al., 1988), in gerbil hippocampus after global forebrain ischaemia (Saito et al.,

1993c) and in a rat epilepsy model (Du et al., 1993). Most of the brain enzymatic activity seems located in the soluble cytosol, but immunoreactivity has also been detected in the mitochondria (Koehler et al., 1988). The mitochondrial enzyme seems to have a different molecular weight and a lower affinity for the substrate. It has been observed that the mitochondria may change 3OH-anthranilate-3,4-dioxygenase's properties: in the presence of these organelles, enzymatic activity is reduced by *o*MBA, a compound which was considered a selective inhibitor of kynureninase and which does not affect purified cytosolic 3OH-anthranilate-3,4-dioxygenase (Chiarugi and Moroni, 1999a). Recently, human 3OH-anthranilate-3,4-dioxygenase has been cloned and functionally expressed. The recombinant enzyme has an affinity for 3OH-anthranilate very similar to that found in purified rat cytosol enzyme (K_m 2 μM; Malherbe et al., 1994). Halogen derivatives of 3OH-anthranilate (see Fig. 3) are potent inhibitors of this enzyme (Parli et al., 1980) and reduce quinolinic acid formation in stimulated monocytes in vitro and in brain or blood of immune-activated mice in vivo (Saito et al., 1993a, 1994).

Quinolinic acid is formed from the spontaneous cyclisation of α-amino-β-carboxymuconate-ϵ-semialdeide, the direct product of 3OH-anthranilate-3,4-dioxygenase activity. Its biological properties are probably responsible for much of the current interest in the regulation of the kynurenine pathway enzymes. Quinolinic acid interacts with a subgroup of NMDA receptors (Stone and Perkins, 1981; Perkins and Stone, 1982a) and when directly injected into brain areas, it destroys most neuronal cell bodies sparing fibres en passant and neuronal terminals (Schwarcz et al., 1983). Much of the original observation showing that cortical NMDA receptors are more sensitive to quinolinic acid than those located in the cerebellum or the spinal cord (Perkins and Stone, 1983; McLennan, 1984) and which suggest the existence of multiple forms of NMDA receptors (Monaghan and Beaton, 1991; Moroni et al., 1995) can probably be explained by considering that quinolinic acid (similar to its analogue homoquinolinic acid) is a relatively poor agonist for the NMDA receptor complex containing either NR2C or NR2D subunits, while it interacts with a relatively high affinity with the NMDA receptor complex containing NR2B subunits (Brown et al., 1998). The neurotoxicity profile found after intrastriatal injection of quinolinic acid closely resembles that found in the basal nuclei of Huntington's disease patients: while most of the intrinsic striatal neurones are destroyed, NADH-diaphorase-staining neurones (which are now considered able to express NO synthetase) and neurones containing neuropeptide Y seem to be spared together with axon terminals and fibres en passant (Ferrante et al., 1985; Beal et al., 1986, 1989). The concentration of quinolinic acid causing neurotoxicity in vivo, after direct local injection, are quite elevated and may be considered at least two orders of magnitude larger than those found in mammalian brains or cerebrospinal fluid (CSF) under basal conditions (Moroni et al., 1984b; Wolfensberger et al., 1984; Heyes and Markey, 1988). In vitro, the neurotoxic effects of the compound have been studied in different model systems with variable results: chronic exposure of organotypic cortico-striatal cultures to submicromolar concentrations of quinolinic acid causes histological signs of pathology (Whetsell and Schwarcz, 1989), but elevated concentrations of quinolinic acid (in the low millimolar range) are necessary to cause excitotoxic neuronal death in primary cultures of mixed cortical neurones when brief (20 min) exposure times are used (Kim and Choi, 1987). Mammalian brain quinolinic acid concentrations are quite low (0.05–0.1 μM) and are approximately one order of magnitude lower that those in the blood, thus suggesting that, under basal conditions, the blood contained in the brain vessels may significantly contribute to the measurement of the overall brain quinolinic acid concentration (Beagles et al., 1998). In models of inflammatory neurological disorders, such as experimental allergic encephalitis (Flanagan et al., 1995), bacterial and viral infections (Heyes et al., 1992; Espey et al., 1996), forebrain global ischaemia or spinal trauma (Heyes and Nowak, 1990; Blight et al., 1995), brain quinolinic acid levels are extremely elevated (Heyes and Morrison, 1997; Beagles et al., 1998). This increased brain quinolinate concentration could be due to either an elevated circulating concentration of the excitotoxin or to an increased de novo synthesis in activated microglia or infiltrating macrophages (Heyes et al., 1996, 1997). In retrovirus-infected macaques, it has been proposed that most of the increased content in brain quinolinic acid (approximately 98%) is due to local production. In fact, a robust increase in the activities of indoleamine 2,3-dioxygenase, kynurenine hydroxylase and kynureninase has been found in areas of brain inflammation (Heyes et al., 1998).

6. Oxindole

In the course of experiments performed to study the neurochemical effects of kynurenine metabolism inhibitors (Carpenedo et al., 1994; Chiarugi et al., 1995), we obtained data suggesting that tryptophan may be metabolised into oxindole (Fig. 1) and that oxindole is physiologically present in blood and other biological fluids in concentrations ranging from 0.1 to 1 μM (Carpenedo et al., 1997). Systemic administration of oxindole to rats, dogs or humans has been shown to cause profound sedation, decreased blood pressure, decreased muscular tone and loss of consciousness (Orcutt et al., 1964). A number of studies have been performed to understand oxindole's mechanism of action, and it has been shown that it does not modify the depolarisation induced by AMPA or NMDA in cortical slices, thus suggesting that it does not affect ionotropic glutamate receptor function. Furthermore, it does not displace the binding of [^{3}H]GABA to either $GABA_A$ or

$GABA_B$ recognition sites in brain membranes, ^{3}H-flunitrazepam to the benzodiazepine recognition sites and ^{3}H-glycine or ^{3}H-glutamate to the NMDA receptor complex. In rat hippocampal slices in vitro, oxindole effects have been evaluated on the neurotransmission of the CA1 region following orthodromic stimulation of the Schaffer collaterals and extra or intracellular recordings. At 0.3–3 mM, the compound decreases the population spike amplitudes extracellularly recorded at a somatic level and the field excitatory postsynaptic potentials recorded at a dendritic level. In intracellular recordings, oxindole (0.1–1 mM) does not affect the resting membrane potential or the neuronal input resistance, but reduces the probability of firing action potentials upon both synaptic or direct activation of the pyramidal cells. It also increases the threshold and the latency of firing action potentials elicited by depolarising steps without changing the duration or the peak amplitude of the spikes. The action potential frequency adaptation (accommodation) induced by long lasting (400 ms) depolarising stimuli is significantly increased by oxindole. It has therefore been proposed that concentrations of oxindole, which may be found in pathological conditions, significantly decreases neuronal excitability by possibly acting on voltage-operated Na^+ channels.

Since rats treated with oxindole or having acute liver impairment share several common signs and since it is widely accepted that tryptophan metabolites are involved in the neurological signs associated with liver damage (see references in: Bengtsson et al., 1991; Bengtsson and Bergqvist, 1996), brain oxindole content in rat models of acute liver impairment have been measured. As expected, a very large increase (10–100 fold) in blood and brain oxindole content has been found in rats with acute liver failure or in patients with cirrhosis and hepatic encephalopathy, thus suggesting that the compound could play an important role in the neurological symptoms associated with liver failure (Carpenedo et al., 1998; Moroni et al., 1998b). Large increases in oxindole concentrations have also been found in urine samples obtained from a subpopulation of patients affected by phenylchetonuria, an hereditary disorder of the amino acid metabolism. Although the biochemical mechanisms responsible for such increases have not been studied, it has been suggested that the compound could have a role in the pathophysiology of the syndrome. It seems, therefore, that oxindole is a neuroactive metabolite that should be added to the already long list of compounds originating from tryptophan which are able to affect brain function in physiology or pathology (Allegri Filippini et al., 1997).

7. Conclusion

The number of tryptophan metabolites which, at reasonable concentrations, have been shown to interact with neurotransmitter receptors, ion channels, carriers and other membrane or intracellular structures of excitable cells is quite elevated. In pathological conditions, the concentrations of these neuroactive compounds may significantly change in blood and brain. It is therefore reasonable to assume that agents which selectively affect the enzymes involved in their formation or metabolism (see Fig. 3) are useful tools to improve our understanding on the role 'kynurenines' play in brain physiology and pathology.

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References

Alberati-Giani, D., Malherbe, P., Koehler, R.C., Lang, G., Kiefer, V., Lahm, H.W., Cesura, A.M., 1995. Cloning and characterization of a soluble kynurenine aminotransferase from rat brain: identity with kidney cysteine conjugate beta-lyase. J. Neurochem. 64, 1448–1455.

Alberati-Giani, D., Buchli, R., Malherbe, P., Broger, C., Lang, G., Koehler, R.C., Lahm, H.W., Cesura, A.M., 1996a. Isolation and expression of a cDNA clone encoding human kynureninase. Eur. J. Biochem. 239, 460–468.

Alberati-Giani, D., Ricciardi-Castagnoli, P., Koehler, R.C., Cesura, A.M., 1996b. Regulation of the kynurenine pathway by interferon-gamma in murine cloned macrophages and microglial cells. In: Allegri Filippini, G., Costa, C.V.L., Bertazzo, A. (Eds.), Recent Advances in Tryptophan Research. Plenum, New York, pp. 171–175.

Alberati-Giani, D., Cesura, A.M., Broger, C., Warren, W.D., Roever, S., Malherbe, P., 1997. Cloning and functional expression of human kynurenine 3-monooxygenase. FEBS Lett. 410, 407–412.

Allegri Filippini, G., Costa, C.V.L., Bertazzo, A., 1997. Recent Advances in Tryptophan Research: Tryptophan and Serotonin Pathways. Plenum, New York.

Baran, H., Schwarcz, R., 1999. Presence of 3-hydroxyanthranilic acid in rat tissues and evidence for its production from anthranilic acid in the brain. J. Neurochem. 55, 738–744.

Beagles, K.E., Morrison, P.F., Heyes, M.P., 1998. Quinolinic acid: in vivo synthesis rates, extracellular concentrations, and intercompartmental distributions in normal and immune-activated brain as determined by multiple-isotope microdialysis. J. Neurochem. 70, 281–291.

Beal, M.F., Kowall, N.W., Ellison, D.W., Mazurek, M.F., Swartz, K.J., 1986. Replication of the neurochemical characteristics of Huntington's disease by quinolinic acid. Nature 321, 168–171.

Beal, M.F., Kowall, N.W., Swartz, K.J., Ferrante, R.J., Martin, J.B., 1989. Differential sparing of somatostatin-neuropeptide Y and cholinergic neurones following striatal excitotoxin lesions. Synapse 3, 38–47.

Bender, D.A., 1989. The kynurenine pathway of tryptophan metabolism. In: Stone, T.W. (Ed.), Quinolinic Acid and the Kynurenines. CRC Press, Boca Raton, FL, pp. 3–24.

Bender, D.A., McCreanor, G.M., 1982. The preferred route of kynurenine metabolism in the rat. Biochim. Biophys. Acta 717, 56–60.

Bengtsson, F., Bergqvist, P.B.F., 1996. Neuropsychiatric implications of brain tryptophan: perturbations appearing in hepatic encephalopathy. In: Allegri Filippini, G., Costa, C.V.L., Bertazzo A. (Eds.), Recent Advances in Tryptophan Research. Plenum, New York, pp. 387–395.

Bengtsson, F., Bugge, M., Johansen, K.H., Butterworth, R.F., 1991. Brain tryptophan hydroxylation in the portocaval shunted rat: a hypothesis for the regulation of serotonin turnover in vivo. J. Neurochem. 56, 1069–1074.

Blight, A.R., Cohen, T.I., Saito, K., Heyes, M.P., 1995. Quinolinic acid accumulation and functional deficits following experimental spinal cord injury. Brain 118, 735–752.

Bonney, E.A., Matzinger, P., 1998. Much IDO about pregnancy. Nature Medicine 4, 1178–1179.

Brown, J.C.l., Tse, H., Skifter, D.A., Christies, J.M., Andaloro, V.J., Kemp, M.C., Watkins, J.C., Jane, D.E., Monaghan, D.T., 1998. [^{3}H]Homoquinolinic acid binds to a subpopulation of NMDA receptors and to a novel binding site. J. Neurochem. 71, 1464–1470.

Buchli, R., Alberati-Giani, D., Malherbe, P., Koehler, R.C., Broger, C., Cesura, A.M., 1995. Cloning and functional expression of a soluble form of kynurenine/alpha-aminoadipate aminotransferase from rat kidney. J. Biol. Chem. 270, 29330–29335.

Burke, F., Knowles, R.G., East, N., Balkwill, F.R., 1995. The role of indoleamine 2,3-dioxygenase in the anti-tumour activity of human interferon-gamma in vivo. Int. J. Cancer 60, 115–122.

Cady, S.G., Sono, M., 1991. 1-Methyl-DL-tryptophan,-(3-benzofuranyl)DL-alanine (the oxygen analog of tryptophan) and -[3-benzo(*b*)thionyl]-DL-alanine (the sulfur analog of tryptophan) are competitive inhibitors for indoleamine 2,3-dioxygenase. Arch. Biochem. Biophys. 291, 326–332.

Carlin, J.M., Ozaki, Y., Byrne, G.I., Brown, R.R., Borden, E.C., 1989. Interferons and indoleamine 2,3-dioxygenase: role in antimicrobial and antitumor effects. Experientia 45, 535–541.

Carpenedo, R., Chiarugi, A., Russi, P., Lombardi, G., Carlà, V., Pellicciari, R., Mattol, L., Moroni, F., 1994. Inhibitors of kynurenine hydroxylase and kynureninase increase cerebral formation of kynurenic acid and have sedative and anticonvulsant activities. Neuroscience 61, 237–244.

Carpenedo, R., Carlà, V., Moneti, G., Chiarugi, A., Moroni, F., 1997. Identification and measurement of oxindole (2-indolinone) in the mammalian brain and other rat organs. Anal. Biochem. 244, 74–79.

Carpenedo, R., Mannaioni, G., Moroni, F., 1998. Oxindole, a neurodepressant tryptophan metabolite, accumulates in blood or brain of rats with hepatic failure. J. Neurochem. 70, 1998–2003.

Cesura, A.M., Alberati-Giani, D., Buchli, R., Broger, C., Koehler, R.C., Vilbois, F., Lahm, H.W., Heitz, M.P., Malherbe, P., 1996. Molecular characterisation of kynurenine pathway enzymes. In: Allegri Filippini, G., Costa, C.V.L., Bertazzo, A. (Eds.), Recent Advances in Tryptophan Research. Plenum, New York, pp. 477–485.

Chiarugi, A., Moroni, F., 1999a. Effects of mitochondria and *o*-methoxybenzoylalanine on 3-hydroxyanthranilic acid dioxygenase activity and quinolinic acid synthesis, J. Neurochem., in press.

Chiarugi, A., Moroni, F., 1999b. Quinolinic acid formation in immune-activated mice: studies with (*m*-nitrobenzoyl)-alanine (mNBA) and 3,4-dimethoxy-[-*N*-4-(-3-nitrophenyl)thiazol-2yl]-benzenesulfonamide (Ro 61-8048), two potent and selective inhibitors of kynurenine hydroxylase. Neuropharmacology, in press.

Chiarugi, A., Carpenedo, R., Molina, M.T., Mattoli, L., Pellicciari, R., Moroni, F., 1995. Comparison of the neurochemical and behavioral effects resulting from the inhibition of kynurenine hydroxylase and/or kynureninase. J. Neurochem. 65, 1176–1183.

Chiarugi, A., Carpenedo, R., Moroni, F., 1996. Kynurenine disposition in blood and brain of mice: effects of selective inhibitors of kynurenine hydroxylase and of kynureninase. J. Neurochem. 67, 692–698.

Choi, D.W., 1988. Glutamate neurotoxicity and diseases of the nervous system. Neuron 1, 623–634.

Connick, J., Heywood, G.C., Sills, G.J., Thomson, G.G., Brodie, M.J., Stone, T.W., 1992. Nicotinylalanine increases cerebral kynurenic acid content and has anticonvulsant activity. Gen. Pharmacol. 23, 235–239.

Cooper, A.J., Abraham, D.G., Gelbard, A.S., Lai, J.C.K., Petito, C.K., 1993. High activities of glutamine transaminase K (dichlorovinylcysteine-lyase) and omega-amidase in the choroid plexus of rat brain. J. Neurochem. 61, 1731–1737.

Cozzi, A., Carpenedo, R., Moroni, F., 1999. Kynurenine hydroxylase inhibitors reduce ischemic brain damage: studies with (*m*-nitrobenzoyl)-alanine (mNBA) and 3,4-dimethoxy-[-*N*-4-(nitrophenyl)-thiazol-2yl]-benzensulfonamide (Ro 61-8048) in models of focal or global brain ischemia. J. Cereb. Blood Flow Metab., in press.

Dai, W., Gupta, S.L., 1990a. Molecular cloning, sequencing and expression of human interferon-gamma-inducible indoleamine 2,3-dioxygenase cDNA. Biochem. Biophys. Res. Commun. 169, 1–8.

Dai, W., Gupta, S.L., 1990b. Regulation of indoleamine 2,3-dioxygenase gene expression in human fibroblasts by interferon gamma. J. Biol. Chem. 265, 19871–19877.

Danesh, U., Hashimoto, S., Renkawitz, R., Schutz, G., 1983. Transcriptional regulation of the tryptophan oxygenase gene in rat liver by glucocorticoids. J. Biol. Chem. 258, 4750–4753.

Danesh, U., Gloss, B., Schmid, W., Schutz, G., Schule, R., Renkawitz, R., 1987. Glucocorticoid induction of the rat tryptophan oxygenase gene is mediated by two widely separated glucocorticoid-responsive elements. EMBO J. 6, 625–630.

DeCastro, F.T., Price, J.M., Brown, R.R., 1956. Reduced triphosphopyridinenucleotide requirement for the enzymatic formation of 3OH-L-kynurenine from L-kynurenine. J. Am. Chem. Soc. 78, 2904–2905.

Decker, R.H., Kang, H.H., Leach, F.R., Henderson, L.M., 1961. Purification and properties of 3-hydroxyanthranilic acid oxidase. J. Biol. Chem. 236, 3076–3082.

Decker, R.H., Brown, R.R., Price, J.M., 1963. Studies on the biological activity of nicotinylalnine, an analogue of kynurenine. J. Biol. Chem. 238, 1049–1053.

Dennison, Z., Ossenkop, K.P., Cain, D.P., 1992. Effects of central administration of kynurenic acid on spontaneous locomotor activity in kindled rat: a multivariate approach of using the automated Digiscan monitoring system. Pharmacol. Biochem. Behav. 43, 807–814.

Dong-Ruyl, L., Kondo, H., Furukawa, S., Nakano, K., 1997. Stimulation of NGF production by tryptophan and its metabolites in cultured mouse astroglial cells. Brain Res. 777, 228–230.

Dong-Ruyl, L., Sawada, M., Nakano, K., 1998. Tryptophan and its metabolite, kynurenine, stimulate expression of nerve growth factor in cultured mouse astroglial cells. Neurosci. Lett. 244, 17–20.

Du, F., Schmidt, W.J., Okuno, E., Kido, R., Koehler, R.C., Schwarcz, R., 1992. Localization of kynurenine aminotransferase immunoreactivity in the rat hippocampus. J. Comp. Neurol. 321, 477–487.

Du, F., Williamson, J., Bertram, E., Lothman, E.W., Okuno, E., Schwarcz, R., 1993. Kynurenine pathway enzymes in a rat model of chronic epilepsy: immunohistochemical study of activated glial cells. Neuroscience 55, 975–989.

Dua, R.K., Taylor, E.W., Phillips, R.S., 1993. *S*-aryl-L-cysteine *S,S*-dioxide: design, synthesis and evaluation of a new class of inhibitors of kynureninase. J. Am. Chem. Soc. 115, 1264–1270.

Eastman, C.L., Guilarte, T.R., 1989. Citotoxicity of 3-hydroxykynurenine in a neuronal hybrid cell line. Brain Res. 495, 225–231.

Erickson, J.B., Reinhard, J.F.J., Flanagan, E.M., Russo, S., 1992. A radiometric assay for kynurenine 3-hydroxylase based on the release of ^3H_2O during hydroxylation of L-[3.5-^{3}H]-kynurenine. Anal. Biochem. 205, 257–262.

Espey, M.G., Tang, Y., Morse, H.C., Moffet, J.R., Namboodiri, M.A.A., 1996. Localization of quinolinic acid in the murine AIDS model of retrovirus-induced immunodeficiency: implications for neurotoxicity and dendritic cell immunopathogenesis. AIDS 10, 151–158.

Feng, G., Taylor, M.W., 1989. Interferon-gamma resistant mutants are defective in the induction of indoleamine 2,3-dioxigenase. Proc. Natl. Acad. Sci. USA 86, 7144–7148.

Ferrante, R.J., Kowall, N.W., Beal, M.F., Richardson, E.P. Jr., Bird, E.D., Martin, J.B., 1985. Selective sparing of a class of striatal neurones in Huntington's disease. Science 230, 561–563.

Flanagan, E.M., Erickson, J.B., Viveros, O.H., Chang, S.Y., Reinhard, J.F.J., 1995. Neurotoxin quinolinic acid is selectively elevated in spinal cord of rats with experimental allergic encephalomyelitis. J. Neurochem. 64, 1192–1196.

Foster, A.C., Collines, J.F., Schwarcz, R., 1983. On the excitotoxic properties of quinolinic acid, 2,3-piperidine dicarboxylic acids and structurally related compounds. Neuropharmacology 22, 1331–1342.

Foster, A.C., White, R.J., Schwarcz, R., 1986. Synthesis of quinolinic acid by 3-hydroxyantranilic acid oxygenase in rat brain tissue in vitro. J. Neurochem. 47, 23–30.

Fukui, S., Schwarcz, R., Rapoport, S.I., Takada, Y., Smith, Q.R., 1995. Blood-barrier transport of kynurenines: implications for brain synthesis and metabolism. J. Neurochem. 56, 2007–2017.

Gill, R., Foster, A.C., Woodruff, G.N., 1987. Systemic administration of MK-801 protects against ischemia-induced hippocampal neuronal degeneration in the gerbil. J. Neurosci. 7, 3344–3349.

Gramsbergen, J.B.P., Hodgkins, P.S., Rassoulpour, A., Turski, W.A., Guidetti, P., Schwarcz, R., 1997. Brain-specific modulation of kynurenic acid synthesis in the rat. J. Neurochem. 69, 290–298.

Green, A.R., Woods, H.F., Knott, P.J., Curzon, G., 1975. Factors influencing effect of hydrocortisone on rat brain tryptophan metabolism. Nature 255, 170.

Guidetti, P., Okuno, E., Schwarcz, R., 1997. Characterization of rat brain kynurenine aminotransferases I and II. J. Neurosci. Res. 50, 457–465.

Guilarte, T.R., Wagner, H.N., 1987. Increased concentrations of 3-hydroxykynurenine in vitamin B-6 deficient neonatal rat brain. J. Neurochem. 49, 1918–1926.

Hayaishi, O., Yoshida, R., Takikawa, O., Yasui, H., 1984. Indoleamine dioxygenase—a possible biological function. In: Schlossberger, H.G., Kochen, W., Linzen B., Steinhart, H. (Eds.), Progress in Tryptophan and Serotonin Research. De Gruiter, W. Berlin, pp. 33–42.

Heyes, M.P., Markey, S.P., 1988. Quantification of quinolinic acid in the rat brain, whole blood and plasma by gas-chromatography and negative chemical ionization mass spectrometry: effects of systemic tryptophan administration on brain and blood quinolinic acid concentrations. Anal. Biochem. 174, 349–359.

Heyes, M.P., Nowak, T.S.J., 1990. Delayed increase in regional brain quinolinic acid follow transient ischemia in the gerbil. J. Cereb. Blood Flow Metab. 10, 660–667.

Heyes, M.P., Morrison, P.F., 1997. Quantification of local de novo synthesis versus blood contributions to quinolinic acid concentrations in brain and systemic tissues. J. Neurochem. 68, 280–288.

Heyes, M.P., Saito, H., Crowley, J.S., Davies, L.E., Demitrak, M.A., Der, M., Dilling, L., Kruesi, M.J.P., Lackner, A., Larsen, S.A., Lee, K., Leonard, H., Markey, S.P., Martin, A., Milstien, S., Mouradian, M.M., Pranzatelli, M.R., Quearry, B.J., Salazar, A.M., Smith, M.L., Straus, S.E., Sunderland, T., Swedo, S., Tourtelotte, W.W., 1992. Quinolinic acid and kynurenine pathway metabolism in inflammatory and non-inflammatory neurologic disease. Brain 115, 1249–1273.

Heyes, M.P., Alchim, C.L., Wiley, A., Major, E.O., Saito, K., Markey, S.P., 1996. Human microglia convert L-tryptophan into the neurotoxin quinolinic acid. Biochem. J. 320, 595–597.

Heyes, M.P., Chen, C.Y., Major, E.O., Saito, K., 1997. Different kynurenine pathway enzymes limit quinolinic acid formation by various human cell types. Biochem. J. 326, 351–356.

Heyes, M.P., Saito, K., Lackner, A., Wiley, A., Achim, C.L., Markey, S.P., 1998. Sources of the neurotoxin quinolinic acid in the brain of HIV-1-infected patients and retrovirus-infected macaques. FASEB J. 12, 881–896.

Hiraku, Y., Inoue, S., Oikawa, S., Yamamoto, K., Tada, S., Nishino, K., Kawanishi, S., 1995. Metal-mediated oxidative damage to cellular and isolated DNA by certain tryptophan metabolites. Carcinogenesis 16, 349–356.

Hirata, H., Hayaishi, O., 1975. Studies on Indoleamine 2,3-dioxygenase. Superoxide anion as substrate. J. Biol. Chem. 250, 5960–5966.

Hodgkins, P.S., Schwarcz, R., 1998. Interference with cellular energy metabolism reduces kynurenic acid formation in rat brain slices: reversal by lactate and pyruvate. Eur. J. Neurosci. 10, 1986–1994.

Kapoor, R., Okuno, E., Kido, R., Kapoor, V., 1997. Immuno-localization of kynurenine aminotransferase in the rat medulla and spinal cord. NeuroReport 8, 3619–3623.

Kawai, J., Okuno, E., Kido, R., 1988. Organ distribution of rat kynureninase and changes of its activity during development. Enzyme 39, 181–189.

Kawata, Y., Tani, S., Sato, M., Katsube, Y., Tokushige, M., 1991. Preliminary X-ray crystallographic analysis of tryptophanase from *Escherichia coli*. FEBS Lett. 284, 270–272.

Kim, J.P., Choi, D.W., 1987. Quinolinic acid neurotoxicity in cortical cell culture. Neuroscience 23, 423–432.

Knox, W.E., Mehler, A.H., 1951. The adaptive increase of the tryptophan peroxidase oxidase system of liver. Science 113, 237–238.

Koehler, R.C., Eriksson, L.G., Okuno, E., Schwarcz, R., 1988. Localization of quinolinic acid metabolising enzymes in the rat brain: immunohistochemical studies using antibodies to 3-hydroxyanthranilic acid oxygenase and quinolinic acid phosphoribosyltransferase. Neuroscience 27, 49–76.

Konan, K.V., Taylor, M.W., 1996. Importance of the two interferon-stimulated response element (ISRE) sequences in the regulation of the human indoleamine 2,3-dioxygenase gene. J. Biol. Chem. 271, 19140–19145.

Koontz, W.A., Shiman, R., 1976. Beef kidney 3-hydroxyanthranilic acid oxygenase. J. Biol. Chem. 251, 368–377.

Kotake, Y., Masayama, T., 1937. Uber den mechanismus der kynurenine-bildung aus tryptophan. Hoppe-Seyler's Z. Physiol. Chem. 243, 237–244.

Lapin, I.P., 1976. Depressor effect of kynurenine and its metabolites in rats. Life Sci. 19, 1479–1484.

Lapin, I.P., 1980. Experimental studies on kynurenine as neuroactive tryptophan metabolites: past, present and future. Trends Pharmacol. Sci. 1, 410–413.

Lapin, I.P., 1981. Kynurenines and seizures. Epilepsia 22, 257–265.

Lapin, I.P., 1982. Convulsant action of intracerebroventricularly administered kynurenine sulphate, quinolinic acid and other derivatives of succinic acid and effects of amino acids: structure–activity relationships. Neuropharmacology 21, 1227–1233.

Long, C.L., Hill, H.N., Weinstock, I.M., Henderson, L.M., 1954. Studies on the enzymatic transformation of 3-hydroxyanthranilic acid to quinolinic acid. J. Biol. Chem. 205, 411–413.

MacKenzie, C.R., Hadding, U., Daubener, W., 1998. Interferon-gamma-induced activation of indoleamine 2,3-dioxigenase in cord blood monocyte-derived macrophages inhibits the growth of group B *streptococci*. J. Infect. Dis. 178, 875–878.

Maezono, K., Tashiro, K., Ichihara, A., 1990. Deduced primary structure of rat tryptophan-2,3-dioxygenase. Biochem. Biophys. Res. Commun. 170, 176–181.

Malherbe, P., Koehler, R.C., Da Prada, M., Lang, G., Kiefer, V., Schwarcz, R., Lahm, H.W., Cesura, A.M., 1994. Molecular cloning and functional expression of human 3-hydroxyanthranilic acid dioxygenase. J. Biol. Chem. 269, 13792–13797.

Malherbe, P., Alberati-Giani, D., Koehler, R.C., Cesura, A.M., 1995. Identification of a mitochondrial form of kynurenine aminotransferase/glutamine transaminase K from rat brain. FEBS Lett. 367, 141–144.

Mason, M., 1954. The kynurenine transaminase of rat kidney. J. Biol. Chem. 211, 839–844.

McDermott, C.E., Casciano, D.A., Gaertner, F.H., 1973. Isolation and characterization of an hydroxykynureninase from homogenates of adult mouse liver. Biochem. Biophys. Res. Commun. 51, 813–818.

McLennan, H., 1984. A comparison of the effects of NMDA and quinolinic acid on central neurones of the rat. Neurosci. Lett. 46, 157–160.

Mehler, A.H., Knox, W.E., 1950. Conversion of tryptophan to kynurenine in liver: II. The enzymatic hydrolysis of formylkynurenine. J. Biol. Chem. 187, 431–433.

Meldrum, B., 1985. Possible therapeutic applications of antagonists of excitatory amino acid neurotransmitters. Clin. Sci. 68, 113–122.

Melillo, G., Cox, G.W., Biragyn, A., Sheffler, L.A., Varesio, L., 1994. Regulation of nitric-oxide synthetase mRNA expression by interferon-gamma and picolinic acid. J. Biol. Chem. 269, 8128–8133.

Miranda, A.F., Boegman, R.J., Beninger, R.J., Jhamandas, K., 1997. Protection against quinolinic acid mediated excitotoxicity in nigrostri-

atal dopaminergic neurones by endogenous kynurenic acid. Neuroscience 78, 967–975.

Monaghan, D.T., Beaton, J.A., 1991. Quinolinic acid differentiates between forebrain and cerebellar NMDA receptors. Eur. J. Pharmacol. 194, 123–125.

Moroni, F., Lombardi, G., Carlà, V., Moneti, G., 1984a. The excitotoxin quinolinic acid is present and unevenly distributed in the rat brain. Brain Res. 295, 352–355.

Moroni, F., Lombardi, G., Moneti, G., Aldinio, C., 1984b. The excitotoxin quinolinic acid is present in the brain of several animal species and its cortical content increases during the aging process. Neurosci. Lett. 47, 51–55.

Moroni, F., Russi, P., Lombardi, G., Beni, M., Carlà, V., 1988. Presence of kynurenic acid in the mammalian brain. J. Neurochem. 51, 177–181.

Moroni, F., Russi, P., Gallo-Mezo, M.A., Moneti, G., Pellicciari, R., 1991. Modulation of quinolinic and kynurenic acid content in the rat brain: effects of endotoxins and nicotinylalanine. J. Neurochem. 57, 1630–1635.

Moroni, F., Galli, A., Mannaioni, G., Carlà, V., Cozzi, A., Mori, F., Marinozzi, M., Pellicciari, R., 1995. NMDA receptor heterogeneity in mammalian tissues: focus on two agonists, (2*S*,3*R*,4*S*) cyclopropylglutamate and the sulphate ester of 4-hydroxy-(*S*)-pipecolic acid. Naunyn-Schmiedeberg's Arch. Pharmacol. 351, 371–376.

Moroni, F., Carpenedo, R., Chiarugi, A., 1996. Kynurenine hydroxylase inhibitors as tools to study the role of kynurenine metabolites in the central nervous system. In: Allegri Filippini, G., Costa, C.V.L., Bertazzo, A. (Eds.), Recent Advances in Tryptophan Research. Plenum, New York, pp. 203–210.

Moroni, F., Carpenedo, R., Venturini, M., Baraldi, M., Zeneroli, M.L., 1998a. Oxindole in pathogenesis of hepatic encephalopathy. Lancet 351, 9119.

Moroni, F., Cozzi, A., Lombardi, G., Sourtcheva, S., Leonardi, P., Carfì, M., Pellicciari, R., 1998b. Presynaptic mGlu1 type receptors potentiate transmitter output in the rat cortex. Eur. J. Pharmacol. 347, 189–219.

Moroni, F., Cozzi, A., Peruginelli, F., Carpenedo, R., Pellegrini-Giampietro, D.E., 1999. Neuroprotective effects of kynurenine-3-hydroxylase inhibitors in models of brain ischemia. In: Steinhart et al., H. (Eds.), ISTRY Meeting 1998. Plenum, in press.

Mosca, M., Cozzi, L., Breton, J., Speciale, C., Okuno, E., Schwarcz, R., Benatti, L., 1994. Molecular cloning of rat kynurenine aminotransferase: identity with glutamine transaminase K. FEBS Lett. 353, 21–24.

Munn, D.H., Zhou, M., Attwood, J.T., Bondarev, I., Conway, S.J., Marshall, B., Brown, C., Mellor, A.L., 1998. Prevention of allogenic fetal rejection by tryptophan catabolism. Science 281, 1191–1194.

Musso, T., Gusella, G.L., Brooks, A., Varesio, L., 1994. Interleukin-4 inhibits indoleamine 2,3-dioxygenase expression in human monocytes. Blood 83, 1408–1411.

Nakagami, Y., Saito, H., Katsuki, H., 1996. 3-Hydroxykynurenine toxicity on the rat striatum in vivo. Jpn. J. Pharmacol. 71, 183–186.

Natalini, B., Mattoli, L., Pellicciari, R., Carpenedo, R., Chiarugi, A., Moroni, F., 1995. Synthesis and activity of enantiopure (*S*) (*m*-nitrobenzoyl) alanine, a potent kynurenine-3-hydroxylase inhibitor. Bioorg. Med. Chem. Lett. 5, 3717–3719.

Nozaki, K., Beal, M.F., 1992. Neuroprotective effects of L-kynurenine on hypoxia-ischemia and NMDA lesions in neonatal rats. J. Cereb. Blood Flow Metab. 12, 400–407.

Okamoto, H., Hayaishi, O., 1970. Intramitochondrial localization of kynurenine aminotransferase. J. Biol. Chem. 245, 3603–3608.

Okuda, S., Nishiyama, N., Saito, H., Katsuki, H., 1998. 3-Hydroxykynurenine, an endogenous oxidative stress generator, causes neuronal cell death with apoptotic features and region selectivity. J. Neurochem. 70, 299–307.

Okuno, E., Kido, R., 1991. Kynureninase and kynurenine,3-hydroxylase in mammalian tissue. In: Schwarcz et al., R. (Eds.), Kynurenine and Serotonin Pathways. Plenum, New York, pp. 167–175.

Okuno, E., Nakamura, M., Schwarcz, R., 1991. Two kynurenine aminotransferases in human brain. Brain Res. 542, 307–312.

Olney, J.W., 1990. Excitotoxic amino acids and neuropsychiatric disorders. Annu. Rev. Pharmacol. Toxicol. 30, 47–71.

Olney, J.W., Labruyere, J., Price, M., 1989. Pathological changes induced in cerebrocortical neurones in phencyclidine related drugs. Science 244, 1360–1362.

Orcutt, J.A., Prytherch, J.P., Konicov, M., Michaelson, S.M., 1964. Some new compounds exhibiting selective CNS-depressant activities: Part 1. Preliminary observations. Arch. Int. Pharmacodyn. 152, 121–131.

Parli, C.J., Krieter, P., Schmidt, B., 1980. Metabolism of 6-chlorotryptophan to 4-chloro-3-hydroxyanthranilic acid: a potent inhibitor of 3-hydroxyanthranilic acid oxidase. Arch. Biochem. Biophys. 203, 161–166.

Pellegrini-Giampietro, D.E., Cherici, G., Alesiani, M., Carlà, V., Moroni, F., 1990. Excitatory amino acid release and free radical formation may cooperate in the genesis of ischemia-induced neuronal damage. J. Neurosci. 10, 1035–1041.

Pellegrini-Giampietro, D.E., Cozzi, A., Moroni, F., 1994. The glycine antagonist and free radical scavenger 7-Cl-thio-kynurenate reduces CA1 ischemic damage in the gerbil. Neuroscience 63, 701–709.

Pellicciari, R., Natalini, B., Costantino, G., Mahmoud, M.R., Mattoli, L., Sadeghpour, B.M., Moroni, F., Chiarugi, A., Carpenedo, R., 1994. Modulation of the kynurenine pathway in search for new neuroprotective agents. Synthesis and preliminary evaluation of (*m*-nitrobenzoyl)alanine, a potent inhibitor of kynurenine-3-hydroxylase. J. Med. Chem. 37, 647–655.

Perkins, M.N., Stone, T.W., 1982a. An iontophoretic investigation of the actions of convulsant kynurenines and their interactions with the endogenous excitant quinolinic acid. Brain Res. 247, 184–187.

Perkins, M.N., Stone, T.W., 1982b. Specificity of kynurenic acid as an antagonist of synaptic transmission in rat hippocampal slices. Neurosci. Lett. 18, 432–436.

Perkins, M.N., Stone, T.W., 1983. Pharmacological and regional variations of quinolinic acid-evoked excitations in the rat central nervous system. J. Pharmacol. Exp. Ther. 226, 551–557.

Perry, S.J., Schofield, M.A., MacFarlane, M., Lock, E.A., Gibson, G.C., Goldfarb, P.S., 1993. Isolation and expression of a cDNA coding for rat kidney cytosolic cysteine conjugate beta-lyase. Mol. Pharmacol. 43, 660–665.

Pfefferkorn, E.R., 1984. Interferon-gamma blocks the growth of toxoplasma Gondii in human fibroblasts by inducing the host cells to degrade tryptophan. Proc. Natl. Acad. Sci. USA, pp. 908–912.

Pinelli, A., Ossi, C., Colombo, R., Tofanetti, O., Spazzi, L., 1984. Experimental convulsions in rats induced by intraventricular administration of kynurenine and structurally related compounds. Neuropharmacology 23, 333–337.

Poeggeler, B., Rassoulpour, A., Guidetti, P., Wu, H.Q., Schwarcz, R., 1998. Potentiation of striatal NMDA excitotoxicity in the rat by D-amphetamine: a role for kynurenic acid?. Soc. Neurosci. Abstr. 24, 461.

Reinhard, J.F.J., Erickson, J.B., Flanagan, E.M., 1994. Quinolinic acid in neurological disease: opportunity for novel drug discovery. In: Advances in Pharmacology. Academic Press, London, pp. 85–118.

Reinhard, J.F.J., Flanagan, E.M., Madge, D.J., Iyer, R., Salter, M., 1996. Effects of 540C91 [(*E*)-3-[2-(4′-pyridyl)-vinyl]-1*H*-indole], an inhibitor of hepatic tryptophan dioxygenase, on brain quinolinic acid in mice. Biochem. Pharmacol. 51, 159–163.

Roberts, R.C., McCarthy, K.E., Du, F., Ottersen, O.P., Okuno, E., Schwarcz, R., 1995. 3-Hydroxyanthranilic acid oxygenase-containing astrocytic processes surround glutamate-containing axon terminals in the rat striatum. J. Neurosci. 12, 1150–1161.

Roever, S., Cesura, A.M., Huguenin, P., Kettler, R., Szente, A., 1997. Synthesis and biochemical evaluation of *N*-(4-phenylthiazol-yl)be-

nzenesulfonamides as high-affinity inhibitors of kynurenine 3-hydroxylase. J. Med. Chem. 40, 4378–4385.
Rogawski, M., 1992. The NMDA receptor, NMDA antagonists and epilepsy therapy. Drugs 44, 279–292.
Russi, P., Alesiani, M., Lombardi, G., Davolio, P., Pellicciari, R., Moroni, F., 1992. Nicotinylalanine increases the formation of kynurenic acid in the brain and antagonizes convulsions. J. Neurochem. 59, 2076–2080.
Saito, K., Chen, C.Y., Masana, M., Crowley, J.S., Markey, S.P., Heyes, M.P., 1993a. 4-Chloro-3-hydroxyanthranilate, 6-chlorotryptophan and norharmane attenuate quinolinic acid formation by interferon-gamma-stimulate monocytes (THP-1 cells). Biochem. J. 291, 11–14.
Saito, K., Crowley, J.S., Markey, S.P., Heyes, M.P., 1993b. A mechanism for increased quinolinic acid formation following acute systemic immune stimulation. J. Biol. Chem. 268, 15496–15503.
Saito, K., Nowak, T.S.J., Markey, S.P., Heyes, M.P., 1993c. Mechanism of delayed increase in kynurenine pathway metabolism in damaged brain regions following transient cerebral ischemia. J. Neurochem. 60, 180–192.
Saito, K., Markey, S.P., Heyes, M.P., 1994. 6-Chloro-D,L-tryptophan, 4-chloro-3-hydroxyanthranilate and dexamethasone attenuate quinolinic acid accumulation in brain and blood following systemic immune activation. Neurosci. Lett. 178, 211–215.
Salter, M., Hazelwood, R., Pogson, C.I., Iyer, R., Madge, D.J., 1995. The effects of a novel and selective inhibitor of tryptophan 2,3-dioxygenase on tryptophan and serotonin metabolism in rat. Biochem. Pharmacol. 49, 1435–1442.
Santamaria, A., Altagracia, M., Solis, F., Kravzov, J., Ordaz, H., Gonzalez, L., Rios, C., 1994. Kynurenine and probenecid prevent quinolinic acid-induced neurotoxicity in rat corpus striatum. Proc. West. Pharmacol Soc. 37, 29–30.
Scharfman, H.E., Ofer, A., 1997. Pretreatment with L-kynurenine, the precursor to the excitatory amino acid antagonist kynurenic acid, suppress epileptiform activity in combined entorhinal/hippocampal slices. Neurosci. Lett. 224, 115–118.
Schmid, W., Scherer, G., Danesh, U., Zentgraf, H., Matthias, P., Strange, C.M., Rowekamp, W., Schutz, G., 1982. Isolation and characterization of the rat tryptophan oxygenase gene. EMBO J. 1, 1287–1289.
Schutz, G., Chow, E., Feigelson, P., 1972. Regulatory properties of hepatic tryptophan oxygenase. J. Biol. Chem. 247, 5333–5337.
Schwarcz, R., Whetsell, W.O., Mangano, R.M., 1983. Quinolinic acid: an endogenous metabolite that produces axon-sparing lesions in rat brain. Science 219, 316–318.
Schwarcz, R., Okuno, E., White, R.J., Bird, E.D., Whetsell, W.O., 1988. 3OH-anthranilic acid oxygenase activity is increased in the brains of Huntington disease victims. Proc. Natl. Acad. Sci. USA 85, 4079–4081.
Schwarcz, R., Guidetti, P., Poeggeler, B., Wu, H.Q., Rassoulpour, A., 1999. Modulation of neuronal viability by neuroactive kynurenines. In: Steinhart et al., H. (Eds.), ISTRY 1998. Plenum, in press.
Sekkai, D., Guittet, O., Lemaire, G., Tenu, J.P., Lepoivre, M., 1997. Inhibition of nitric oxide synthase expression and activity in macrophages by 3-hydroxyanthranilic acid, a tryptophan metabolite. Arch. Biochem. Biophys. 340, 117–123.
Shemer, Y., Kol, R., Sarov, I., 1987. Tryptophan reversal of recombinant human gamma-interferon inhibition of *C. Trachomatis* growth. Curr. Microbiol. 16, 9–13.
Simon, R.P., Swan, J.H., Griffiths, T., Meldrum, B.S., 1984. Blockade of *N*-methyl-D-aspartate receptors may protect against ischemic damage in the brain. Science 226, 850–852.
Sing, L., Oles, R.J., Tricklebank, M.D., 1990. Modulation of seizure susceptibility in the mouse by the strychnine-insensitive glycine recognition site of the NMDA receptor/ion channel complex. Br. J. Pharmacol. 99, 285–288.
Small, D.L., Buchan, A.M., 1997. NMDA antagonists: their role in neuroprotection. In: Green, A.R., Cross, A.J. (Eds.), Neuroprotective Agents and Cerebral Ischaemia. Academic Press, San Diego, pp. 137–148.
Speciale, C., Hares, K., Schwarcz, R., Brooks, A., 1989. High affinity uptake of L-kynurenine by a Na+-independent transporter of neutral amino acids in astrocytes. J. Neurosci. 9, 2066–2072.
Speciale, C., Wu, H.Q., Cini, M., Marconi, M., Varasi, M., Schwarcz, R., 1996. (*R,S*)-3,4-dichlorobenzoylalanine (FCE 28833A) causes a large and persistent increase in brain kynurenic acid levels in rats. Eur. J. Pharmacol., pp. 263–267.
Stevens, J.L., 1985. Isolation and characterization of a rat liver enzyme with both cysteine conjugate beta-lyase and kynureninase activity. J. Biol. Chem. 260, 7945–7950.
Stevens, J.L., Ayoubi, N., Robbins, J.D., 1988. The role of mitochondrial matrix enzymes in the metabolism and toxicity of cysteine conjugates. J. Biol. Chem. 263, 3395–3399.
Stone, T.W., 1993. Neuropharmacology of quinolinic and kynurenic acids. Pharmacol. Rev. 45, 309–379.
Stone, T.W., Perkins, M.N., 1981. Quinolinic acid: a potent, endogenous excitant at amino acid receptors in CNS. Eur. J. Pharmacol. 72, 411–412.
Stone, T.W., Connick, J., 1985. Quinolinic acid and other kynurenines in the central nervous system. Neuroscience 15, 507–617.
Swartz, K.J., During, M.J., Freese, A., Beal, M.F., 1990. Cerebral synthesis and release of kynurenic acid: an endogenous antagonist of excitatory amino acid receptors. J. Neurosci. 10, 2965–2973.
Takikawa, O., Habara-Ohkubo, A., Yoshida, R., 1991. Induction of indoleamine 2,3-dioxygenase in tumor cells transplanted into allogenic mouse: interferon-gamma is the inducer. In: Schwarcz, R., Young, S.N., Brown, R.R. (Eds.), Kynurenine and Serotonin Pathways. Plenum, New York, pp. 437–444.
Taylor, M.W., Feng, G., 1991. Relationship between interferon gamma, indoleamine 2,3-dioxygenase, and tryptophan catabolism. FASEB J. 5, 2516–2522.
Thomas, S.R., Mohr, D., Stocker, R., 1994. Nitric oxide inhibits indoleamine 2,3-dioxygenase activity in interferon gamma primed mononuclear phagocytes. J. Biol. Chem. 269, 14457–14464.
Toma, S., Nakamura, M., Tone, S., Okuno, E., Kido, R., Breton, J., Avanzi, N., Cozzi, L., Speciale, C., Mostardini, M., Gatti, S., Benatti, L., 1997. Cloning and recombinant expression of rat and human kynureninase. FEBS Lett. 408, 5–10.
Tone, S., Takikawa, O., Habara-Ohkubo, A., Kadoya, A., Yoshida, R., Kido, R., 1990. Primary structure of human indoleamine 2,3-dioxygenase deduced from the nucleotide sequence of its cDNA. Nucleic Acid Research 18, 367.
Uemura, T., Hirai, K., 1998. L-kynurenine 3,monooxygenase from mitochondrial outer membrane of pig liver: purification, some properties and monoclonal antibodies directed to the enzyme. J. Biochem. 123, 253–262.
Varesio, L., Clayton, E., Blasi, R., Ruffman, R., Radzioch, D., 1990. Picolinic acid, a catabolite of tryptophan, as the second signal in the activation of interferon gamma primed macrophages. J. Immunol. 145, 4265–4269.
Vartanian, M.G., Taylor, C.P., 1991. Different stereoselectivity of the enantiomers of HA-966 (3-amino-1-hydroxy-2-pyrrolidinone) for neuroprotective and anticonvulsant actions in vivo. Neurosci. Lett. 133, 109–112.
Vécsei, L., Miller, J., MacGarvey, U., Beal, M.F., 1991. Kynurenine and probenecid inhibit pentylenetetrazol and NMDA-induced seizures and increase kynurenic acid concentrations in the brain. Brain Res. Bull. 28, 233–238.
Vederas, J.C., Schleicher, E., Tsai, M., Floss, H.G., 1978. Stereochemistry and mechanism of reactions catalyzed by tryptophanase from *Escherichia coli*. J. Biol. Chem. 253, 5350–5354.
Watanabe, Y., Yoshida, R., Sono, M., Hayaishi, O., 1981. Immunohisto-

chemical localization of indoleamine 2,3-dioxygenase in the argyrophilic cells of rabbit duodenum and thyroid gland. J. Histochem. Cytochem. 29, 623–632.

Whetsell, W.O., Schwarcz, R., 1989. Prolonged exposure to submicromolar concentrations of quinolinic acid causes excitotoxic damage in organotypic cultures of rat cortico-striatal system. Neurosci. Lett. 97, 271–275.

Wolf, H., 1974. Studies on tryptophan metabolism in man. Scand. J. Clin. Lab. Invest. 136S, 1–186.

Wolfensberger, M., Amsler, U., Cuenod, M., Foster, A.C., Whetsell, W.O., Schwarcz, R., 1984. Identification of quinolinic acid in rat and human brain tissue. Neurosci. Lett. 41, 247–252.

Yoshida, R., Hayaishi, O., 1987. Indoleamine 2,3-dioxygenase. Meth. Enzymol. 142, 188–195.

Yoshida, R., Urade, Y., Tokuda, M., Hayaishi, O., 1979. Induction of indoleamino 2,3-dioxygenase in mouse lung during virus infection. Proc. Natl. Acad. Sci. USA 76, 4084–4086.

Yoshida, R., Oku, T., Imanishi, J., Kishida, T., Hayaishi, O., 1986. Interferon: a mediator of indoleamine 2,3-dioxygenase induction by lipopolysaccharide, Poly(I)–Poly(C) and pokeweed mitogen in mouse lung. Arch. Biochem. Biophys. 249, 596–604.

Young, S.N., 1981. Mechanism of decline in rat brain 5-HT after induction of liver tryptophan pyrrolase by hydrocortisone: roles of tryptophan catabolism and kynurenine synthesis. Br. J. Pharmacol. 74, 695–700.

Yuan, W., Collado-Hidalgo, A., Yufit, T., Taylor, M.W., Varga, J., 1998. Modulation of cellular tryptophan metabolism in human fibroblasts by transforming growth factor beta: selective inhibition of indoleamine 2,3-dioxygenase and tryptophanyl-tRNA synthetase gene expression. J. Cell. Physiol. 177, 174–186.