

Invited review in histology and cell biology

Ruggero Bortolami (1926-2014): in memoriam

Ermanno Manni

Professor Emeritus of Human Physiology, Medical School, Catholic University, Rome.

Key words

Central Nervous System, Comparative Anatomy, History of Medicine, Neuroanatomy, Physiology, Cranial Nerves

This short writing is a deep-felt commemoration of a dearest Colleague and very skilled researcher who has worked with me along more than 35 years and who died July 18, 2014.

I met prof. Bortolami, chairman of the Veterinary Anatomical Institute, for the first time in the afternoon of the far October 21, 1962, in Sassari. The chair of Human Physiology of that medical School was vacant and I had applied in order to obtain the appointment as professor in charge for the new academic year. On November 14 the Faculty appointed me as professor in charge of Human Physiology for academic year 1962-63, and by the end of November I filled my new position. In this way I had the opportunity to meet frequently professor Bortolami: thus our friendship began. In the University of Sassari a great fellowship linked the professors to each other. Bortolami had a very solid scientific background: he had carried out important research on the conducting system of the heart in numerous vertebrates under the guide and in collaboration with his chairman professor Valentino Chiodi (Chiodi and Bortolami, 1967). I was impressed by his intelligence and by his scientific preparation obtained also through his stay at the University of Paris. He became interested in the trigeminal nerve, particularly in the degenerations following destruction of some trigeminal fibres in the central nervous system. His results brought relevant contribution to odology. However, he was unsatisfied because by means of histological techniques only it was difficult to arrive at function. Thus the idea of carrying out experiments with a physiologist was born: the collaboration of an anatomist with a physiologist would have been useful and fruitful for both. Our collaboration began in 1964 and continued for about 35 years, even after his return to Bologna (1969) and my transfer to Rome (1973), and involved also many of our young collaborators.

The first investigation we carried out together concerned the analysis of the posterior intertectal commissure in ducks. Bortolami and Veggetti (1965) had observed that the section of some trigeminal branches coming from "masticatory" muscles induced the degeneration of cells contained in the posterior intertectal commissure. We decided to explore the electrical activity of single cells of the layer of posterior intertectal commissure during the movements of the jaw, with a tungsten microelectrode (Manni et al., 1965): 85% of these cells responded with high frequency discharge (260/sec)

to lowering the jaw of the duck, while the remaining 15% cells were influenced by raising the jaw. The conclusion was reached that the cells of the posterior intertectal commissure of birds exhibit the same functions as those of the mesencephalic trigeminal nucleus of mammals. Passatore et al. (1979b) were able to discover a somatotopic arrangement in the posterior intertectal commissure of ducks, corresponding to movements of the jaw. Azzena and Palmieri (1967) showed that the masseteric reflex is monosynaptic also in ducks as in mammals. Azzena et al. (1970) observed that the proprioceptive "masticatory" information reaches the cerebellum. In later years these investigations were extended in our Institutes to other species that had not been studied previously. Desole et al. (1970, 1971) in caiman sclerops and Marini and Bortolami (1982) in the frog confirmed our results.

We then studied the control exerted on the mesencephalic trigeminal nucleus by different nervous structures. In lambs and birds the vagal nerve and the bulbo-pontine reticular formation (Manni et al., 1978a), and in the rabbit the masticatory cerebral cortex (Passatore et al., 1979a; Manni et al., 1980) may induce different effects, however excitation was the prevalent influence.

On the contrary the area postrema inhibits the mesencephalic trigeminal nucleus (Manni et al., 1982). Nevertheless the cell bodies of the primary afferents supplying jaw raising muscles and the periodontal or gingival mechanoreceptors exhibit a somatotopic localization in the rabbit mesencephalic nucleus (Passatore et al., 1983).

A second group of our investigations concerned the peripheral and central organization of the extraocular muscle proprioception in various animals (Manni et al., 1966, 1968). The question was a matter of controversy. Are the perikarya of afferent proprioceptive fibres located in the mesencephalic trigeminal nucleus, or in the nuclei of the III, IV and VI cranial nerves? It was also admitted that the cells could be identified within ganglia scattered along the III, IV and VI nerves. Bortolami and I focused on the semilunar ganglion of lamb, pig and calf, whose eye muscles contain spindles, and were able to recognize for the first time a medial dorsolateral area in the semilunar ganglion of the lamb, pig and calf, that contains a cell pool from which it is very easy to record typical responses to stretch of a given eye muscle, by means of a tungsten microelectrode. Chronic cutting of any of the oculomotor, trochlear and abducent nerves did not abolish the responses (Manni et al., 1966, 1968, 1970b, 1971a). These cells did not respond to other trigeminal stimulations (Manni et al., 1966, 1968). The response of a given ocular muscle was suppressed by cutting the ipsilateral trigeminal ophthalmic branch which provoked also degeneration of the corresponding eye muscle spindles (Manni et al., 1968). The chronic section of the trigeminal ipsilateral pontogasserian tract did not abolish the semilunar responses to stretching the extraocular muscles (Manni et al., 1970a): this fact shows that we did not record the electrical activity of nervous trigeminal fibres in transit within the semilunar ganglion. Manni and Pettorossi (1976) demonstrated a somatotopic arrangement of the cells responding to stretch of eye muscles within the medial dorsolateral pool of the semilunar ganglion of the lamb. Such an organization was confirmed histochemically following injection of horseradish peroxidase into single eye muscles of lamb and pig (Bortolami et al., 1987b).

Further investigation carried out in lambs by Manni et al. (1971a, 1972a, 1972b, 1974) traced the central pathway of eye muscle proprioception in the spinal cord and in the encephalic trunk.

The conclusion was reached that the perikarya of 2° order axons of eye muscle proprioception are located in the pontine main trigeminal nuclei and in the oral portion of the descending spinal trigeminal nuclei and are connected with ipsilateral thalamus through the medial lemniscus and the dorsal trigemino-thalamic tract. Marini and Bortolami (1979, 1980, 1982) were able to demonstrate a somatotopic organization of the afferents from the extraocular muscles in the main trigeminal nucleus, in the oral portion of the descending trigeminal nucleus and in the superior colliculus. In lambs, fibres of the 2° order neurons of the extraocular muscles proprioception project to paravermian areas of the cerebellum (Azzena et al., 1970).

We then addressed the structural basis of eye muscle proprioception in spindle-less animals, as the cat. In this animal responses to stretch of single eye muscles were recorded from both the semilunar ganglion and trigeminal mesencephalic nucleus; they were prevalent in the mesencephalic nucleus, where they exhibited a low threshold and a slow adaptation (Bortolami et al., 1987b)

Furthermore we demonstrated (Manni et al., 1970c) that the ganglion cells scattered along the trunk of the three eye nerves (III,IV,VI) cannot represent the somata of fibres innervating the eye muscle spindles (Tozer, 1912). The experiments were carried out on calves since these animals are the mostly endowed with this type of cells (Palmieri and Asole, 1967). All the calves studied presented typical responses of the cells of medial dorsolateral gasserian pool to stretching a single eye muscle. No response to stretching eye muscles was recorded from the ganglion cells scattered along the oculomotor nerve.

To complete this area of investigation we studied the influence on ocular motility of the destruction of gasserian cells that control the proprioception of the medial or the lateral rectus muscles of lambs and observed an eye deviation towards or away from the damaged side, respectively (Pettorossi et al., 1995). In other investigations the unilateral electrolysis destruction of all gasserian medio-dorso-lateral cells provoked marked alterations, more marked for the trajectories of the quick phase and less so for the slow phase of the horizontal and vertical vestibulo-ocular reflex (Pettorossi et al., 1997).

A third group of investigations dealt with the central course and the functional role of trigeminal fibres present along the trunk of the oculozygic nerves. The results showed that the III, IV and VI cranial nerves must be considered as mixed nerves since they contain not only motor fibres but also a minority of afferent axons that have the perikarya in the semilunar ganglion of the trigeminal nerve.

We were able to trace the course of these axons through the encephalic trunk and their termination in the two first cervical segments of spinal cord (Bortolami et al., 1977; Manni et al., 1976, 1978b, 1979, 1984, 1987, 1989). The terminations of the aberrant trigeminal fibres in the substantia gelatinosa are axo-axonal on classical trigeminal axons just before the latter in turn terminate, *i.e.* they are presynaptic fibres. It is well known that such presynaptic fibres may be inhibitory and the aberrant fibres might exert this effect on classical trigeminal fibres. Bortolami et al. (1987a) pointed out that there is an analogy between the trigeminal afferent fibres running within the oculomotor nerve and the afferent fibres contained within the ventral roots of spinal cord. We proposed an explanation for the physiological meaning of such aberrant trigeminal axons: they might transmit painful signals, since they are of small diameter and may be enclosed in groups A delta and C, which conduct nerve impulses at

low speed. According to our experiments such axons have a role in transporting thermal painful stimuli from the eyelid areas: in fact they supply the extraocular muscles, cornea and superior eyelid and contain neuropeptides which are usually associated with pain sensation (Bortolami et al., 1991). These axons are influenced by bradykinin and histamine that may reduce nociception, and by capsaicin capable of increasing nociception (Manni et al., 1989). The presence of aberrant trigeminal fibres in the oculomotor nerve may explain the reappearance of pain after retrogasserian rhizotomy in patients suffering for trigeminal neuralgia (Manni et al., 1979).

Our results have been accepted and quoted by the international literature. We have been invited to deliver lectures on our results in national and international Congress and meetings: twice at Wenner Gren Center of Stockholm, in 1974 (Manni, 1975) and 1981 (Manni and Bortolami, 1982); twice in Pisa, in 1978 (Manni and Bortolami, 1979) and 1981 (Bortolami and Manni, 1981); and in Schloss Reisingen (Ulm) 1980 (Manni et al., 1981).

Prof. Bortolami has been a precise, deep and passionate researcher and with his scientific contributions he must be considered one of the most illustrious Italian anatomist. His death has been a great loss not only for his Family, but also for the Italian Science and for us who have known and appreciated him.

Acknowledgements

The author is indebted to professor A. M. Barazzoni Bortolami for revising the English text.

The publication of this paper was supported by the Italian Association of Anatomists and Histologists, in honour to its distinguished member Prof. Ruggero Bortolami.

References

- Azzena G.B., Desole C., Palmieri G. (1970) Cerebellar projection of the masticatory and extraocular muscle proprioception. *Exp. Neurol.* 27: 151-161.
- Azzena G.B., Palmieri G. (1967) A trigeminal monosynaptic reflex in birds. *Exp. Neurol.* 18: 184-193.
- Bortolami R., Manni E. (1981) Aspetti morfofunzionali del nucleo mesencefalico del trigemino e del ganglio di Gasser. *Atti della Società italiana di Anatomia, XXXVII Convegno nazionale*: 63-121.
- Bortolami R., Veggetti A. (1965) Etude expérimental sur le noyau mésencéphalique du trijumeau dans les Oiseaux. *Compt. Rend. Assoc. Anat.* 124: 307-310.
- Bortolami R., Veggetti A., Palmieri G. (1970) Ulteriori osservazioni sui rapporti tra branca oftalmica ed oculomotore comune. *Atti Soc. Ital. Sci. Vet.* 24: 189-195.
- Bortolami R., Veggetti A., Callegari E., Lucchi M.L., Palmieri G. (1977) Afferent fibres and sensory ganglion cells within the oculomotor nerve in some mammals and man - I. Anatomical investigations. *Arch. Ital. Biol.* 115: 355-385.
- Bortolami R., Lucchi M.L., Callegari E., Calzà C., Pettorossi V.E., Manni E. (1987a) Analogies existing between the primary trigeminal afferent fibres running within the oculomotor nerve and ventral root primary afferent fibres. In: Tiengo M.,

- Eccles J., Cuello A.C., Ottoson D. (Eds.) Pain and Mobility. Raven Press, New York. *Advances in Pain Research and Therapy* 10: 59-63.
- Bortolami R., Lucchi M.L., Pettorossi V.E., Callegari E., Manni E. (1987b) Localization and somatotopy of sensory cells innervating the extraocular muscles of lamb, pig and cat. Histochemical and electrophysiological investigation. *Arch. Ital. Biol.* 125: 1-15.
- Bortolami R., Calzà C., Lucchi M.L., Giardino L., Callegari E., Manni E., Pettorossi V.E., Barazzoni A.M., Lalatta Costerbosa G. (1991) Peripheral territory and neuropeptides of the trigeminal ganglion neurons centrally projecting through the oculomotor nerve demonstrated by fluorescent retrograde double-labeling combined with immunocytochemistry. *Brain Res.* 547: 82-88.
- Chiodi V., Bortolami R. (1967) The conducting system of the vertebrate heart. Edizioni Calderini, Bologna.
- Desole C., Palmieri G., Veggetti A. (1970) Mesencephalic trigeminal nucleus and jaw muscle proprioception in reptilians. *Arch. Ital. Biol.* 108: 121-129.
- Desole C., Palmieri G., Veggetti A. (1971) A trigeminal monosynaptic reflex in reptilians. *Exp. Neurol.* 32: 287-294.
- Manni E. (1975) Discussion remarks. In: Lennerstrand G., Bach-y-Rita P., Collins C.C., Jampolsky A., Scott A.B. (Eds.) *Basic Mechanisms of Ocular Motility and their Clinical Implications*. Pergamon Press, Oxford, New York. Pp. 109-110.
- Manni E., Bortolami R. (1979) Peripheral and central organization of the extraocular muscle proprioception in the Ungulata. In: Granit R., Pompeiano O. (Eds.) *Reflex Control of Posture and Movement* Elsevier/North Holland Biomedical Press, Amsterdam, New York. *Progr. Brain Res.* 50: 291-299.
- Manni E., Bortolami R. (1982) Proprioception in eye muscles. In: Lennerstrand G., Zee D.S., Keller E.L. (Eds.) *Functional Basis of Ocular Motility Disorders*. Pergamon Press, Oxford, New York. Pp. 55-67.
- Manni E., Pettorossi V.E. (1976) Somatotopic localization of the eye muscle afferents in the semilunar ganglion. *Arch. Ital. Biol.* 114: 178-187.
- Manni E., Bortolami R., Azzena G.B. (1965) Jaw muscle proprioception and the mesencephalic trigeminal cells in birds. *Exp. Neurol.* 12: 320-328.
- Manni E., Bortolami R., Desole C. (1966) Eye muscle proprioception and the semilunar ganglion. *Exp. Neurol.* 16: 226-236.
- Manni E., Bortolami R., Desole C. (1968) Peripheral pathway of the eye muscle proprioception. *Exp. Neurol.* 22: 1-12.
- Manni E., Bortolami R., Deriu P.L. (1970a) Presence of cell bodies of the afferents from the eye muscles in the semilunar ganglion. *Arch. Ital. Biol.* 108: 106-120.
- Manni E., Bortolami R., Deriu P.L. (1970b) Superior oblique muscle proprioception and the trochlear nerve. *Exp. Neurol.* 26: 543-550.
- Manni E., Desole C., Palmieri G. (1970c) On whether eye muscle spindles are innervated by ganglion cells located along the oculomotor nerves. *Exp. Neurol.* 28: 333-343.
- Manni E., Palmieri G., Marini R. (1971a) Peripheral pathway of the proprioceptive afferents from the lateral rectus muscle of the eye. *Exp. Neurol.* 30: 46-53.
- Manni E., Palmieri G., Marini R. (1971b) Extraocular muscle proprioception and the descending trigeminal nucleus. *Exp. Neurol.* 33: 195-204.
- Manni E., Palmieri G., Marini R. (1972a) Pontine trigeminal terminations of proprioceptive afferents from the eye muscles. *Exp. Neurol.* 36: 310-318.

- Manni E., Palmieri G., Marini R. (1972b) Mesodiencephalic representation of the eye muscle proprioception. *Exp. Neurol.* 37: 412-421.
- Manni E., Palmieri G., Marini R. (1974) Central pathway of the eye muscle proprioception. *Exp. Neurol.* 42: 181-190.
- Manni E., Bortolami R., Pettorossi V.E., Callegari E. (1976) Trigeminal afferent fibres in the trunk of the oculomotor nerve of lambs. *Exp. Neurol.* 50: 465-476.
- Manni E., Bortolami R., Pettorossi V.E., Callegari E. & Lucchi M.L. (1977) Reflex and reticular modulation of first-order proprioceptive neurons of the mesencephalic trigeminal nucleus. *Arch. Ital. Biol.* 115: 20-37
- Manni E., Bortolami R., Passatore M., Perinetti Casoni R., Lucchi M.L. (1978a) Vagal and reticular influence on the first-order proprioceptive neurons of the mesencephalic trigeminal nucleus in birds. *Arch. Ital. Biol.* 116: 25-42.
- Manni E., Bortolami R., Pettorossi V.E., Lucchi M.L., Callegari E. (1978b) Afferent fibres and sensory ganglion cells within the oculomotor nerve in some mammals and in man. II. Electrophysiological investigations *Arch. Ital. Biol.* 116: 16-24.
- Manni E., Bortolami R., Pettorossi V.E., Lucchi M.L., Callegari E. (1979) Una via alternativa per la conduzione di impulsi trigeminali dolorifici. In: Pagni C.A., Procacci C., Ventafridda V. (eds.): *Il Dolore: Problemi di Fisiopatologia e Terapia*. Cortina, Verona. Pp. 51-55.
- Manni E., Bortolami R., Passatore M., Lucchi M.L., Filippi G.M. (1980) Epileptogenic stimulation of cortical masticatory area and the mesencephalic trigeminal nucleus. *Arch. Ital. Biol.* 118: 89-104.
- Manni E., Bortolami R., Pettorossi V.E., Lucchi M.L., Callegari E. (1981) A direct trigeminal-oculomotor pathway. In: Fuchs A. F., Becker W. (Eds.) *Progress in Oculomotor Research*. Elsevier/ North Holland, Amsterdam. *Developments in Neuroscience* 12: 281-287.
- Manni E., Lucchi M.L., Filippi G.M., Bortolami R. (1982) Area postrema and the mesencephalic trigeminal nucleus. *Exp. Neurol.* 77: 39-55.
- Manni E., Bortolami R., Pettorossi V.E., Callegari E., Lucchi M.L., Ferraresi A. (1984) Afferent trigeminal fibres in the oculomotor nerve and their physiological role. *Documenta Ophthalmologica* 58: 101-107.
- Manni E., Bortolami R., Pettorossi V.E., Lucchi M.L., Callegari E., Draicchio F. (1987) Influence of oculomotor nerve afferents on central endings of primary trigeminal fibres. *Arch. Ital. Biol.* 126: 29-39.
- Manni E., Bortolami R., Pettorossi V.E. (1989a) Afferent trigeminal fibres in the oculomotor nerve. In: Gainotti G., Bentivoglio M., Bergonzi P., Ferro F.M. (Eds.) *Neurologia e Scienze di Base, Scritti in Onore di G. Macchi*. Vita e Pensiero, Milano. Pp. 63-77.
- Manni E., Draicchio F., Pettorossi V.E., Carobi C., Grassi S., Bortolami R., Lucchi M.L. (1989b) On the nature of afferent fibers of oculomotor nerve. *Arch. It. Biol.* 127: 99-108.
- Marini R., Bortolami R. (1979) Somatotopic organization of second order neurons of the eye muscle proprioception. *Arch. Ital. Biol.* 117: 45-57.
- Marini R., Bortolami R. (1980) Somatotopic representation of eye muscle proprioception within the superior colliculus of the lamb. *Exp. Neurol.* 69: 226-238.
- Marini R., Bortolami R. (1982) A somatotopic and functional organization of the masticatory proprioceptors in the mesencephalic trigeminal nucleus of the frog. *Arch. Ital. Biol.* 120: 385-396.

- Palmieri G., Asole A. (1967) Sulla presenza di cellule gangliari nei nervi oculomotori di alcuni mammiferi ed uccelli. *Riv. Biol.* 60: 117-137.
- Passatore M., Bortolami R., Manni E. (1979a) Somatotopic arrangement of the proprioceptive afferents from the jaw muscles in the mesencephalic trigeminal nucleus of the duck. *Arch. Ital. Biol.* 117: 123-139.
- Passatore M., Bortolami R., Lucchi M.L., Filippi G.M., Manni E. (1979b). Corticofugal influences of the cerebral masticatory area on the mesencephalic trigeminal nucleus of the rabbit. *Arch. Ital. Biol.* 117: 340-360.
- Passatore M., Lucchi M.L., Filippi G.M., Manni E., Bortolami R. (1983) Localization of neurons innervating masticatory muscle spindle and periodontal receptors in the mesencephalic trigeminal nucleus and their reflex actions. *Arch. Ital. Biol.* 121: 117-130.
- Pettorossi V.E., Ferraresi A., Draicchio F., Errico P., Santarelli R., Manni E. (1995) Extraocular muscle proprioception and eye position. *Acta Otolaryngol. (Stockh.)* 115: 137-140.
- Pettorossi V.E., Manni E., Errico P., Ferraresi A., Bortolami R. (1997) Otolithic and extraocular muscle proprioceptive influences on the spatial organization of the vestibulo- and cervico-ocular quick phase. *Acta Otolaryngol. (Stockh.)*, 117: 139-142.
- Tozer F.M. (1912) On the presence of ganglion cells in the roots of III, V and VI cranial nerves. *J. Physiol. (London)* 45: 15-16.