氏名	VIDYA VARATHAN（ヴィディヤ ヴァラタン）
博士の専攻分野の名称	博士（歯学）
学位記番号	第 17751 号
学位授与年月日	平成15年3月25日
学位授与の要件	学位規則第4条第1項該当 歯学研究科歯学臨床系専攻
学位論文名	Nitric Oxide Synthase/Nicotinamide Adenine Dinucleotide Phosphate-Diaphorase in the Brainstem Trigeminal Nuclei after Transection of the Masseteric Nerve in rats. （ラット咬筋神経切断後の脳幹三叉神経核における一酸化窒素合成酵素/ニコチンアミド アデニンジヌクルオチド フォスフェート ジアホラーゼ）
論文審査委員	（主査） 教授 古川 惣平 （副査） 教授 脇坂 聡　助教授 米原 典史　講師 竹村 元秀

論文内容の要旨

Abstract

Nitric Oxide (NO) is a short-lived free radical which has its actions in the nervous, cardiovascular and immune systems. NO synthase (NOS), the enzyme responsible for NO synthesis, requires nicotinamide adenine dinucleotide phosphate (NADPH) to be enzymatically active. Neuronal NOS (nNOS) and NADPH-diaphorase (NADPH-d) have been demonstrated to be present in edentical cell populations and their activity has been shown to parellel NO production and the presence of cyclic guanosine monophosphate.

Peripheral nerve transection has been reported to induce a number of changes of NADPH-d and/or nNOS in both peripheral sensory neurons and motoneurons, as well as transsynaptic changes in neurons within the central nervous system. The changes are primarily seen on the ipsilateral side, but bilateral changes have been reported in both cranial and spinal neuronal systems. In general, the contralateral changes are qualitatively similar to those on the ipsilateral side but are usually generally smaller.

In this study, the responses of NADPH-d and nNOS activities were quantitatively analyzed at different time course in both ipsilateral and contralateral sides of trigeminal nuclei, after unilateral trigeminal muscle nerve transection, in Sprague Dawley rats.

Methods

A total of 48 male Sprague Dawley rats were anesthetized eith sodium pentobarbital (60 mg/kg ; i.p.), and the masseter nerve was ligated as centrally as possible at two points and the intervening nerve was transected and removed. At fixed post-operative periods, animals were deeply anesthetized and a fixative containing 0.5% paraformaldehyde in phosphate buffer was used for the NADPH-d histochemistry whereas 4% paraformaldehyde

in phosphate buffer was used for nNOS immunohistochemistry.

Numbers of NADPH-d-positive and nNOS-immunoreactive neurons in the trigemino-solitary complex were counted bilaterally and the labeled neuronal soma was traced with the aid of light microscope equipped with a camera lucida drawing tube.

Results

In the control animals, both NADPH-d- and nNOS-positive neurons were constitutively distributed in the rostrolateral solitary tract nucleus, dorsomedial part of trigeminal nucleus oralis (Vo/Sn), and superficial layers (VcI/II) of the trigeminal nucleus caudalis (Vc). NADPH-d-positive neurons appeared in the trigeminal mesencephalic nucleus (Vmes), ipsilaterally at 5 days (mean±SEM=30.5±5.6) and maintained until 8 weeks (33±10.6) after the denervation. In the trigeminal motor nucleus (Vmo), NADPH-d-positive neurons appeared transiently and bilaterally, peaking at one week (663.5±156.2, ipsilateral side ; 687.5±118.6, contralateral side) after unilateral denervation of the masseteric nerve. In both Vo/Sn and Vc, the number of NADPH-d-positive neurons in the control animals showed a decrease at 3 days, but significantly increased from fifth day to 1 week and gradually came down to the control values by 8 weeks, after the denervation. There were no significant differences observed between the two sides in both Vo/Sn and Vc. nNOS-positive neurons were similarly distributed and the number of labeled neurons changed similar of NADPH-d-positive neurons after the denervation, thougt the changes were delayed by approximately 1 week.

Conclusion

Unilateral masseteric nerve transection demonstrated conspicuous similarities of NADPH-d and nNOS activity in the trigemino-solitary complex, but the peak NADPH-d activity occurred 1 week prior to nNOS activity. NADPH-d and nNOS were freshly induced in the ipsilateral side of Vmes within one week after the nerve transection and the induction lasted upto 4-8 weeks. In the Vmo, transient and bilateral induction was observed in both NADPH-d and nNOS neurons. Unilateral nerve transection initially decreased both NADPH-d and nNOS neurons in the Vo/Sn and Vc bilaterally, but increased the numbers significantly thereafter which came back to the baseline level in 4-8 weeks duration.

論文審査の結果の要旨

本研究は、咬筋神経切断後の三叉神経感覚・運動核における NADPH-diaphorase（NADPH-d）および神経型一酸化窒素合成酵素（nNOS）の発現変化を組織化学的に明らかにした。NADPH-d は運動核では両側性に、中脳路核では切断側で新たに発現したが、三叉神経脊髄路核吻側亜核および尾側亜核では両側性に同様に発現を増減した。nNOS の発現は NADPH-d の発現様式と同様であったが、約一週間遅れた。

以上の結果は、三叉神経系における一酸化窒素の中枢制御機構の解明に重要な知見をあたえるものであり、博士（歯学）の学位を授与するに値するものと認める。