Morphometric analysis of the neuronal numbers and densiti the inferior olivary complex in the donkey (Equus asinus)

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Abstract

The morphometric interrelations between the compartments of the inferior complex (IOC) in the donkey (Equus asinus) were ascertained by examining sections throughout the entire length of the IOC for both sides. Nissl-stained ce sections of four donkey's brainstems were used. The IOC consisted of three nuclei and four small cell groups. The total neuronal count in both sides of the IO 202,040 + 8,480 cells. The medial accessory olivary nucleus (MAO) had the l relative area (46%) and the highest number of neurons (90,800 + 7,600). The accessory olivary nucleus (DAO) had the second largest relative area (33%), wh principal olivary nucleus (PO) had the lowest relative area (21%). However, th neuron count in the PO was larger (60,840 + 1,840) than DAO (50,360 + 4,040 average neuronal density was 2,700 + 400 cells/ mm3. The numerical values current study of the IOC in the donkey were similar to that of other mammals.

Keywords: *Donkey, Inferior olivary complex, Neuronal number and density*

Introduction

The morphology of the inferior olivary complex (IOC) has been demonstrated in mammals (Kooy, 1916; Kappers et al., 1960; Taber, 1961; Moatamed, 1966; Bre 1967; Schild, 1970; Bowman and King ,1973; Bowman and Sladek, 1973; Martin 1975; Watson and Herron, 1977; Rutherford and Gwyn, 1980; Saigal et al., 1983 Woodward, 1987; Tan et al., 1995; Bozhilova-Pasirova and Ovstcharoff, Bukowska et al., 2002; Rashed et al., 2007), including donkey (Rashed et al., 2 However the neuronal number of the IOC in the donkey still unknown.

In donkey the IOC is divided into three main nuclei; medial, and dorsal acce olivary nuclei (MAO and DAO, respectively) and a principal olivary nucleus (PC four small cell groups; the dorsal cap (DC), the ventro-lateral outgrowth (VLO nucleus β and the dorsal medial cell column (DMCC) (Rashed et al., 2006).

Generally, it is recognized that the IOC is the sole source of the climbing (Szentagothai and Rajkovits, 1959; Desclin, 1974; Brodal et al. 1975; Freedman 1977), and nearly all of the neurons in the IOC are projection neurons t cerebellum (De Zeeuw et al., 1998). A single olivocerebellar fiber projects with m climbing fibers to a single narrow longitudinal band-shaped area in the cere cortex and, with its collateral axons to a small area in the cerebellar nuclei (Sugih al., 1999).

Each climbing fiber innervates a single Purkinje cell (Eccles et al., 1966). olivocerebellar fiber branches into about 4 – 7 (rat), to 14 – 17 (human) and

(chicken) (Rashed et al., 2005). Thus we can estimate the number of Purki direct counting or by the neuronal numbers of the IOC.
In continuing study of the IOC in the donkey, we try to find out mo information about this nuclear complex by morphometrical observation of th number of each main nucleus.

Materials and Methods

Four donkeys (Equus asinus), 2-3 years old, were used in this study. Tl were anesthetized with an over dose of pentobarbital sodium, and then pe physiological saline followed by 10% formalin via the carotid artery. The b were removed; post fixed in formalin for 3 days or more, dehydrated using series of ethanol, and embedded in celloidin. The brain stem was serially transverse plan at 50 mm thick. Serial sections were stained with toluidi cresyl violet. Methods of staining are those quoted from Bancroft ar (1990).

Histology and procedure for neuron count:

The sections were observed under a light microscope at a final magnificati for cell counting. The IOC neurons were counted in every tenth section c right and left sides of each animal. All neurons in a given section wer whether or not a nucleolus could be identified. The total neuronal number of the donkey was then calculated by the method of Escobar et al., (1968) as number of neuron (A) in each section counted was multiplied by half the sections not counted (B) between the section counted and the next count By adding the products of AB/2 for all sections counted, the total number of the IOC of each specimen was obtained. The reliability of this method wa before by Rashed et al., (2005).

Procedure for neuronal density:

The neuronal density was obtained by projecting the microscopic sections o micrometer (VM-29; Olympus, Tokyo, Japan) at a final magnification 90X. T areas of each nucleus and small cell group of the IOC were measured in section on the left side (two cases) and on the right side (one case). The neurons per square millimeter in each counted section was obtained, an number of neurons per cubic millimeter was obtained by multiplying the me of neuron /mm2 by the Escobar coefficient from the following formula:

Escobar coefficient = 1,000 / (section thickness X 2*)

2*: the neuronal count for one section is equal to the neuronal count for two sections (Escobar et al., 1968). In this study the Escobar coefficient = 1,00 = 10

Photos for the nuclei of the IOC were taken at different levels and cropped t processing application, to correct the brightness and contrast and nothing el

Results

Histological findings:

The IOC in donkey was divided into three main nuclei; medial, and dorsal olivary nuclei (MAO and DAO, respectively) and a principal olivary nucleu four small cell groups; the dorsal cap (DC), the ventro-lateral outgrowth nucleus β and the dorsal medial cell column (DMCC).

Neuron number, size and density in the IOC:

The neuron numbers in the IOC of the donkey was estimated (Table 1 differences between the neuron numbers in the right and left sides of the IO non significant. Among the three major nuclei, the MAO had the largest cell r (90,800 + 7600 cells). Although the relative area of the PO was smaller than tha DAO the former had the second largest neuron number (Table 1). Over 500 different levels of each nucleus in the IOC were measured. The average neur (represented by diameter) was 25 µm (Plate I).

The average neuron density of the IOC of the donkey was calculated to be 2700 with its highest value in the PO and its lowest value in the DAO (Table 1).

Discussion

The total numbers of the IOC neurons have been estimated at about 90 1,025,000 or 1,060,000 in humans (Escobar et al., 1968; Moatamed, 1966; Futa Okamoto, 1968), 27,000 in vampire bat (Escobar et al., 1968), 140,000 or 150, cat (Escobar et al., 1968;Mlonyeni, 1973), 49,000 or 57,000 in rat (Delhaye-Bou et al., 1985; Schild, 1970) and 211,000 in water buffalo (Rashed et al., respectively. In the present study, the IOC in the donkey contained 202,000 cells the number of IOC neurons in the donkey showed more or less morphom similarities to that of the water buffalo.

The MAO is the largest nucleus of the IOC in most of the studied mammals exc human in which the PO is the largest nucleus (Moatamed, 1966; Armstrong, Azizi and Woodward, 1987). Previous studies showed that the MAO, DAO a contain neurons at proportions of 10%, 4% and 86%, respectively in hu (Moatamed, 1966), 49%, 24% and 27% or 46%, 25% and 29% in rat (De Bouchaud et al., 1985; Schild, 1970), 47%, 26% and 27% in water buffalo (Ras al., 2007), and 45%, 25% and 30% in this study. Therefore, the IOC in the donke nearly similar to that of rat and water buffalo in the proportions of its three major n Estimates have been made for the packing density of cell within the IOC. estimated as 65,000 cells /mm3 in the vampire bat and carp (Escobar et al., Bozhilova-Pasirova and Ovtscharoff, 2000), 44,000 cell / mm3 in the rat (Esco al., 1968; Schild, 1970), 28,000 cells / mm3 in the pigeon (Bozhilova-Pasirov Ovtscharoff, 2000), 23,000 cells /mm3 in the ground squirrel (Bozhilova-Pasirov Ovtscharoff, 2000), 8,000 ~ 15,000 cells / mm3 in the cat (Escobar et al., Bozhilova-Pasirova and Ovtscharoff, 2000), 5,000 ~ 15,000 cells / mm3 in h (Escobar et al., 1968; Bozhilova-Pasirova and Ovtscharoff, 2000) and 3,000 mm3 in the water buffalo (Rashed et al., 2007). The neuron density in the IOC donkey was 2,700 + 400 cells /mm3. The previous and current studies showed th neuron densities correlate inversely with the body weight. Since the donkey is h than human, but lighter than water buffalo, the neuron density of the donkey IO be lower than that of human and higher than that of water buffalo. Actually the n densities in the IOC of the donkey and water buffalo were nearly the same. This be attributed to the decreased neuron density in the DAO of the donkey which aff the average neuron density for the three major nuclei.

In the present study the average neuron size (represented by diameter) was 2 This reflects the fact that the olivary neurons in the donkey are within the animal (Armstrong, 1974). The study of the neuron size in the donkey did not revea

significant regional differences. This contrasts with the finding that humai were about twice as large as DAO or MAO cells (Moatamed, 1966).

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Figure legends:

Plate I: Photomicrographs of the inferior olivary complex at different levels: pole (a), middle part (b) and caudal pole (c).

Fig. 1: Photomicrographs of medial accessory olive (MAO) showing large neurons in the cranial pole (1a), neurons within the average size and density ran the middle part (1b) and average size range in the caudal pole (1c).

Fig. 2: Photomicrographs of dorsal accessory olive (DAO) showing low ne density as indicated from low neuron numbers within a large area of IOC in the and caudal poles (2a and 2c). The density within the middle part (2b) was comp with the counterparts in the other major nuclei, but with relatively large-sized neu

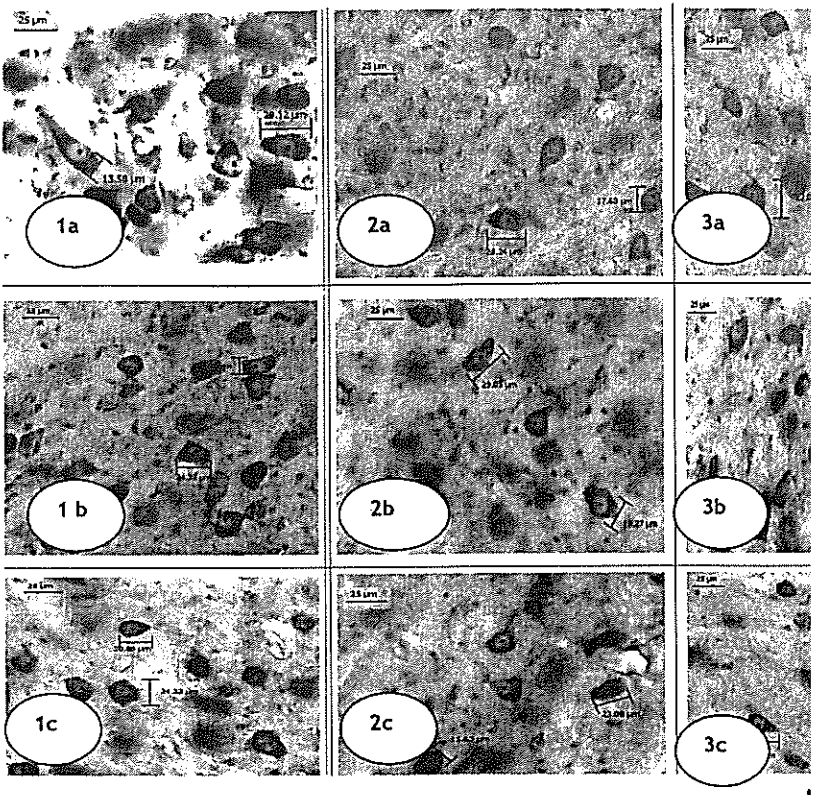
Fig. 3: Photomicrographs of principal olive (PO) showing large-sized cells in the pole (3a), neurons within the average size and density ranges in the middle pa and relatively small neurons within the caudal pole (3c).

Table 1: Numbers and densities of the IOC neurons in the donkey

Major nuclei	MAO		DAO		PO		Total
Sub-nuclei	a, b and c	nucleus b	DMCC	PO	DC	VLO	
Neuron number	83900	4970	1970	50360	53710	1970	5160	
Total + SD		90840 + 7620		50360 + 4040		60840 + 1840	202040 + 8480
Percentage – 1 (%)	42	2	1	25	26.5	1	2.5	100
Percentage – 2 (%)		45		25		30		
CV (%)		8.4		8		3		4
Relative area (%)		46		33		21		100
Neuronal density cells / mm3 + SD		2600 + 600		1900 + 140		3600 + 500		2700 + 400

SD; standard deviation, Percentage -1 ; proportion of the neuronal number in the subnuclei to the total neuronal number of the IOC, Percentage -2; proportion of th neuronal number in the major nuclei to the total neuronal number of the IOC, CV; coefficient of variation, Relative area (%); the proportion of the area of the major to the total area of the IOC in a given number of sections.

Plate I



العربى

رفومترى لعدد و كثافة الخلايا العصبية فى الجسم الزيتونى فى الحُمُر

(١)، رضا راشد(٢)، حسام عطية(٣)
نسجة (١) و التشريح و الأجنة (٢) بكلية الطب البيطرى جامعة المنوفية فرع السادات و قسم
(٣) بكلية الطب البيطرى جامعة بنها

لعلاقة العددية بين أجزاء الجسم الزيتونى فى الحُمُر الأهلية من خلال فحص قطاعات
سلة من جذع المخ- و تحديداً من النخاع المستطيل- مصبوغة بطريقة نيسل بطول الجسم
جهتين اليُمنى و اليُسرى. أظهرت الدراسة أن الجسم الزيتونى يتكون من ثلاثة فصوص
صغرى. و كان المجموع الكلى لأعداد الخلايا العصبية فى الجهتين اليُمنى و اليُسرى
وني (٢٠٢،٠٤٠ ± ٨،٤٨٠) خلية. يمثل الفص الداخلى أكبر مساحة نسبية (٤٦%)
يا (٩٠،٨٠٠ ± ٧،٦٠٠) خلية. ويحتل الفص العلوى المركز الثانى من حيث المساحة
) في حين أن الفص الرئيسى يمثل أقل مساحة نسبية (٢١%) و رغم ذلك يحتوى
د خلايا أكبر (٦٠،٨٤٠ ± ١،٨٤٠) من الفص العلوى (٥٠،٣٦٠ ± ٤،٠٤٠) خلية. و
افة العددية للخلايا العصبية في الجسم الزيتوني (٢٧٠٠± ٤٠٠) خليةامم٣. و قد خلصت
إلى أن القيم العددية للجسم الزيتوني في الحُمُر الأهلية كانت شبيه بمثيلاتها في الثدييات