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Morphometric Analysis of Appendicular and Spinal Ventral Horn Growth -A Correlative Study in Human Foetuses.

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ABSTRACT

Background: Literature showing growth pattern of cervical and lumbar enlargements of spinal cord and the limbs to which they supply and their inter-relationships do not exist in literature in human samples. **Methods:** Spinal cords were dissected out from 30 normal human foetuses of different gestational ages, divided into five equal groups. Mean length of arm, forearm and thigh were determined in each group. Sections obtained from cervical and lumbar enlargements were stained with hematoxylin and eosin to determine mean transverse diameters of their ventral horns in each foetal group. **Results:** Length of the forearm was smaller than that of the arm till third trimester when a relative spurt in growth of forearm was noticed. **Conclusion:** Direct correlation exhibited in growth patterns between transverse diameters of ventral horn of cervical enlargement and forelimb measurements on one hand and that of lumbar enlargement and hind limb parameters on the other hand with a spurt of growth between second and third groups of foetuses.

Keywords: Limb growth, Ventral horn, Histomorphometry, Human foetuses.

INTRODUCTION

Cervical and lumbar enlargements of spinal cord control motor and sensory functions of upper and lower limbs respectively in adult.[1] Neurons connected to musculature of limbs are located in the lateral group in the ventral horn of these enlargements.[2] Obviously these neural groups are specifically responsible for aforementioned enlargements. Phenomenon of cephalocaudal gradient in embryo makes the forelimbs to appear earlier than the hind limbs.[3]

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The hind limbs in spite of late appearance compared to forelimbs in early embryo grow faster to become larger which is well appreciated in postnatal life in human. A correlation is supposed to exist between growth of ventral horns of spinal cord and those of limbs. Foetal anatomy seems to be emerging speciality in recent days due to being indispensable with in utero foetal surgery. The latter is made possible due to development of highly sophisticated surgical instruments and imaging techniques. The early diagnosis of foetal disorders that may be irreversible in the neonate can by means of in utero surgery.[4] Scientists do agree that in spite of detailed information related to foetal anatomy available in literature, there are enormous facts of the subject yet to be explored.[5-7] Step in the development of limbs based on simple observations are well documented[8] but quantitative approach to different parts of developing limbs in developing foetuses is lacking in literatures. The same is true for ventral horns of enlargements of spinal cord directly related to developing limbs. Some earlier correlative studied were performed in developing lower animals. Ghazi and Gholmani studied growth of spinal cord in relation to ventral column in sheep.[9] Hifry et.al. showed relation between the ventral column and spinal cord in Equus asinus.[10] Salka did quantitative studies on postnatal growth of spinal cord and ventral column of albino mice.[11] Therefore present information in human foetuses will be unique of its kind. In the present study, we

have planned to perform quantitative measurements on different parts of both upper and lower limbs and ventral horns of cervical and lumbar enlargements of spinal cord simultaneously in foetuses of different age groups to find out the patterns of their growth. The attempt will also be made to find out correlation between them if any.

MATERIALS AND METHODS

Thirty formalin fixed human foetuses of either sex preserved in the museum of department of anatomy were procured. All the foetuses were devoid of any apparent anomaly. The length of foot from tip of second toe to back of heel, was measured by Streeter's method to determine the gestational age of foetuses.[12] Foetuses in Groups I, II, III, IV and V were <17 weeks 17-20 weeks, 21-25 weeks, 26-30 weeks and >30 weeks respectively. Permission was sought from Institutional Ethical Committee before considering the human foetuses for research purposes. Length of arm, forearm and thigh were measured with the help of measuring tape.[13] Reference points for measuring arm were angle of acromion process and lateral epicondyle of humerus. Forearm was measured from lateral epicondyle of humerus to radial styloid process. Length of thigh was considered between anterior superior iliac spine and joint line of knee on medial aspect. Spinal cord was exposed in the vertebral canal by laminectomy starting from sacral hiatus to the posterior arch of atlas on each side of midline. Two mm thick slices of spinal cord were obtained at maximum widths of cervical and lumbar enlargements by cross incisions using sharp knife. Seven-micron thick sections received by wax embedding technique were stained with hematoxylin and eosin. The width of the ventral horn perpendicular to its long axis (transverse diameter) was measured with the help of slide micrometer in both cervical and lumbar enlargements of spinal cord. The measurements of arm and forearm lengths of forelimbs (upper limbs) and ventral horn transverse diameters of cervical enlargements were plotted on the graph to find out their pattern of growths and interrelationships. Graphs were also plotted for measurements of thigh length and transverse diameter of ventral grey horn of lumbar enlargement for the same purpose. Transverse diameters of the ventral horns of cervical and lumbar enlargements were also interrelated by finding out their per cent differences in different groups of foetuses.

RESULTS & DISCUSSION

Lengths of all the three appendicular regions i.e. arm, forearm and thigh, showed steady growth throughout intrauterine life [Figure 4]. Invariably a spurt of growth was noticed in all said regions between second and third group of foetuses. The readings were minimum, as expected in first group i.e. 32.5 mm, 25.0 mm and 37.8 mm respectively which were raised to maximum in group five foetuses i.e. 64.7 mm, 66.7 mm and 74.5 mm respectively [Table 2]. The striking feature in the said readings was relative lengths of arm and forearm. Length of arm was larger compared to forearm in first three groups but in last two groups due to relative spurt in growth of forearm, the later equaled and then slightly increased in length compared to the arm [Table 2], [Figure 4]. Surprisingly this period coincided with quickening which was known to affect growth of limb bones.

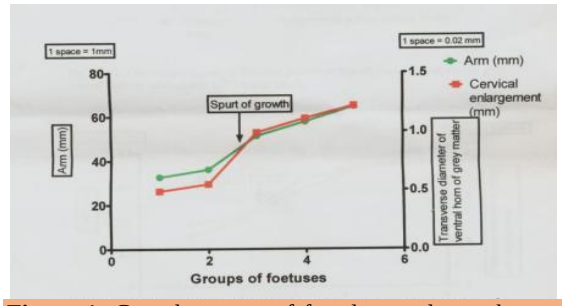

Figure 1: Growth pattern of foetal ventral gray horn of cervical enlargement and arm length.

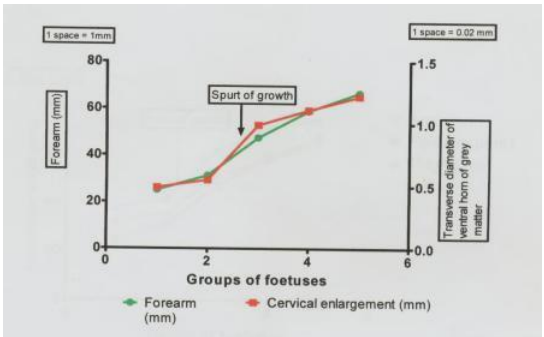

Figure 2: Growth pattern of foetal ventral gray horn of cervical enlargement and forearm length.

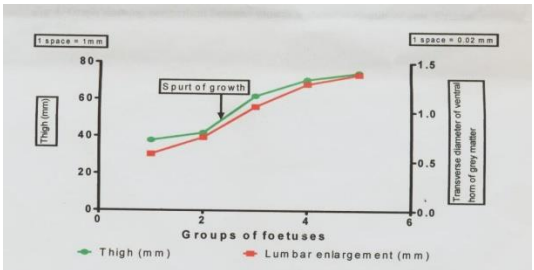

Figure 3: Growth pattern of foetal ventral gray horn of cervical enlargement and thigh length.

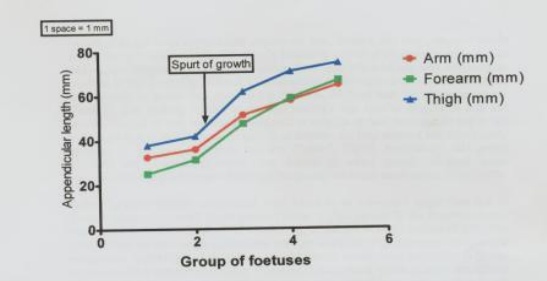

Figure 4: Growth pattern of in length of arm, forearm and thigh.

Ventral horns of lumbar enlargement were found to be constantly larger than that of cervical enlargement in all foetal groups [Table 1]. This pattern in the interrelationship was exactly the same as observed between upper and lower limbs lengths [Figure 4]. The graphs plotted for arm length and transverse diameter of ventral horn of grey matter of cervical enlargement [Figure 1], forearm and that of cervical enlargement [Figure 2] and thigh length and lumbar enlargement [Figure 3] further support our aforementioned findings. The most striking result which strongly strengthen this correlation was spurt of growth observed in both ventral horn size and limb length simultaneously between second and third group of foetuses [Figure 1,2,3]. Related work was lacking except following ones, which only indirectly supported our conclusions. Although limb was non functional during intrauterine life, but its spontaneous embryonic activity had been shown to play role in neuron and muscle development.[15] This indirectly explained the quantitative correlation between muscle and ventral horn. Direct evidence of such finding was established by Clowry et al[16] who found muscle afferents crossing the dorsal horn in human foetuses of 7.5 weeks of gestation and innervating motor neurons by 9th gestational weeks. By 13th weeks motor neurons were segregated into distinct columns. This investigation contributed towards a time table for the functional development of human motor control.

Table 1: Mean transverse diameter (mm) of ventral horn of grey matter of spinal cord in human Foetuses

Groups of foetuses	Cervical enlargement	Lumbar enlargement	Per cent difference
I	0.49	0.57	16
II	0.55	0.74	35
III	0.99	1.05	06
IV	1.11	1.28	15
V	1.22	1.38	13

Table 2: Mean lengths (mm) of different regions of limbs in human foetuses

Groups of foetuses	Arm	Forearm	Thigh
I	32.5	25.0	37.8
II	36.0	31.3	41.9
III	51.3	47.4	61.8
IV	57.7	58.5	70.8
V	64.7	66.7	74.5

CONCLUSION

Direct correlation exhibited in growth patterns between transverse diameters of ventral horn of cervical enlargement and forelimb measurements on one hand and that of lumbar enlargement and hind limb parameters on the other hand with a spurt of growth between second and third groups of foetuses.

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