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Effects of α-Tocopherol in Rats with a Neuropathic Pain Syndrome

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The rat model of a neuropathic pain syndrome (transection of the sciatic nerve with encapsulation of its central end) was used to evaluate the efficacy of the antioxidant α-tocopherol in such syndromes. It was found that administration of α-tocopherol 3 days before nerve transection and then for 3 weeks thereafter it delayed the development of the pain syndrome, which subsequently tended to subside. In contrast, a 3-week α-tocopherol treatment started when the pain syndrome had already set in failed to influence its evolution. α-Tocopherol markedly reduced manifestations of inflammatory and degenerative processes in the denervated limb.

Key Words: *generator of pathologically enhanced excitation; neuropathic pain syndrome; α-tocopherol; lipid peroxidation*

Damage to peripheral nerves and the consequent hyperactivity of central neurons in the system of pain sensitivity lead to the development of pain syndromes. It has been shown that the formation of a generator of pathologically enhanced excitation (GPEE) [1,2] and the establishment of a pathological algetic system (PAS) are of key importance in the pathogenesis of these syndromes [4, 16]. When the neurons have become hyperactive, the regulation of free-radical lipid peroxidation (LPO) is impaired, and this leads to substantial changes in cellular metabolism [9,17,18]. Agents that prevent or diminish imbalance of the LPO-regulating system, in particular antioxidants, delay the pathological process [6,9,10] and promote suppression of neuronal activity [8].

The present work was undertaken to study how the antioxidant α-tocopherol might influence the time course of the neuropathic pain syndrome arising after transection of the sciatic nerve [5,6].

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MATERIALS AND METHODS

A total of 56 random-bred male rats weighing 180-200 g were used. In all rats, the sciatic nerve was

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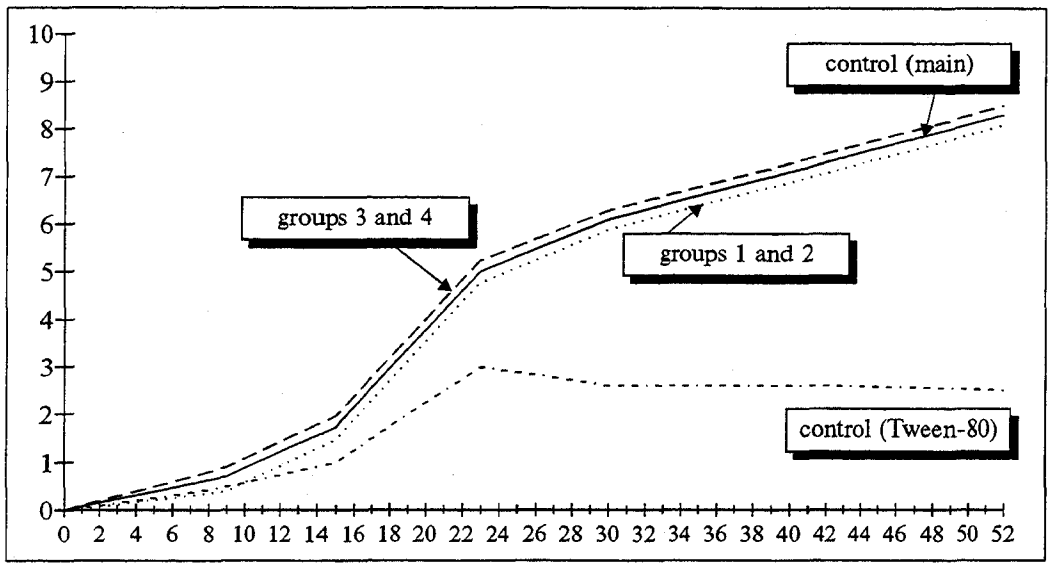

Fig. 1. Time course of the pain syndrome resulting from sciatic nerve transection in rats administered α-tocopherol and in control rats. Abscissa: days after nerve tran-section; ordinate: scores.

transected under ether anesthesia at the popliteal fossa level and its central end was placed in a sealed polyethylene tube and left in the wound, which was sutured [5,19]. The rats were divided into 7 groups, 8 animals in each. The first two groups consisted of rats given α-tocopherol (dissolved in Tween-80) in a dose of 100 mg/kg by the intraperitoneal route (group 1) or by mouth (group 2) 3 days before the operation and then once daily by the same route in a dose of 50 mg/kg for 21 days. Groups 3 and 4 comprised rats administered α-tocopherol at 50 mg/kg intraperitoneally (group 3) or by mouth (group 4) once daily for 21 days starting with postoperative days 13-14, when the pain syndrome had already set in. Groups 5 and 6 (control groups) consisted of rats given Tween-80 alone intraperitoneally (group 5) or orally (group 6). Finally, group 7 was composed of operated rats given neither α-tocopherol nor Tween-80 (the main control group).

The severity of the developing pain syndrome was scored by noting signs of autotomy on the deafferented hind paw. In all animals, pain sensitivity thresholds were determined by the hot-plate test. For this, rats were placed on a hot (55°C) plate and the times at which they began licking the paw or jumped off the plate were recorded.

RESULTS

During the first week after sciatic nerve transection, control rats (groups 5-7) and those pretreated with α-tocopherol (groups 1 and 2) developed motor disturbances of the paw paresis type. On postoperative days 6-7, rats began to use the operated paw for moving and scratching and to lick and bite the tips of digits; they became restless and irritable. Periodically, they were seen shaking the injured limb and pressing it against the belly; 1 or 2 rats in each group were found to have bitten off the tips of claws. On days 10-14, autotomies appeared and became increasingly pronounced in 3-4 rats of each group. Initially, no differences in the development of the pain syndrome were observed between the control and test animals. The hot plate test showed lowered pain sensitivity thresholds (rats were licking both the injured and intact paws), indicating that hyperalgesia had developed (Table 1). With progression of the pain syndrome, some animals in all groups developed inflammatory changes, edema of the paw or of digit tips, and ulcerations on the plantar side of the foot (trophic ulcers). On days 20-25, when 5-6 of the 8 rats from the main control (untreated) group had bitten off all claw tips and had gnawed away pieces of interphalangeal joints, less marked signs of autotomic damage were seen in the two groups administered α-tocopherol before nerve transection - the tips of only one or two claws had been bitten off and only 2 or 3 rats out of 8 had gnawed away pieces of interphalangeal joints, i.e., the pain syndrome was less severe than in the control rats. After days 20-25, no further progression of the pain syndrome was observed in the α-tocopherol-pretreated groups (Fig. 1). From that time on, the inflammatory process in the affected limb subsided, the trophic ulcers began to heal, and the hyperalgesia, as assessed by the hot-plate test, became less pronounced. Oral α-tocopherol administration was as effective as intraperitoneal.

In groups 3 and 4, where α-tocopherol treatment was started on postoperative day 13 or 14, when the severity of the pain syndrome was assigned score 2 or 3, the further development of this

TABLE 1. Effects of α-Tocopherol on Pain Sensitivity Thresholds as Measured (in Seconds) in Hot-Plate Testing of Rats with a Neuropathic Pain Syndrome Caused by Chronic Damage to the Sciatic Nerve ($M \pm m$)

Time measurement	Main control group	Tween-80-treated control group	Group given α-tocopherol orally on day 3 before operation and during 3 weeks after it	Group given α-tocopherol orally for 3 weeks after operation in the presence of pain syndrome
Before operation	9.3±0.42	9.6±0.6	9.2±0.4	9.4±0.8
After operation:				
day 3	9.4±0.6	9.0±0.7	9.7±0.6	9.2±0.8
day 7	8.8±0.8	9.0±0.6	8.9±0.6	8.9±0.5
day 10	8.2±0.7	8.6±0.4	6.8±0.6	7.8±0.5
day 14	6.8±0.9	7.4±0.6	6.3±0.7	6.8±0.6
day 17	6.1±0.4	6.2±0.5	6.1±0.4	6.3±0.6
day 21	5.4±0.6	5.8±0.6	6.8±0.5	5.8±0.6
day 24	5.1±0.8	5.4±0.5	7.4±0.6	5.4±0.6
day 28	5.2±0.5	5.0±0.5	8.0±0.7	5.3±0.6
day 31	5.0±0.6	5.3±0.6	8.4±0.8	5.2±0.5
day 35	5.8±0.7	6.1±0.5	8.3±0.7	5.9±0.6
day 38	6.4±0.4	6.6±0.4	8.9±0.8	6.4±0.5

syndrome did not differ from that in the controls (Fig. 1 and Table 1). In the treated animals, however, the inflammatory changes in the denervated paw were much less conspicuous.

Thus, as the results presented above indicate, the pretreatment of rats with α-tocopherol has a beneficial effect by reducing the severity of the pain syndrome developing after sciatic nerve transection. This effect was apparent 2 weeks after the operation and was more strongly marked one weak later, when the pain syndrome continued to progress in the control rats. In the two groups where α-tocopherol treatment was started after the pain syndrome had already set in, its further evolution did not differ from that in the untreated groups. Inflammatory and degenerative changes, however, were less pronounced in all α-tocopherol-treated groups.

During the formation of a denervational GPEE a hyperalgesia develops, brought about by structural, morphological, and biochemical changes in the dorsal horns of the spinal cord [11-13, 15]. This results in impaired postsynaptic inhibition in the dorsal horn's nociceptive neurons and promotes GPEE formation [3]. Many of the changes that arise are standard, typical intracellular changes [2]. One of these is intensified LPO in neuronal membranes, which leads to the appearance of excessive amounts of peroxides and free-radical oxidation products that exert toxic effects on cellular structures and favor the production of free fatty acids with the result that the neuronal membranes sustain further damage. The primary changes occurring in the system of LPO regulation induce secondary changes including accelerated prostaglandin synthesis which, in turn, by sensitizing pain receptors, results in increased action of bradykinin [16]. Administration of α-tocopherol has been shown to correlate directly with the lowering of LPO products in the central nervous system (CNS) to normal levels [10]. A protective action of α-tocopherol has been demonstrated in animal models of emotional pain stress [7]. Administration of α-tocopherol to rats for preventive purposes raises its levels in brain tissue and blood plasma [8] and prevents LPO activation in the CNS. As a consequence, GPEE formation is delayed so that the pain syndrome develops more slowly and may subsequently subside. If α-tocopherol is administered when the GPEE has already developed, further progression of the pain syndrome cannot be prevented or arrested. This indicates that LPO activation is a secondary process during pain syndrome development.

The results of the present study permit the conclusion that α-tocopherol may be found useful for combined pathogenetic therapy of a neuropathic pain syndrome associated with nerve injury, and that α-tocopherol treatment should be initiated as early as possible after the injury.

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Adaptive Stabilization of Structures and Adaptive Protection of the Heart in Rats of Two Different Genetic Strains: Role of Heat-Shock Proteins hsp70

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Periodic exposures of Wistar rats to a stressor resulted in the accumulation of five isoforms of heat-shock proteins (hsp70) in their myocardia and, as a consequence, in increased resistance to thermal damage shown by their isolated hearts, whereas in rats of the August strain such exposures did not lead to hsp70 accumulation and the heart's resistance was not increased. It is concluded that the ability of a given genetic strain to develop adaptive protection of the heart appears to depend on the ability of that strain to boost the expression of hsp70 genes in response to stressors.

Key Words: *stress; adaptation; heart; Wistar rats; August rats*

It has been established that adaptation to periodic stressful exposures is characterized by a broad spectrum of protective, notably cardioprotective, effects [1] and that important roles in adaptive protection of the heart are played, in addition to alterations in neurohumoral regulation, by mechanisms originating in the heart itself [2]. As a result, hearts isolated from stress-adapted animals have been found to exhibit greatly increased resistance to damage inducible by reperfusion, toxic concentrations of calcium and catecholamines, and heat [7,8]. Enhanced resistance to injurious factors is also displayed by intracellular structures isolated from such hearts, including sarcoplasmic reticulum elements [5] and mitochondria and nuclear DNA [7]. The set of alterations leading to enhanced

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