

The left-right side-specific endocrine signaling: implications for neurological deficits in stroke and neurodevelopmental disorders

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Brain injury-induced neurological deficits typically develop on the contralateral side of the body and include abnormal posture, motor weakness, and spasticity. It is believed that the interruption of descending neural pathways that convey supraspinal commands to the motoneurons in the spinal cord is the main cause of these deficits. This long-standing paradigm was challenged by our recent findings: a unilateral injury of the hindlimb sensorimotor cortex of rats with prior complete transection of the spinal cord produced hindlimb postural asymmetry (HL-PA), asymmetric hindlimb withdrawal reflexes, and asymmetry in gene expression patterns in the lumbar spinal cord (Figure 1; Lukoyanov et al., 2021). Strikingly, the contralateral hindlimb was flexed as usually seen in rats with intact spinal cord after brain injury. Hypophysectomy abolished the injury-induced effects, whereas hindlimb postural asymmetry was induced by serum from animals with brain injury transfused into animals with the intact brains. Arg-vasopressin and β -endorphin, two pituitary neurohormones, induced the right side hindlimb responses in naïve animals, while their antagonists blocked HL-PA in rats with the left-brain injury. Thus, in addition to motor pathways descending from the brain to spinal circuits, the side-specific humoral

signaling mediates the effects of unilateral brain injury (UBI) on hindlimb posture and reflex asymmetries.

The phenomenon is robust, firmly established, and may be relevant for neurological deficits after stroke and traumatic brain injury, and for neurodevelopmental disorders such as cerebral palsy. Yet a precise mechanism of side-specific endocrine signaling is to be established.

The left-right side-specific humoral signaling from injured brain: Brain lesions interrupt descending neural pathways from the cerebral cortex to motoneurons in the brain stem and the spinal cord. Deficits in voluntary and skilled movements are linked to impairment of the corticospinal and rubrospinal tracts, whereas spasticity, changes in reflexes and postural abnormalities to aberrant activity of the reticulospinal and vestibulospinal tracts.

An alternative pathway that does not engage descending neural tracts but may convey side-specific signals from the brain to the paired endocrine glands, the left and right pituitary gland, and the left and right extremities had been suggested (for references, see Lukoyanov et al., 2021) and supported by early evidence (Bakalkin et al., 1986).

We hypothesized that this pathway, the side-specific neuroendocrine signaling may operate in parallel with the descending neural tracts and mediate the effects of a UBI on the hindlimb posture and motor functions. To test this hypothesis, we disabled the descending neural influences in order to reveal the endocrine signaling (Lukoyanov et al., 2021). For this purpose, the spinal cord was completely transected before the brain injury was performed. We observed that a unilateral injury of the hindlimb sensorimotor cortex of rats with completely transected thoracic spinal cord produced HL-PA with contralateral flexion and asymmetric hindlimb withdrawal reflexes, as well as asymmetry in gene expression patterns in the lumbar spinal cord. The brain injury-induced postural effects were abolished by hypophysectomy and were induced in animals with the intact brains by transfusion of serum from animals with brain injury. Thus, the effects of brain injury in this experimental setting were mediated by the left-right side-specific humoral signaling (Figure 1).

The left-right side-specific effects of neurohormones: Pharmacological experiments demonstrated that the effects of a unilateral brain lesion may be mimicked by administration of neurohormones including Arg-vasopressin and opioid peptides into animals with an intact brain (Bakalkin et al., 1986; Bakalkin and Koblyyansky, 1989; Watanabe et al., 2020, 2021). The remarkable finding is that the side of the flexed limb depends on the compound administered. The left hindlimb was flexed after administration of the μ - δ -agonist Met-enkephalin, and the selective κ -agonists dynorphin and U-50488. Conversely, Leu-enkephalin (acting through δ -receptor), β -endorphin, and Arg-vasopressin caused the right hindlimb flexion. The asymmetric response may be mediated through the opioid system lateralized in the spinal cord (Kononenko et al., 2017; Watanabe et al., 2021). The levels of opioid peptides and expression of opioid genes were lateralized, and the inter-regional co-expression patterns of these genes were side-specific in the spinal cord. The neurohormonal effects were evident at their intrathecal, intracisternal, and intravenous administration suggesting that the “non-directional” molecular messengers circulating in the blood or cerebrospinal fluid may convey topographical information that is converted into left-right side-specific postural and motor responses upon activation of lateralized receptors.

The pituitary gland is the main source of Arg-vasopressin and opioid peptides including

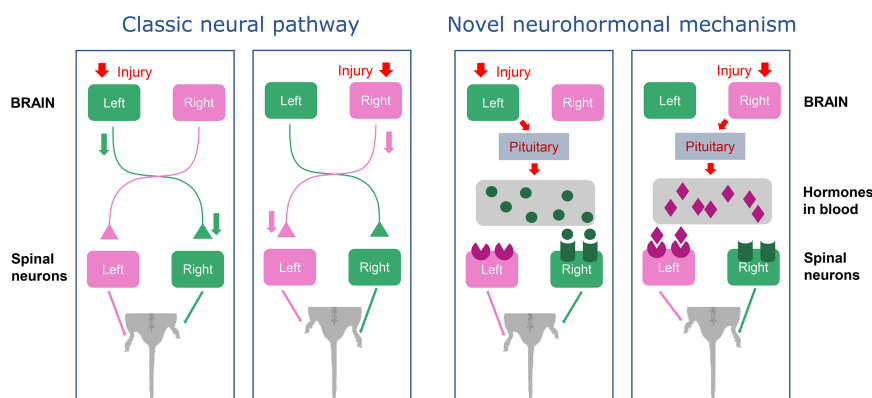


Figure 1 | The effects of brain injury on contralateral motor function. Injury to the hindlimb region on one side of the brain impairs the function of the hindlimb on the opposite side of the body resulting in the abnormal asymmetric posture. The effect may be mediated by two different mechanisms. In the classic mechanism (left panel), brain injury impairs the descending neural tracts that decussate and control the opposite hindlimb. In the novel humoral mechanism (right panel), brain injury stimulates the release of side-specific neurohormones from the pituitary gland into the blood, they bind to neuronal receptors that are lateralized in the spinal cord and affect the contralesional hindlimb. The neurohormonal signaling may operate in addition to the neural mechanism, by contributing to long-lasting side-specific neuroplastic changes that underlie asymmetric postural and motor deficits such as hemiparesis and hemiplegia (Lukoyanov et al., 2021). Modified from Wolpaw and Carp (2021).

β -endorphin and dynorphins in the body that are secreted into the bloodstream suggesting their role in neurological responses induced by a UBI. Evidence that either SSR-149415 (an antagonist of vasopressin V1B receptor, which is expressed in the pituitary gland) or naloxone (a general opioid antagonist) administered to animals with a left UBI with transected spinal cord blocks the formation of HL-PA, supports this hypothesis. Similarly, HL-PA induced by serum from animals with left UBI was abolished by administration of either SSR-149415 or naloxone. Thus, the pituitary neurohormones Arg-vasopressin opioid peptides released into the systemic circulation may serve as the side-specific signals that mediate UBI effects on hindlimbs.

In animals with the intact spinal cord, selective antagonists of the μ - and κ -opioid receptors β -funaltrexamine and nor-binaltorphimine, respectively, inhibited HL-PA formation after the right but not left UBI (Watanabe et al., 2020, 2021). In contrast, the δ -antagonist naltrindole inhibited HL-PA after the left but not right-side brain injury. These findings together with the aforementioned effects of Met- and Leu-enkephalins and the selective κ -agonists dynorphin and U-50488 suggest that the postural and motor deficits resulting from the right-side brain injury are mediated through the μ - and κ -opioid receptors whereas those from the left-side injury through the δ -receptors.

The left-right side-specific endocrine signaling may have four key phases: 1) signaling from injured cortical sites to the hypothalamus; 2) converting the side-specific signals in the hypothalamus and the pituitary gland into a topographically specific hormonal message; 3) humoral transmission of this message to its target(s) on peripheral nerve endings or spinal neurons; and 4) translation of this message into left-right side-specific responses through lateralized receptors to these hormones.

In summary, these studies describe a novel, unusual phenomenon, the side-specific endocrine mechanism that mediates asymmetric effects of a UBI on hindlimb posture and motor functions (Figure 1). It would be worthwhile to ascertain whether signals mediated by the endocrine and neural pathways are additive, synergistic, or even antagonistic with respect to each other. Analysis of asymmetric sensorimotor deficits transmitted by neurohormonal signals versus those mediated by neural pathways is essential for understanding mechanisms of sensorimotor deficits and could facilitate new therapeutic discoveries. From a biological standpoint, the mechanism may serve to maintain a balance between the left-

right physiological and molecular processes in bilaterally symmetric animals.

The endocrine mechanisms in intergenerational transmission (IGT) of neurological signals: Effects of environmental impact may be transmitted to the following generation and cause neuropsychiatric disorders including depression, anxiety, and posttraumatic stress disorder in the offspring (for references, see [Carvalho et al., 2021]). The transmission may be at least in part mediated by the neuroendocrine system. Having evidence for the left-right side-specific neurohormonal signaling from the injured brain, we examined if the effects of UBI are intergenerationally transmitted from pregnant rats to the offspring, and whether the resulting neurological deficits are asymmetrical (Carvalho et al., 2021). Ablation of the hindlimb sensorimotor cortex in pregnant rats resulted in the development of HL-PA and impairment of balance and coordination in the offspring. Strikingly, the effects depended on the UBI side. After left UBI in pregnant dams, the motor deficits in the offspring were apparent on the contralesional (i.e., right) side and were exhibited both before and after spinal cord transection. In contrast, the effects of right UBI in the offspring were cryptic; the HL-PA was evident only after spinalization and randomly lateralized to the left and right side. The lateralized effects of brain injury may be transmitted from mother to fetus by the maternal left-right side specific endocrine mechanism. The persistence of the deficits after spinalization suggests that the asymmetry is either encoded in spinal circuits or developed due to impaired balance of the left and right neuroendocrine pathways in the offspring. The revealed phenomenon requires a detailed analysis of neurological deficits in the offspring, assessment of the role of the maternal endocrine system in the formation of pup's neurological status, and analysis of neuroplasticity in spinal neurocircuits as a basis of long-lasting neurological changes in the offspring.

Implementation of IGT of neurological deficits for cerebral palsy (CP): Unilateral CP is a common syndrome and is characterized by asymmetric impairments of muscle tone, balance, coordination, and posture. The rat IGT findings suggest that small cryptic localized brain injuries or neuroinflammation in pregnant individuals may signal through the left-right side-specific endocrine system resulting in pathological changes in the developmental central nervous system and asymmetric neurological deficits in the adult offspring.

Risk factors that account for 70–80% of CP cases occur in the prenatal period. They may include prematurity and low birth weight, intrauterine infections and inflammation, multiple gestations, and pregnancy complications in the mother such as preeclampsia. However, two-thirds of CP patients are born at term, and birth asphyxia happens in less than 10% of CP cases, and the estimated upper limit of unconfirmed CP causality is 80% of all cases (Li et al., 2021). Thus, there might be another mechanism that underlies the majority of CP cases and that has not yet been revealed.

Animal models of CP (Cavarsan et al., 2019) generally focus on developmental insults including hypoxia, ischemia, and neuroinflammation that result in anatomically severe injuries. Most of them do not model prenatal mechanisms and severe motor phenotype that includes spasticity and weakness, and that is generally life long-lasting. Intra-amniotic infection and inflammation are the only prenatal mechanism supported by experimental evidence.

The IGT of neurological deficits may be considered as a model which exhibits several CP features that are not or are only partially addressed in previous animal studies. In contrast to previous CP models (but excluding intrauterine inflammation), the IGT model may account for antenatal causes of 70–80% CP cases. Other features modeled by the IGT are the asymmetric postural and motor deficits that are typical for CP patients. In animal experiments, these deficits were evident during the whole observation period that lasted for 2 months and persisted even after spinal cord transection (Carvalho et al., 2021). CP is associated with an increased risk of neurodevelopmental disorders in siblings. Consistently, in the IGT model, most progenies in the same litter did not differ in extent of neurological deficits after brain injury to pregnant rats suggesting that they are similarly affected by the intergenerational mechanism. These CP features were not apparently modeled in the previously developed animal approaches to study CP pathology.

A large fraction of subjects with CP do not relax their muscles – they are tonically constricted without any voluntary command. This phenomenon is called spastic dystonia and is defined as “stretch- and effort-unrelated sustained involuntary muscle activity following central motor lesions” (Lorentzen et al., 2018). Spastic dystonia may have a central mechanism which does not depend on afferent input in contrast to spasticity that is based on exacerbated reflex excitability. It is postulated that

rhizotomy which abolishes sensory input and diminishes stretch reflexes would have weak or no effect on spastic dystonia. Indeed, the effects of dorsal rhizotomy on muscle contractures in children with CP are still in question (for references see Lorentzen et al. 2018).

Postural changes induced by a UBI in rats were partially resistant to bilateral deafferentation from the L1 to S2 spinal levels (Zhang et al., 2020). Thus, modification of spinal reflexes is not solely a cause of HL-PA. These findings suggest that formation of HL-PA in the offspring of pregnant rats with brain injury may represent a small animal model for the investigation of spastic dystonia and its role in CP.

Preeclampsia, one of the key factors contributing to CP, may be developed due to dysregulation of Arg-vasopressin and lead to the formation of aberrant asymmetric neurocircuit connectivity in the offspring in humans. Thus, Arg-vasopressin infusion has been used to model clinical features of preeclampsia in animals (Sandgren et al., 2018). Considering that Arg-vasopressin also induces asymmetric posture in intact animals and mediates the effects of brain injury on abnormal posture and reflexes (Lukoyanov et al. 2021), we may hypothesize that this neurohormone may be one of the causal factors in IGT of the effects of brain injury from mother to fetus resulting in asymmetric motor deficits in the offspring.

Slow progress in the development of pharmacological treatments of CP may be due to the absence of small animal models that uncover mechanisms of this disorder and allow for screening of pharmacological agents. The IGT model may fill this gap by modeling several clinical features of this neurodevelopmental disorder, and by prioritization of hypothalamic-pituitary hormones with left-right side-specific actions as pharmacological targets. Dysregulation of this neuroendocrine system in mothers by brain injury or by other factors, e.g., preeclampsia, may be one of the causes of neurological deficits in the offspring.

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