more easily if displaced vertically with respect to one another by about onesixth of the period. Inspection of Fig. 1 shows that this displacement is indeed seen in the micrographs.

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## Visceral Reflex Activity: Development in Postnatal Rabbit

Abstract. In postnatal rabbits there is progressive development of central regulation of the micturition reflex. During the first 1 to 2 days of postnatal life a coordinated sustained reflex may be obtained from the bladder which has been isolated from the central nervous system. Changes in reflex integration occur over the following 7 to 12 days, which result in a coordinated reflex dependent upon supraspinal influences.

Stimulation and ablation studies have indicated the significance of specific brainstem and spinal cord areas in the regulation of the micturition reflex in mammals (1). Transection of the brainstem of the cat below the level of the rostral pons results in abolition of the high-amplitude, sustained, contractile response of the urinary bladder elicited by slow retrograde filling (2). This contractile response with sequential passive relaxation of the sphincter effects completes evacuation of the bladder. The ontogeny of this reflex has not been investigated. Forty-eight New Zealand rabbits ranging in age from 1 day to 6 months were studied (3). Rabbits were anesthetized with ether;

tracheostomy was performed, and a polyethylene catheter of appropriate size was inserted through the distal urethra into the bladder. The urethral catheter was connected to a T-shaped cannula; one arm of the "T" was connected to the output of a positive displacement pump (4) and the other arm to a strain gauge whose output was recorded on a Model 680 Moseley Autograf strip chart recorder. Warm, normal saline was pumped into the bladder slowly at known infusion rates. A cystometrogram and a determination of the reflex threshold and amplitude of the contractile response were made while the intact animal was under light ether anesthesia. A laminectomy was per-



Fig. 1. Micturition reflex activity in a 1-day-old rabbit. 1, Cystometrogram in intact rabbit; 2, cystometrogram after complete transection of mid-thoracic portion of spinal cord; 3, cystometrogram after suction ablation of lumbo-sacral portion of the spinal cord. There is persistence of organized reflex response of the bladder after complete neurological isolation.

formed and the spinal cord was transected either at the cervico-medullary junction or at the mid-thoracic level. Transection of the spinal cord was performed by suction ablation, and more cystometrograms were made. This procedure was followed by laminectomy and suction ablation of the lumbo-sacral portion of the spinal cord, after which a third set of cystometrograms were made. The animals were killed and autopsied, and completeness of spinal cord transection was checked. Results of these experiments are shown in Fig. 1.

In the first 1 to 2 days of postnatal life neither section of the cervical and thoracic cord nor destruction of the lumbo-sacral cord abolished the micturition reflex. To the contrary, the micturition reflex was frequently increased in amplitude after these procedures (5). From the 3rd to the 13th or 14th day of postnatal life the reflex was inconsistently abolished by transection of the spinal cord at the mid-thoracic level. Subsequent ablation of the lumbosacral cord at this time did not abolish the bladder contractile response in one of the two 10-day-old animals that we tested. After the 2nd week of postnatal life, transection of the spinal cord at either the cervico-medullary junction or the mid-thoracic portion of the spinal cord resulted in loss of the micturition reflex. No consistent change was observed in either low-amplitude vesical rhythmic activity or slope of the tonus limb after the ablation procedures throughout this interval.

Developmental studies of tonic postural mechanisms have indicated that maturational changes occur in the first 2 to 3 weeks of postnatal life in a craniocaudal direction (6). These maturational changes are due to developmental changes in afferent reflex pathways. Studies in adult animals have indicated a close association between tonic postural mechanisms and micturition reflex threshold (7). A similar time course in developmental sequence of the micturition reflex and postural mechanisms would be anticipated. Our results confirm the similarity of temporal sequence in development of supraspinal regulation of the micturition reflex.

It is concluded from these experiments that the urinary bladder of the rabbit is in functional isolation from the central nervous system during the first 1 to 2 days of postnatal life. After this interval, activity of the bladder reflex is functionally integrated into the lumbo-sacral spinal cord. Following the integration into brainstem neural networks by the 2nd week of postnatal life, the bladder is not capable of organized reflex response if these central pathways are acutely interrupted.

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## Naphthaleneacetic Acid: Localization in the Abscission Zone of the Bean

Abstract. When 14C- naphthaleneacetic acid, labeled in the ring, was applied to the petiolar stub of debladed bean (Phaseolus vulgaris L.) plants, it accumulated in or between cell walls of tissue immediately adjacent and distal to the abscission layer. There was no localization in the abscission layer per se. Similar distribution patterns were observed in the abscission zone after naphthaleneacetic acid applications, which either delayed  $(10^{-3}M)$ or accelerated  $(10^{-5}M)$  abscission.

It is generally accepted that of the many factors influencing abscission, auxin appears most important. Two theories have been advanced to explain the action of auxin in abscission. Addicott et al. (1, 2) suggested that a gradient from relatively high auxin, distal to the abscission layer, to relatively low auxin, proximal to the abscission layer, is necessary to prevent abscission. A lowering of this gradient initiates or accelerates abscission. Gaur and Leopold (3), however, proposed that the total quantity of auxin applied was the controlling factor, and not the gradient. Using naphthaleneacetic acid (NAA), Biggs and Leopold (4) demonstrated a two-phase action: low concentrations accelerated, and high concentrations delayed, abscission. Significantly, they concluded that the primary action of auxin was directly on the abscission zone. Our experiments provide a test for both the auxin gradient and concentration theories and establish the localization of NAA in the abscission zone.

Bean (Phaseolus vulgaris L. cv 'Contender') seedlings, started in sand, were placed in nutrient solution and transferred to a controlled environment at 25°C and a light intensity of 4400  $lu/m^2$  (fluorescent, cool white). The

seedlings were debladed, and approximately 15 mg of a  $10^{-3}$  or  $10^{-5}M$ lanolin emulsion of ring-labeled (5) naphthaleneacetic acid (14C-NAA) was applied to the 1-cm petiolar stub.

Two plants were harvested from each concentration daily from 2 to 6 days (inclusive) after treatment, and a tissue explant containing the lower pulvinus was removed from the treated petiole. The tissues were quickly frozen on a Freon block, sectioned (58  $\mu$ ) with a cryostat, and affixed with adhesive onto glass slides. After the slides were dried



Fig. 1. Microradioautograms prepared from the lower pulvinus of the bean illustrating the localization of <sup>14</sup>C-NAA: (A) 2 days, (B) 3 days, (C) 4 days, (D) 5 days, and (E) 6 days after deblading at  $10^{-3}M$  NAA; (F) application of  $10^{-5}M$  NAA 6 days after deblading. P, petiole; AL, abscission layer; S, stem ( $\times$  200).

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