Hearing Research, 40 (1989) 65–74
Elsevier

HRR 01221

The effects of kainic acid on the cochlear ganglion of the rat

Jose M. Juiz [1,2], Joaquin Rueda [1], Jaime A. Merchán [1] and Maria Luisa Sala [1]

[1] *Department of Histology, University of Alicante, Alicante, Spain and* [2] *Kresge Hearing Research Institute, The University of Michigan, Ann Arbor, Michigan, U.S.A.*

(Received 3 August 1988; accepted 24 January 1989)

The effects of locally applied kainic acid on cells and fibers in the rat cochlea were examined in a quantitative and ultrastructural study. Doses of 5 nM per μl of artificial perilymph destroyed part of the spiral ganglion type I cell population, with no ototoxic effects on cochlear hair cells or supporting cells. Type II cells also appeared unaffected. A quantitative evaluation of the cell loss with the 5 nM dosage showed that 34% of spiral ganglion neurons were lost 10 days after treatment. Doses of 20 nM per μl and 40 nM per μl did not result in increasing neuronal loss.

This differential toxicity could reflect the presence of a sub-population of spiral ganglion cells with an increased number of KA receptors.

Kainic acid; Cochlear ganglion; Rat; Neurotoxin; Spiral ganglion neuron

Introduction

Kainic acid (KA) is a conformational restricted analogue of glutamate with well known depolarizing and neurotoxic effects, particularly on neurons receiving rich glutamatergic innervation (Coyle, 1983). In the central nervous system KA has been shown to destroy neurons in a number of areas, including the striatum (Biziére and Coyle, 1978), cerebellum (Herndon and Coyle, 1978), retina (Yazulla and Kleinschmidt, 1980), olfactory cortex (Olney and Gubareff, 1978) and cochlear nucleus (Bird et al., 1978). This has led to a wide use of KA as a tool for investigating neuronal circuitry (Moore, 1981).

The excitatory and toxic properties of KA seem to depend on specific KA receptors located on the neuronal receptive fields (Coyle, 1983; Bird and Gulley, 1979). In any case, KA receptors constitute a distinct pharmacological type of excitatory aminoacid receptor, whose physiological role is still unresolved (for review see Foster and Fagg, 1984).

L-glutamate is a strong candidate for the role of neurotransmitter in the first auditory synapse (inner hair cell-spiral ganglion type I cell dendrite) (Godfrey et al., 1976; Bobbin, 1979; Comis and Leng, 1979; Bobbin et al., 1985; Eybalin and Pujol, 1983; Ryan and Schwartz, 1984; Wiet et al., 1986; Klinke, 1986; Bledsoe et al., 1988). As glutamate-ceptive neurons seem to be particularly prone to the effects of KA (Coyle, 1983), a certain sensitivity to this toxic might be expected on the spiral ganglion (SG) neuronal population (Bledsoe et al., 1981). Pujol et al. (1985) have reported the selective swelling of SG type I cell dendrites (connected with inner hair cells) shortly after the intracochlear infussion of KA. Basically, this is in accordance with previous morphological reports on the initial cytotoxic effects of kainate in the nervous system (Olney and Gubareff, 1978; Rieke and Bowers, 1982; Sperk et al., 1983).

In the present study we have carried out quantitative and ultrastructural studies on the long-term effect of KA on the cochlea of the rat, focusing on a quantitative analysis of SG cells.

Correspondence to: Dr. Joaquin Rueda, Departamento de Histología, Facultad de Medicina, Universidad de Alicante, 03690 Alicante, Spain.

Material and Methods

Sixty four six-week old healthy Wistar albino rats, with normal pinna reflex, were used. After Ekithesin (Green, 1982) anaesthesia, the right tympanic bullae was exposed, and the lateral bony wall visualized under a dissecting microscope. 'Inlet' and 'outlet' holes were drilled in the basal and apical regions, respectively. These holes allowed for the replacement of the natural perilymph, by means of a micropippete attached to a Hamilton syringe.

In 36 of the animals, the perilymph was replaced by 5 μl of Konishi's artificial perilymph (Konishi and Hamrick, 1978) containing 5 nM of KA per μl. The other 28 rats received 5 μl of kainate-free perilymph. The pH and osmolarity of the solutions were adjusted to physiological ranges (pH 7.3–7.4, 310–330 mOsm/l). After slowly replacing the natural perilymph, the holes were closed with a non-toxic material (Peripac R, De Trey, Ash. N.Y., U.S.A.), and the surgical wound sutured.

From the 36 rats receiving the KA solution, 20 were distributed in five groups and sacrificed 24 h, 10 days, 20 days, 30 days and nine months after the exposure to the neurotoxin. The right cochleas were removed, fixed in 10% buffered formaldehyde and embedded in methacrylate for quantitative studies of the SG neuronal population. The remaining 16 rats were distributed in four groups that were sacrificed 24 h, 10 days, 20 days and 30 days respectively after the infusion of the neurotoxin. The right cochleas were quickly removed and fixed by perilymphatic perfusion with 3% *p*-formaldehyde and 2.5% glutaraldehyde in 0.1 M cacodylate buffer, postfixed in 2% osmium tetroxide, dehydrated in graded acetones and embedded in Epon- Araldite for transmission electron microscope. The left, non- manipulated, cochleas were processed under the same conditions and used as controls.

From the 28 rats receiving kainate-free artificial perilymph, and used as controls, 12 were distributed in four groups of 3 rats each, sacrificed respectively 24 h, 10 days, 20 days and 30 days later. The right cochleas were processed for light microscopy as described above, in order to perform SG quantitation. The remaining 16 rats were distributed in four sets, comprising 4 rats each one, sacrificed 24 h, 10 days, 20 days and 30 days after the fluid replacement. The right (manipulated) and left (non-manipulated) cochleas were processed for electron microscopy as previously described.

In addition, two groups of 5 animals each-one, which received artificial perilymph containing 20 nM and 40 nM of KA per μl respectively, were killed 30 days after the KA exposure.

Quantitative studies were performed in 5 μm thick serial sections, stained with hematoxiline-eosine, using a method described elsewhere (Rueda et al., 1987).

Results

Quantitative results

Results of quantitative analysis of SG neurons are shown in Fig. 1 and Table I. Twenty four hours after the cochlear infusion of artificial perilymph, the average number of neurons in the SG is 17945 $\pm$ 819.5. Cell-counts 10, 20 and 30 days later show very similar results (Table I). These numbers are within the range of the normal population of the Wistar rat SG (Rueda et al., 1987).

Twenty four hours after the exposure to the KA solution, the mean SG neuronal population

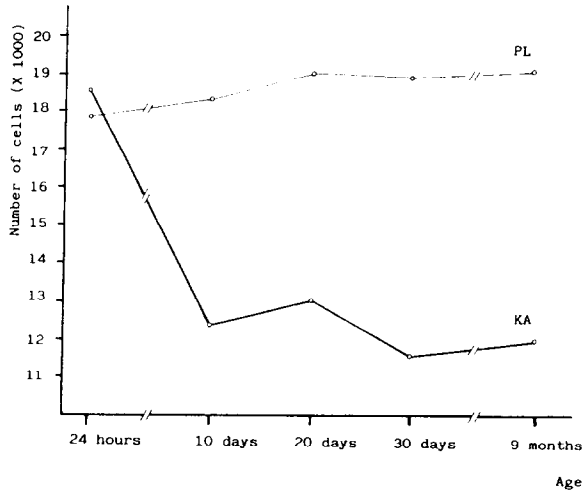

Fig. 1. Neuronal death in the spiral ganglion after kainic acid exposure. There is an evident neuronal loss in animals exposed to kainic acid, that takes place between 24 h and 10 days after the exposure. Control animals show no neuronal loss.

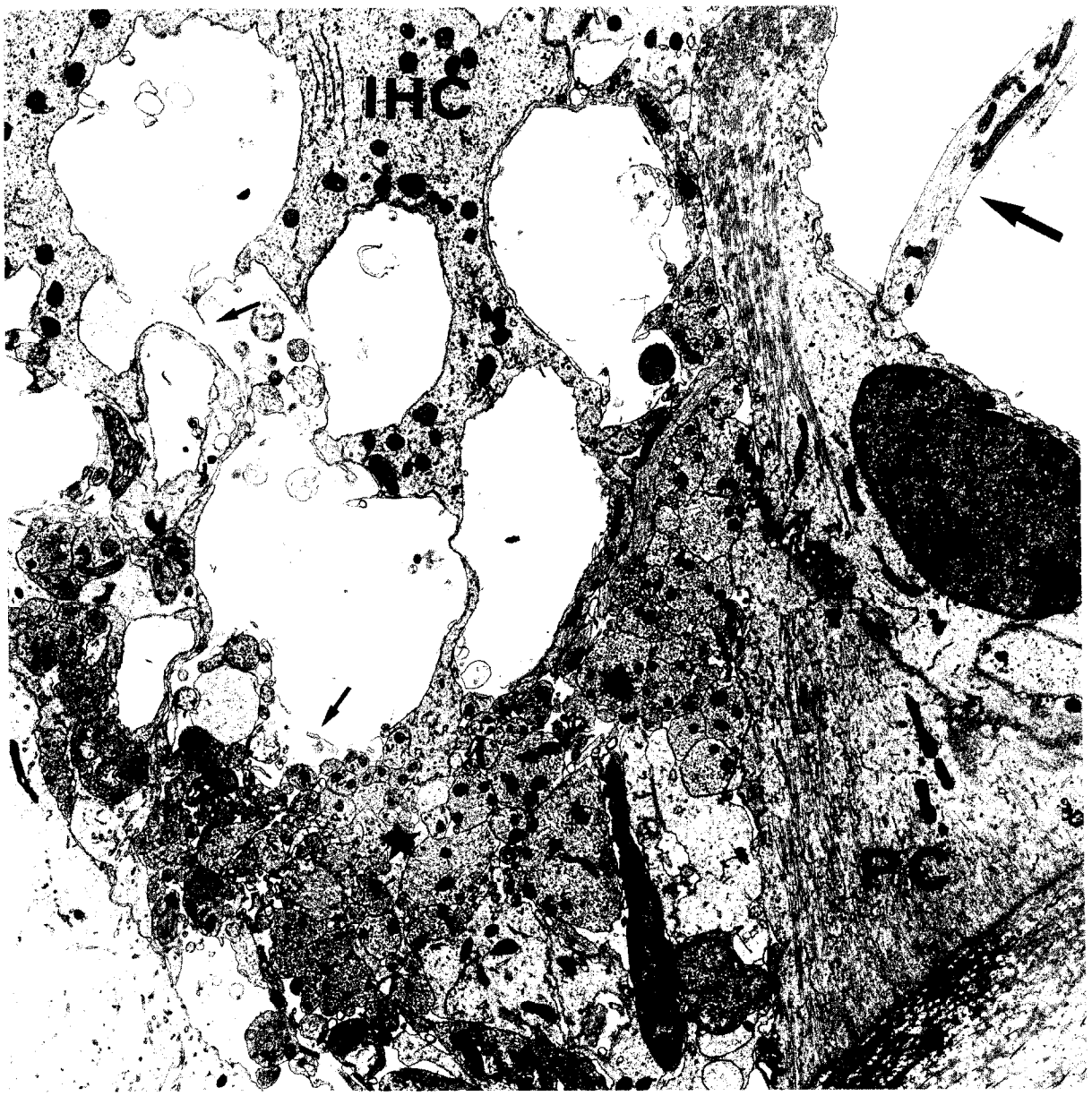

Fig. 2. Inner region 24 h after kainic acid exposure. Swollen inner radial fibers occupy most of the inner spiral bundle area. Among them, spiral fibers (stars) show no evidence of alteration. The large arrow points to a normal tunnel fiber. Small arrows are to show membrane disruptions of the swollen dendrites. IHC (Inner hair cell), PC (Pillar cell). ×8700.

(18635 ± 516.7) resembles that one of the control group (Table I). However, ten days later there is a loss in the number of SG neurons (12464 ± 438) (Table I, Fig. 1). This neuronal loss, which accounts for the 34% of the ganglion cell population is well established 10 days after the exposure to KA, and is not modified further (Fig. 1).

The average number of neurons in the animals receiving 20 nM and 40 nM of KA per μl and killed 30 days later is 13395 ± 715.3 and 12406 ±

TABLE I

CELL COUNTS IN THE SPIRAL GANGLION AFTER KAINIC ACID AND PERILYMPH EXPOSURE

Exposure time	Artificial perilymph	Kainic acid
24 hours	17945 ± 819.5	18635 ± 516.7
10 days	18848 ± 638.6	12464 ± 438
20 days	19141 ± 468.4	13046 ± 1362
30 days	18994 ± 483.2	11670 ± 609.5
9 months		12006 ± 646.6

A significant neuronal loss is evident 10 days after kainic acid exposure. $P < 0.001$ between 24 hours and 10 days exposure-time.

518.1, respectively, which is similar to that obtained with the infussion of perilymph containing 5 nM of KA per μl. ($P < 0.001$ between animals exposed 30 days to perilymph containing KA 5 nM, 20 nM and 40 nM, respectively).

Ultrastructural results

Twenty four hours after KA placement in the cochlea, inner radial fibers are swollen and almost empty of cytoplasmic content (Fig. 2). It is worth noting that some of these fibers are disrupted (Fig. 2 small arrows). Among these altered inner radial fibers, elements running in the inner spiral bundle, i.e. the fibers of the efferent system and the afferents to outer hair cells, appear to be normal (Fig. 2). The same is true for the remaining neuronal elements, including afferent and efferents crossing the tunnel, as well as their endings below outer hair cells (Fig. 3). Receptor and supporting cells are completely normal (Figs. 2, 3). Ten and 20 days after the intracochlear placement of KA, ultrastructural examination reveals the presence of some swollen inner radial fibers, some of them also disrupted. However, an important finding is that almost (or completely) normal inner radial fibers are not uncommon. This fact is particularly evident 30 days after the exposure to KA. At this time no swellings are evident, and only cell debris (probably degenerated afferents) can be seen below inner hair cells, intermingled with normal nerve fibers (Fig. 5). In addition, the remaining nerve elements in the acoustic receptor are completely normal in these groups, as it occurred in the 24 h kainate-exposed animals. The same is true for the receptor and supporting structures.

Fig. 3. Basal pole of an outer hair cell (OHC) 24 h after kainic acid exposure. Both afferent (A) and efferent (E) endings appear normal. ×17000.

Fig. 4. Spiral ganglion, 24 h after kainic acid exposure. Type I cells exhibit large vacuolizations (asterisks). Swollen mitochondria are also evident. ×3500.

Fig. 5. Inner region 30 days after KA injection. Membrane debris (arrows) can be seen. There are no swollen inner radial fibers. The stars indicate a normal one approaching to the basal pole of an inner hair cell (IHC). PC (Pillar cell). ×14000.

In the SG, some of the type I cells show some pathology, cytoplasmic vacuoles (Fig. 4) being the most prominent feature. Swollen or collapsed mitochondria and dispersion of organelles are less evident findings. Swollen axoplasms are also present. However, an abundant number of type I cells exhibit a normal appearance, as well as all the type II cells. In the SG, 10, 20 and 30 days after the exposure to the neurotoxic, there are type I cells displaying signs of severe damage. These include collapsed protoplasmic masses intermingled with membrane debris and enclosed by

Schwann cells (Fig. 6). Myelin figures, large vacuoles and lysosomes may crowd the cytoplasm, which is often reduced to amorphous masses. Besides this, type I cells with no evidence of alterations are frequently seen . Type II cells appear completely normal (Fig. 6).

In the control groups, 24 h after the kainate-free perilymph placement in the cochlea, swollen inner radial fibers are evident. However, as opposed to what was seen in the 24 hour KA-exposed animals, no disrupted fibers are obvious (Fig. 7). Other nerve elements (efferent fibers and afferents to outer hair cells) are completely normal. No signs of damage are found in the ganglion cell bodies in the SG. Ten and twenty days later, the area below inner hair cells still shows some swollen radial fibers. However, 30 days later, the ultrastructure of this region changes, and no membrane debris are found underneath inner hair cells. In fact, the morphological aspect of this area is completely normal (Fig. 8). The same is true for the remaining cochlear fibers, as well as the cell bodies in the SG.

Fig. 6. Spiral ganglion, 20 days after intracochlear kainic acid infusion. There are some collapsed type I cell somatas (I). Membrane debris are evident (arrowheads). The arrow points to a degenerating axonal element. (II) is a normal type II cell. ×3000.

Fig. 7. Inner region 24 h after perilymph exposure. There are swollen inner radial fibers (stars). Some of them have mitochondria and a flocculent material. IHC (Inner hair cell), PC (Pillar cell). ×4700.

Discussion

The present results show that KA exerts a toxic effect on part of the rat spiral ganglion cell population. The intracochlear injection of KA at a concentration of 5 nM per μl (5 μl in total), causes a SG cell loss that accounts for a 34% of the normal neuronal population. This is a high concentration of KA (Moore, 1981) which suggests that the unaffected neurons (66%) may be relatively resistant to the neurotoxin. Supporting this, we have found that higher concentrations of the neurotoxin (20 nM per μl and 40 nM per μl) do not modify the 34% neuronal loss.

An important question arises as to which neuronal population is affected by KA. Dendritic swelling is the first morphological evidence of KA toxicity (Herndon and Coyle, 1978; Nadler et al.,

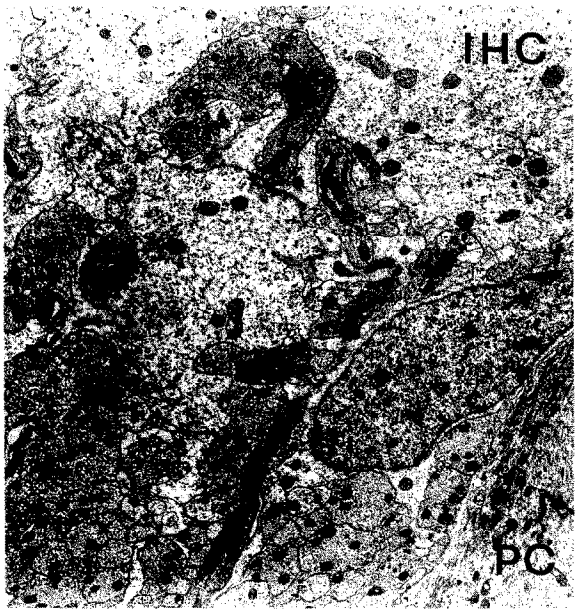

Fig. 8. Inner region 30 days after Ka-free perilymph exposure. Swelling is not present, and the inner spiral bundle area is completely normal. There are no membrane debris scattered among the nerve fibers. IHC (Inner hair cell), PC (Pillar cell). ×4700.

1981; Rieke and Bowers, 1982; Coyle, 1983; Sperk et al., 1983; Pujol et al., 1985). Our ultrastructural results show a prominent swelling of inner radial fibers in the first 24 h after the infusion of KA (Fig. 2), while both the afferents to outer hair cells are normal (Fig. 3) and the fibers of the efferent system (Figs. 2, 3) look normal; no alterations are evident in the acoustic receptor itself. All of these results are in good accordance with the proposal that the cochlear toxicity of KA is restricted to the SG type I cell population (Pujol et al., 1985).

However, this initial type I cell dendritic swelling is massive (Fig. 2), as if all of these neurons were affected by the neurotoxin. Once an excitotoxic process begins, many studies have shown it to be essentially non reversible, leading to neuron degeneration and death (Coyle, 1983; Mayer and Westbrook, 1987; Rothman and Olney, 1988). However, our quantitative results show only a 34% neuronal loss. An explanation for this discrepancy could be that not all the dendritic swelling is related to the KA toxicity, as otherwise all of the type I neurons should be expected to degenerate and die, and not just a part of them. Actually, dendritic swelling is not restricted to KA toxicity. In the case of the cochlea, and regardless the mechanism, it is known that inner radial fibers swell after a variety of injuries, such as acoustic trauma (Beagley, 1965; Spoendlin, 1968, 1971, 1975), postmorten anoxia (Nadol and Burgess, 1985) or the effects of some ototoxic drugs (Terayama et al., 1977). Some surgical manipulations of the cochlea (Kelly and Khana, 1984; Juiz et al., 1988) can also lead to swelling of inner radial fibers, that seems to be reversible in some cases (Juiz et al., 1988; Rueda et al., 1989).

Swollen inner radial fibers were present in the control animals of the present study, mainly in the 24 h group (Fig. 7). As the left, non-manipulated, cochleas of these animals are normal, the origin of this alteration could be related to the cochlear manipulation, as discussed elsewhere (Juiz et al., 1988). Thus, the massive dendritic swelling could be atributed not only to the neurotoxin but also to conditions related to the surgical procedure.

The 34% of the SG neurons destroyed by KA would also display an initial swelling directly related to the excitoxin, but this could not be distinguished from the non-specific swelling that, on the other hand, seems to be reversible with time (Juiz et al., 1988, Rueda et al., 1989). In fact, until a method is found to differentiate between KA-induced swelling and the reversible swelling, we cannot determine whether KA only causes initial swelling in afferent fibers of the subpopulation of SG neurons (34%) that will disappear, or if it affects the reversibility of the non-specific lesion. A further possibility is an atypical behaviour of SG cells, so that KA-induced swelling occurs in all afferent fibers, but in two-thirds of them it is reversible.

In any case, with or without the contribution of other sources of injury, KA seems to selectively destroy part of the type I SG neuronal population, and this is supported from the findings in the remaining groups of animals. The cell debris (probably degenerated inner radial fibers) below inner hair cells 10, 20 and 30 days after KA treatment (Fig. 5) together with some severely degenerated type I cell somatas (Fig. 6) and intact type II cells (Fig. 6) indicates the selectivity of the neuronal loss for the type I cell population. That

there are also type I cells relatively resistant to the neurotoxin is shown by the normal somatas and peripheral endings (Fig. 5), which also indicated that 30 days after the treatment the initial massive swelling had already dissapeared. In fact, control animals show no cochlear alterations 30 days after manipulation (Fig. 8). The swollen fibers still present 10 and 20 days after the KA treatment are, most probably, non-specific in origin, as the neuronal loss is already established at this time, and the swellings also appear in control animals.

Previous studies have shown how KA, in the micromolar range, selectively abolishes the cochlear nerve action potential (CAP) (Bledsoe et al., 1981; Kusakari et al., 1984). This would imply that all the cochlear neurons are affected by the neurotoxin, and not just a part (i.e. 34%) of it. This discrepancy deserves further studies, but some hypothesis may be advanced. First of all, species differences cannot be ruled out, previous studies mentioned here were carried out on guinea pigs. Also, the supression of the CAP does not necessarily reflect a toxic process, it could be the result of excitatory action on primary afferents (Bledsoe et al., 1981; Jenison and Bobbin, 1985). Excitation is only one of the possible steps in the poorly known cascade of events leading to the kainate-induced neuronal degeneration (Coyle, 1983; Mayer and Westbrook, 1987; Rothman and Olney, 1988). In fact, and under certain conditions, it is possible to supress KA toxicity while maintaining its excitatory effects (Mc Lennan, 1980). Another factor to consider is the possibility that cochlear neurons are homogeneously sensitive to the excitatory effects of KA (Bledsoe et al., 1981; Kusakari et al., 1984), but not to the toxic ones. This might reflect a differential density of KA receptors (Foster and Fagg, 1984), with the highest concentration on the 34% of auditory neurons which are sensitive to the neurotoxin. Another possibility could be the presence of a 'receptor set' including both pre- and postsynaptic receptors (Ferkany et al., 1982; Coyle, 1983) only on the auditory synapses sensitive to the neurotoxin. The partial toxicity could also be a reflection of the lack of *N*-methyl-D aspartate receptors of the cochlea (Fex and Martin, 1980; Jenison et al., 1986), as they seem to be involved, to some exent, in the KA toxicity (Bledsoe, personal communication; Rothmann and Olney, 1988). In any case, further studies are needed to clarify this point.

In conclusion, KA destroys 34% of the rat SG neurons, most probably type I. This finding supports not only the existence of a difference between type I and type II SG neurons (Pujol et al., 1985), but also the presence of some degree of heterogeneity in the type I cell population itself: the differential sensitivity to the neurotoxin might reflect different densities of KA receptors among type I neurons. As it has been pointed out by Liberman (1980), neurons innervating inner hair cells are not morphologicaly homogeneous. Although the heteregeneous behaviour of these neurons to KA also support this point of view, its significance remains as an open question, as the physiological role of KA receptors regarding synaptic transmission is still controversial (Foster and Fagg, 1984; Stone and Burton, 1988). To what extent these results can be extrapolated to other species, must be also addressed in the future.

Acknowledgements

The authors are indebted to Drs. R.A. Altschuler, S. C. Bledsoe Jr. and J.J. Prieto for their valuable comments and suggestions during the writing of the manuscript. Our acknowledgements to Drs. D.J. Lim and M.A. Ruggero for the critical reading of a previous version of this paper, and to Mrs. Dolores Segura for its expert technical assistance. Mrs. Mercedes Garcia Encinas provided the illustration.

This work was partially supported by grants from the Spanish Government (CICYT PB86-0277, FISS 1646/87).

References

Beagley, H.A. (1965) Acoustic trauma in the guinea pig. Acta Otolaryngol. (Stockh.) 60, 479–495.

Bird, S.J., Gulley, R.L., Wenthold, R.J. and Fex, J. (1978) Kainic acid injections results in degeneration of cochlear nucleus cells innervated by the auditory nerve. Science 202, 1087–1089.

Bird, S.J. and Gulley, R.L. (1979) Evidence against a pre-synaptic mechanism for kainate neurotoxicity in the cochlear nucleus. Neurosci. Lett. 15, 55–60.

Biziére, K. and Coyle, J.T. (1978) Influence of cortico-striatal afferent on striatal kainic acid neurotoxicity. Neurosci. Lett. 8, 303–310.

Bledsoe, S.C., Bobbin, R.P. and Chihal, D.M. (1981) Kainic acid: an evaluation of its action on cochlear potentials. Hear. Res. 4, 109–120.

Bledsoe, S.C., Bobbin, R.P. and Puel, J.L. (1988) Neurotransmission in the inner ear. In: A.F. Jahn and J.R. Santos-Sacchi (Eds.), Physiology of the ear. Raven Press. N.Y. pp. 385–406.

Bobbin, R.P. (1979) Glutamate and aspartate mimic the afferent transmitter in the cochlea. Exp. Brain Res. 34, 389–393.

Bobbin, R.P., Bledsoe, S.C. and Jenison, G.L. (1985) Neurotransmitters of the cochlea and the lateral line organ. In: C.I. Berlin (Ed.), Hearing Science, College Hill Press, pp. 159–180

Comis, S.D. and Leng, G. (1979) Action of putative neurotransmitters in the guinea pig cochlea. Exp. Brain Res. 36, 119–128.

Coyle, J.T. (1983) Neurotoxic action of kaininc acid. J. Neurochem. 41, 1–11.

Eybalin, M. and Pujol, R. (1983) A radioautographic study of H3L-glutamate and H3L-glutamine uptake in the guinea pig cochlea. Neurosci. 9, 863–871.

Ferkany, J.W., Zaczek, R. and Coyle, J.T. (1982) Kainic acid stimulates excitatory aminoacid neurotransmitter release at presynaptic receptors. Nature 298, 757–759.

Fex, J. and Martin, M.R. (1980) Lack of effect of DL-α-aminoadipate, an excitatory amino acid antagonist, on cat auditory nerve responses to sound. Neuropharmacology 19, 809–811.

Foster, A.C. and Fagg, G.E. (1984) Acidic aminoacid binding sites in mammalian neuronal membranes: their characteristics and relationships to synaptic receptors. Brain Res. Review 7, 103–164.

Godfrey, D.A., Carter, J.A., Berger, S.I. and Matschinsky, F.M. (1976) Levels of putative transmitter aminoacids in the guinea pig cochlea. J. Histochem. Cytochem. 24, 468–470.

Green, C.J. (1982) Animal Anaesthesia. Laboratory Animal Handbooks 8, pp. 131–163. Laboratory Animals Ltd.

Herndon, R.M. and Coyle, J.T. (1978) Glutamatergic innervation, Kainic acid and selective vulnerability in the cerebellum. In: E.G. Mc Geer, J.W. Olney and P.L. Mc Geer (Eds.), Kainic acid as a tool in Neurobiology, Raven Press, N.Y. pp. 189–200

Jenison, G.L. and Bobbin, R.P. (1985) Quisqualate excites spiral ganglion neurons of the guinea pig. Hear. Res. 20, 261–265.

Jenison, G.L., Winberg, S. and Bobbin, R.P. (1986) Comparative actions of quisqualate and *N*-methyl-D-aspartate, excitatory amino acid agonists, on guinea-pig cochlear potentials. Comp. Biochem. Physiol. 84C, 385–389.

Juiz, J.M., Rueda, J. and Merchán, J.A. (1988) Reversible damage to the nerve fibres in the organ of Corti after surgical opening of the cochlea in the rat. Acta Otolaryngol. (Stockh.) 106, 29–33.

Kelly, J.P. and Khanna, S.M. (1984) Distribution of cochlear damage caused by the removal of the round window membrane. Hear. Res. 16, 109–126.

Klinke, R. (1986) Neurotransmission in the inner ear. Hear. Res. 22, 235–243.

Konishi, T. and Hamrick, P.E. (1978) Ion transport in the cochlea of guinea pig. I: Potassium and sodium transport. Acta Otolaryngol (Stockh.) 86, 22–34.

Kusakari, J., Arakawa, E., Ohyama, K, Rokugo, M. and Inamura, M. (1984) Effect of kainic acid upon N_1 latency. Laryngoscope 94, 1365–1369.

Liberman, M.C. (1980) Morphological differences among radial afferent fibers in the cat cochlea: an electron-microscopic study of serial sections. Hear. Res. 3, 45–63.

Mayer, M.L. and Westbrook, G.L. (1987) Cellular mechanisms underlying excitotoxicity. Trends in Neurosci. 10, 59–61.

Mc Lennan, H. (1980) The effect of decortication on excitatory aminoacid sensitivity of striatal neurons. Neurosci. Lett. 18, 313–316.

Moore, R.Y. (1981) Methods for selective, restricted, lesion placement in the central nervous system. In: L. Heimer and M.J. Robbards (Eds.), Neuroanatomical Tract-Tracing Methods. Plenum Press. N.Y. pp. 55–85.

Nadler, J.V., Evenson, D.A. and Cuthberson, G.J. (1981) Comparative toxicity of kainic acid and other acidic aminoacids toward rat hippocampal neurons. Neuroscience 6, 2505–2517.

Nadol, J.B. and Burgess, B. (1985) A study of post-mortem autolysis in the human organ of Corti. J. Comp. Neurol. 237, 333–342.

Olney, J.W. and Gubareff, T. (1978) Extreme sensitivity of olfactory cortical neurons to kainic acid toxicity. In: E.G. Mc Geer, J.W. Olney and P.L. Mc Geer (Eds.), Kainic acid as a tool in Neurobiology, Raven Press, N.Y. pp. 201–217.

Pujol, R., Lenoir, M., Robertson, D., Eybalin, M. and Johnstone, B.M. (1985) Kainic acid selectively alters auditory dendrites connected with inner hair cells. Hear. Res. 18, 145–151.

Rieke, G.K. and Bowers, D.E. (1982) Acute effects of the neurotoxin kainic acid on neurons of the pigeon basal ganglia. Acta Neuropathol. (Berlin) 56, 123–135.

Rothman, S.M. and Olney, J.W. (1988) Excitotoxicity and the NMDA receptors. Trends Neurosci. 10, 299–302.

Rueda, J., Sen, C., Juiz, J.M. and Merchán, J.A. (1987) Neuronal loss in the spiral ganglion of young rats. Acta Otolaryngol. (Stockh.) 104, 417–421.

Rueda, J., Juiz, J.M. and Merchán, J.A. (1989) Surgical trauma to the cochlea results in reversible damage to spiral ganglion type I neurons. Acta Otolaryngol. (Stockh.) 107, 59–62.

Ryan, A.F. and Schwartz, I.R. (1984) Preferential glutamine uptake by cochlear hair cells: implications for the afferent cochlear transmitter. Brain Res. 29, 376–379.

Sperk, G., Lassman, H., Baran., Hish, S.J., Seitelberg, F. and Hornykiewicz, A. (1983) Kainic acid induced seizures: neurochemical and histopathological changes. Neuroscience 10, 1301–1315.

Spoendlin, H. (1968) Ultrastructure and peripheral innervation pattern of the receptor in relation to the first coding of the acoustic message. In: A.V.S. de Reuck and J. Knight (Eds.), Hearing Mechanisms in Vertebrates, Churchill, London, pp. 89–119.

Spoendlin, H. (1971) Degeneration behaviour of the cochlear nerve. Arch. Klin. Exp. Ohren. Nasen. Kehlkopfheild 200, 275–291.

Spoendlin, H. (1975) Retrograde degeneration of the cochlear nerve. Acta Otolaryngol. (Stockh.) 79, 266–275.

Stone, T.W. and Burton, N.R. (1988) NMDA receptors and ligands in the vertebrate CNS. Progress Neurobiol. 30, 333–368.

Terayama, I., Kaneko, Y., Kawamoto, K. and Sakai, N. (1977) Ultrastructural changes of nerve elements following disruption of the organ of Corti. I: Nerve elements in the organ of Corti. Acta Otolaryngol. (Stockh.) 83, 291–302.

Wiet, G.J., Godfrey, D.A., Ross, C.D. and Dunn, J.D. (1986) Quantitative distribution of aspartate aminotransferase and glutaminase activities in the cochlea. Hear. Res. 24, 137–150.

Yazulla, S. and Kleinschmidt, J. (1980) The effects of intraocular injection of kainic acid on the synaptic organization of the goldfish retina. Brain Res. 82, 287–301.