Comparative histomorphometric study of the various segments of the spinal cord in the adult male and female mongoose (Herpestes edwardsii)

Rasouli, B.*, Gholami, S.

Department of Basic Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran

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Correspondence

Rasouli, B.

Department of Basic Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran Tel: +98(71) 36138823 Fax: +98(71) 32286940 Email: babrs32@yahoo.com

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Abstract:

BACKGROUND: Anatomical and histological studies of the spinal cord have always garnered anatomists' attention because of their high importance in various fields of veterinary medicine, zoology and behavioral science. **OBJECTIVES:** This work was conducted to understand the detailed histomorphometric aspects of the spinal cord of Indian gray mongoose (Herpestes edwardsii). METHODS: Six adult Indian grey mongooses in the terminal stages of disease and the status of approaching death were used in the present study. The spinal cords were dissected and fixed in 10% buffer formalin then paraffinized and sections of 6 µm thick were cut and stained with haematoxylin and eosin. In this study, the vertical and transverse diameters of the spinal cord, central canal, the length of the ependymal cells and the ratio of gray matter to white matter in the selected spinal cord segments, were measured with standard micrometric method using light microscope. RESULTS: In male and female mongooses, the longest transverse and vertical diameters of spinal cord segments were observed in the lumbar region. Although this stability and readability of the data were not seen in the transverse and vertical diameters of the central channel. Also, the highest ependymal cells in both sexes were observed in the lumbar region. CONCLUSIONS: It can be concluded that the transverse diameter of spinal cord is larger than the vertical one and the largest measured diameter and ratio of gray matter to white matter were identified in lumbosacral area.

Introduction

Mongooses are small carnivores occupying various regions from Africa to Southeast Asia. The genus Herpestes contains 10 species and is considered the oldest genus within the order Carnivora, dating back to approximately 30 million years (Shil et al., 2012). The Indian gray mongoose (Herpes-

tes edwardsii) is the specie of mongoose that is mainly found in southern Asia particularly India, Pakistan, the south of Iran, Sri Lanka and some other parts of Asia (Choudhury et al., 2011). The Indian gray mongoose is one of the few animals that can survive a cobra attack, which makes it one of the few predators of this deadly snake. This will be a platform for better understanding the physiology and pathology of diseases (Rasouli et al., 2015 and Wu et al., 2013).

The widest spinal cord segment of adult human was identified at 6th cervical segment and 12th thoracic segment (Williams et al., 1989). In addition, Sharma and Rao (1971) and Malinska et al. (1976) measured the spinal cord of buffalo and mole and reported different diameters of the spinal cord segments. The spinal cord segments were also studied microscopically in sheep (Ghazi and Gholami, 1994), camel (Ghazi et al., 1998), duck (Gholami et. al., 1999), cat and male dog (Mansouri et al., 2002), pigeon (Khaksar et al., 2002), guinea pig (Ghazi et al., 2005) and rabbit (Farag et al. 2012). No information appears to be available on histological study of the various segments of the spinal cord in mongoose. Therefore the present study was undertaken.

Materials and Methods

Six adult mongooses (3 male and 3 female) which have been found in the surrounding areas of Shiraz were used for this study during last two years. The age of the animals was estimated to be more than one year old by teeth inspection. These fresh specimens were in terminal stages of disease or the status of approaching death (severe hypothermia; 36°C body temperature) and were admitted to the Department of Anatomy and Embryology, Shiraz University, Iran.

All experiments conducted on animals were in accordance with the guidance of ethical committee for research on laboratory animals of Shiraz University.

The animals were euthanized with Ketamine 10% and Xylazine 2% and then 10%

buffered formalin solution was injected into the lateral ventricles of brain. This way, the fixative was spread easily in all ventricles, central canal of spinal cord and subarachnoid space. The specimens were then transferred into big containers of phosphate buffered formaldehyde solution, and 24 hours later, the spinal cord segments were further dissected free without damaging the meningeal layers. In the following, after discerning the spinal cord segments, the 1st, 4th and 8th cervical, 6th and 12th thoracic, 4th and 7th lumbar and 2nd sacral spinal cord segments were taken. Subsequently, 6 µm thick serial paraffin sections were prepared from each spinal segment and stained with haematoxylin and eosin. The transverse and vertical diameters of the spinal cord and central canal and the length of the ependymal cells in the sampled segments were measured by ocular micrometer. The ocular graticular lattice line was used to determine the proportion of gray matter to white matter (Farag et al., 2012). Analysis of morphometric data was carried out with Student's t-test using SPSS program (version 16).

Results

The longest to shortest transverse diameter of spinal cord segments in male mongoose were identified as follows: 4th lumbar, 1st and 4th cervical, 12th and 6th thoracic, 8th cervical, 7th lumbar and 2nd sacral segments. The 1st cervical and 4th lumbar spinal cord segment had the longest transverse diameter in female mongoose followed by 12th thoracic, 4th cervical, 6th thoracic, 8 cervical, 7th lumbar, and 2nd cervical segments (Table 1).

The 7th lumbar spinal cord segment had the longest vertical diameter in male mongoose followed by 12th thoracic, 4th, 1st and

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| Segments | Vertical diameter | Vertical diameter | | Transverse diameter | |
|----------|-------------------|-------------------|-------------------|---------------------|--|
| | Female | Male | Female | Male | |
| C1 | 2.25 ± 0.50 | 2.50 ± 0.35 | 5.50 ± 0.55 | 6.25 ± 1.00 | |
| C4 | $2.75 \pm 0.00*$ | 3.00 ± 0.05 | $3.50 \pm 0.50 *$ | 5.00 ± 0.75 | |
| C8 | 2.50 ± 0.45 | 2.50 ± 0.125 | 3.15 ± 0.125 | 3.25 ± 0.125 | |
| Т6 | $2.125 \pm 0.25*$ | 2.50 ± 0.40 | $3.25 \pm 0.20*$ | 4.50 ± 0.45 | |
| T12 | 2.75 ± 0.30 | 3.635 ± 0.25 | 4.25 ± 0.90 | 4.75 ± 0.00 | |
| L4 | $3.75 \pm 0.05*$ | 4.25 ± 0.40 | $5.50 \pm 0.35*$ | 6.75 ± 0.125 | |
| L7 | 1.875 ± 0.20 | 2.125 ± 0.35 | $2.50\pm0.05*$ | 3.15 ± 0.05 | |
| S2 | 1.875 ± 0.00 | 2.00 ± 0.25 | 1.75 ± 0.45 | 2.15 ± 0.90 | |

Table 1. Transverse and vertical diameters of different segments (Mean \pm SD, in mm) of the spinal cord in different segments in male and female mongooses (*Significant at p<0.05).

Table 2. Transverse and vertical diameters (Mean \pm SD, in mm) of central canal of the spinal cord in different segments in male and female mongooses (*Significant at p<0.05).

| Segments | Vertical diameter of central canal | | Transverse diameter of central canal | |
|----------|------------------------------------|-----------------|--------------------------------------|-----------------|
| | Female | Male | Female | Male |
| C1 | $0.11 \pm 0.03*$ | 0.14 ± 0.01 | 0.09±0.03* | 0.11±0.02 |
| C4 | 0.06±0.04 | 0.07±0.01 | 0.15±0.01* | 0.19±0.1 |
| C8 | 0.06±0.03 | 0.08 ± 0.03 | 0.10±0.00 | 0.10±0.04 |
| Т6 | 0.15±0.10 | 0.17±0.07 | 0.12±0.03 | 0.15±0.04 |
| T12 | 0.05±0.00* | 0.10±0.06 | $0.08 \pm 0.00*$ | 0.12±0.09 |
| L4 | 0.14±0.20 | 0.18±0.04 | 0.13±0.05 | $0.14{\pm}0.02$ |
| L7 | 0.15±0.20 | 0.17±0.02 | 0.12±0.02 | 0.12±0.01 |
| S2 | 0.09 ± 0.04 | 0.10 ± 0.00 | 0.05±0.01 | 0.05±0.03 |

8th cervical, 6th thoracic, 7th lumbar and 2nd sacral segments. The 4th lumbar spinal cord segment had the longest vertical diameter in female mongoose too, followed by 4th cervical and 12th thoracic, 8th and 1st cervical, 6th thoracic, 7th lumbar and 2nd sacral segments (Table 1).

Comparing the vertical and transverse and diameter of spinal cord segments in the two groups revealed that the transverse diameter is consistently longer than the vertical diameter except for the 2nd sacral segment in female animals (Table 1 and Figure 1).

The longest to shortest transverse diameter of central canal of spinal cord segments in the male mongoose was identified as follows: 4th cervical, 6th thoracic, 4th lumbar, 12th thoracic and 7th lumbar, 1st and 8th cervical and 2nd sacral segments. The 4th cervical and lumbar spinal cord segments had the longest transverse diameter of central canal of spinal cord in female mongoose followed by 6th thoracic and 7th lumbar, 8th cervical, 1st cervical, 12th thoracic and 2nd sacral segments (Table 2).

The 4th lumbar spinal cord segment had the longest vertical diameter of central canal in the male mongoose followed by 4th lumbar and 6th thoracic, 1st cervical, 2nd sacral, 12th thoracic and 8th and 4th segments. The 6th thoracic and 7th lumbar spinal cord segments had the longest transverse diameters of central canal in the female mongoose, followed by 4th lumbar, 1st cervical, 2nd sacral, 4th and 8th cervical and 12th thoracic segments (Table 2).

Statistical analysis showed significant differences in vertical and transverse di-



Figure 1. Transverse sections of the spinal cord in the male and female mongoose, based on the length of the spinal cord transverse and vertical diameters.

ameters of some spinal cord segments and their central canal between male and female mongoose (p<0.05) which are shown in Table 1 and Table 2.

The height of ependymal cells in all of the cervical spinal cord segments was similar in male and female mongooses and was measured 0.0075 mm. This height in the 6th and 12th thoracic spinal segments was 0.0100±0.00 mm and 0.0125± 0.0025 mm in male mongoose and 0.0100±0.00 mm and 0.0125 ± 0.0025 mm in females. In the 4th lumbar spinal segment, the height of these cells was 0.0150±0.005 mm in male, and 0.0125 ± 0.0025 in female mongoose. In both sexes, the height of ependymal cells in the 7th spinal segment was 0.0125. In the 2nd sacral spinal segment, the height of these cells was 0.0100±0.00 mm in male, and 0.0075±0.00 mm in female mongoose. Statistical analysis showed no significant differences in the height of the ependymal cells between male and female mongoose (p<0.05) (Table 3).

The largest to smallest ratio of gray matter to white matter of spinal cord segments in male and female mongoose are as follows: 2nd sacral, 4th and 7th lumbar, 1st, 4th and 8th



Figure 2. The proportion of gray matter to white matter of the spinal cord in different segments in male and female mongoose.

cervical, and 6th and 12th thoracic segments. The results showed that the proportion of gray matter to white matter of the spinal cord in sacral and lumbar segments was far more than other areas (Figure 2).

Discussion

The longest vertical and transverse diameters of spinal cord segments in male and female adult mongoose was found to be in the 4th lumbar segment. On the other hand, the 2nd sacral spinal segment had the shortest vertical and transverse diameter.

The long transverse and vertical diameters in lumbar region, in both groups, may be due to involvement of this region in the formation of the lumbosacral plexus and therefore, its innervations of the hind limb. Also, the small dimensions of the sacral segment, can be due to the limited innervations of this region.

The results of this work about the longest transverse and vertical diameters, were not in agreement with the previous studies in some other species. Previous studies in sheep (Ghazi and Gholami, 1994), goat (Chandna et al., 1998), cat (Mansouri et al., 2002) and camel (Ghazi et al., 1998), found the biggest dimensions of the spinal cord to be in the cervical region. However in rat, dog, guinea pig and rabbit the final lumbar segment has the maximum dimension in the spinal cord (Ghazi et al., 2001, Mansouri et al. 2002, and Farag et al., 2012). The results of this study about the shortest transverse and vertical diameters, were in agreement with the studies on rat and guinea pig (Ghazi et al., 2001).

The present results showed that in both sexes, the longest transverse diameter of the spinal central canal was in the fourth cervical segment, and the shortest in the second sacral segment. This study contradicts the previous studies on guinea pigs and rats about the longest transverse diameter of the central canal. However, concerning the shortest diameter, this study confirms the previous studies (Ghazi et al., 2001 and Farag et al., 2012).

As for the vertical diameter of central canal, our study showed that the maximum diameter of the central canal in male and female mongoose was in fourth and seventh lumbar segment respectively. Also, in male mongoose the 4th cervical segment, and in female the 12th thoracic segment had the shortest vertical diameter of central canal. In rabbit the 12th thoracic and the 2nd sacral, in rat the 2nd sacral and 6th thoracic, in male guinea pig 2nd sacral and 4th cervical, and in female guinea pig spinal segments have the longest and the shortest diameter of central canal respectively (Ghazi et al., 2001).

It seems that there is no consistency between different studies about the maximum and minimum vertical diameter of the central canal.

The present study about the ratio of gray matter to white matter revealed that the

maximum ratio in male and female mongoose was related to 2nd sacral and 7th lumbar segments. In addition, the ratio of gray matter to white matter in lumbar and sacral region was much more than cervical and thoracic regions. The present results were in accordance with previous studies. Also, in rabbit, Guinea pig, Rat (Ghazi et al., 2001) and even in birds such as pigeons (Khaksar et al., 2002) the maximum proportion of gray matter to white matter was found in lumbosacral region. This could be due to the special role of this region in innervation of pelvic limb and showing the potential activity of the lumbosacral area (Masouri.et al., 2008).

According to our study, it can be concluded that the transverse diameter of each spinal cord segment is larger than the vertical diameter. The largest measured diameters and ratio of gray matter to white matter were identified in lumbar and sacral areas.

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مطالعه مقايسهاي هيستومور فومتريك سكمنتهاي مختلف نخاعي در خدنگ نر و ماده بالغ

بابک رسولی^۰ صغرا غلامی گروه علوم پایه، دانشکده دامپزشکی دانشگاه شیراز، شیراز، ایران (دریافت مقاله: ۱۱ مهر ماه ۱۳۹۶، پذیرش نهایی: ۴ دی ماه ۱۳۹۶)

*چکید*ہ

زمینه مطالعه: مطالعات آناتومیکی و بافت شناسی نخاع به دلیل اهمیت بالای آن در زمینههای مختلف دامپزشکی، جانور شناسی و رفتارشناسی همواره مورد توجه متخصصین علوم تشریح بوده است. **هدف:** مطالعه حاضر به منظور ادراک بهتر از جنبههای منظور از ۶ نمونه خدنگ هندی خاکستری که در مراحل پایانی زندگی بر اثر تصادف و یا بیماری در شیراز و نواحی اطراف یافت شدند، استفاده گردید. در ادامه ساختار نخاع شوکی به دقت تشریح گردید، در فرمالین ۱۰٪قرار گرفت و قالبهای پارافینی و سپس برشهای استفاده گردید. در ادامه ساختار نخاع شوکی به دقت تشریح گردید، در فرمالین ۱۰٪قرار گرفت و قالبهای پارافینی و سپس برشهای بافتی با ضخامت μ ۶ از آنها تهیه شد و نمونه ها با رنگ آمیزی هماتو کسلین و ائوزین رنگ آمیزی گردید. در این مطالعه قطرهای عمودی و عرضی، کانال مرکزی، طول ارتفاع سلولهای اپندیم و نسبت ماده خاکستری نخاع به ماده سفید در سگمنتهای منتخب با استفاده از روشهای میکرومتری و میکروسکوپ نوری اندازه گیری شد. **نتایج:** در خدنگ نر وماده بیشترین قطرهای عرضی و مودی سگمنتهای منتخاعی در ناحیه کمری دیده شدند. اگرچه این ثبات و هم خوانی دادهها در قطرهای عرضی و عرضی و مرکزی دیده نمی شود. هم چنین بیشترین ارتفاع سلولهای اپاندیم و نمات و هم خوانی دادهها در قطرهای عرضی و مرکزی دیده نمی شود. هم چنین بیشترین ارتفاع سلولهای اپاندیم در هر دو جنس در ناحیه کمری دیده شد. **نتیجه گیری نه** بر مرکزی دیده نمی شود. هم چنین بیشترین ارتفاع سلولهای اپاندیم در هر دو جنس در ناحیه کمری دیده شد. **نتیجه گیری نه** بر توجه به دادههای بدست آمده این گونه استنتاج گردید که در تمامی سگمنتهای نخاعی قطر عرضی طویل تر از قطر طولی بود و بیشترین ابعاد سگمنتی و هم چنین بزرگترین نسبت ماده خاکستری به ماده سفید در ناحیه کمری دیده شد. که مولی که به دلیل بیشترین ابعاد سگمانها و سوعی برای سانه ماده کردید که در تمامی می ماده مونی در میم می می می و مولی تر از قطر طولی بود و بیشترین ابعاد سگمنتی و هم چنین بزرگترین نسبت ماده خاکستری به ماده سفید در ناحیه کمری – خاجی مشاهده گردید که به دلیل

واژههای کلیدی: مقایسه ای، هیستومور فومتری، خدنگ، سگمنت، نخاع شوکی

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