ABSTRACT

Compared to the rest of the brain, the cerebellum is a small but outstanding structure which is present in all vertebrates. It is a dorsally located metencephalic structure of the rhombencephalon and plays important roles in movements. As in other vertebrates, the cerebellum is expected to be regulating the control of fish movements; related to maintenance of posture and balance, and coordinated movements. Emphasis of the study was on the gross morphology and neurohistology of the cerebellum of selected freshwater fishes. Preliminary studies revealed the existence of different cerebellar orientations in fishes, either rostrally or caudally directed. Four Malaysian freshwater fish species, with two species representing each cerebellar orientation, were looked at as part of an ongoing study in developing Malaysian freshwater fish central nervous system database. *Keli* (*Clarias* sp.) and *baung* (*Mystus nemurus*) represented fish with rostrally oriented cerebellum, while *tilapia* (*Oreochromis* sp.) and *jelawat (Leptobarbus hoeveni)* represented fish with caudally oriented cerebellum. The shape of the body and features of each fish were correlated to the cerebellar orientation. Both *keli* and *baung*, which have rostrally directed cerebellum, demonstrated the same 'torpedo-shaped' body form; while *jelawat* and *tilapia* with caudally directed cerebellum displayed the same compressed body shape. The external morphological features of the cerebellum of these fishes were observed and measured. Each cerebellum exhibited morphological features common to specific cerebellar orientation. For *keli's* and *baung's* rostrally oriented cerebellum, the median-longitudinal sulcus and gyrus were found on their cerebellar surfaces respectively; while for *tilapia's* and *jelawat's* caudally oriented cerebellum, neither sulcus nor gyrus was found. Each fish cerebellum was histologically shown to be made up of mostly cortex area. Stained with H&E and Nissl, the architecture of cerebellar cortex in those fishes appeared similar,

i.e. consisting of molecular layer at the outermost, followed by Purkinje and granular neuronal layers towards the inner area. The granular layer made up the inner region of densely packed stained neuronal somas in cerebellum of these fishes, while the outer region was not intensely stained. Transverse and longitudinal sections of cerebellar tissue of each fish portrayed unique structure of the innermost area. Transverse sections of *keli's*, *baung's* and *jelawat's* cerebellum showed a 'butterfly like' pattern of the innermost area. In contrast, a 'dome like' pattern of innermost area pointing dorsally was observed in *tilapia's* cerebellum transverse sections. Longitudinal cerebellum sections stained with Thionin staining showed fibers in the white matter, which existed within the core portion of the innermost area. These fibers could be mossy and climbing fibers as had been reported in higher vertebrates. It is also possible that cerebellum of fish represents one primitive convolution / folium of higher vertebrate cerebellum. Architectural 3D details of the unique pattern of cerebellar granular layer and possible correlation to physiological cerebellar functions should be further studied.

ABSTRAK

Berbanding dengan bahagian otak yang lain, serebelum ialah suatu struktur kecil tetapi menyerlah yang wujud pada semua vertebrat. Ia merupakan struktur metensefalik yang terletak secara dorsal pada rombensefalon dan memainkan peranan yang penting dalam pergerakan. Seperti pada vertebrat yang lain, serebelum dijangka mengawalatur kawalan gerakan ikan; berkaitan dengan penyelenggaraan postur dan keseimbangan, serta koordinasi gerakan. Penekanan kajian adalah ke atas morfologi luaran dan neurohistologi serebelum ikan air tawar yang terpilih. Kajian-kajian rintis mendedahkan kewujudan orientasi serebelum pada ikan yang berbeza, iaitu sama ada mengunjur ke arah rostral atau kaudal. Empat spesies ikan air tawar Malaysia, dengan dua spesies mewakili setiap orientasi serebelum, telah dipilih sebagai sebahagian daripada kajian yang dijalankan secara berterusan dalam membentuk pengkalan data sistem saraf pusat ikan air tawar Malaysia. Ikan keli (*Clarias* sp.) dan baung (*Mystus nemurus*) mewakili ikan dengan serebelum berorientasi rostral, manakala ikan tilapia merah (*Oreochromis* sp.) dan jelawat (*Leptobarbus hoeveni*) mewakili ikan dengan serebelum berorientasi kaudal. Bentuk badan dan ciri-ciri setiap ikan dikolerasikan dengan orientasi serebelum. Kedua-dua keli dan baung yang mempunyai serebelum mengarah ke rostral memiliki bentuk badan yang sama, iaitu bentuk ‘torpedo’; sementara jelawat dan tilapia yang mempunyai serebelum mengarah ke kaudal memiliki bentuk badan yang sama, iaitu pipih (*compressed*). Ciri-ciri morfologi luaran serebelum ikan-ikan ini telah diamati dan diukur. Setiap serebelum menunjukkan ciri-ciri morfologi lazim bagi orientasi serebelum yang khusus. Bagi serebelum keli dan baung yang berorientasi rostral, sulkus dan girus median-longitudinal telah ditemui pada permukaan serebelum masing-masing; manakala bagi serebelum yang berorientasi kaudal yang terdapat pada tilapia dan jelawat, kedua-dua sulkus atau girus tidak

ditemui. Secara histologi telah ditunjukkan bahawa sebahagian besar setiap serebelum ikan-ikan tersebut terdiri daripada kawasan korteks. Melalui pewarnaan H&E dan Nissl, arkitektur korteks serebelum ikan-ikan tersebut kelihatan serupa, iaitu terdiri daripada lapisan molekul di bahagian paling luar, diikuti oleh lapisan neuron Purkinje dan lapisan neuron granular mengarah ke bahagian paling dalam. Lapisan granular membentuk bahagian dalam serebelum ikan tersebut yang tersusun padat dengan soma neuron yang diwarnakan kuat; manakala bahagian luar tidak diwarnakan dengan kuat. Keratan-keratan rentas dan memanjang tisu serebelum setiap ikan menunjukkan struktur kawasan paling dalamnya yang unik. Keratan-keratan rentas serebelum keli, baung dan jelawat menunjukkan kawasan paling dalam yang berbentuk seperti ‘rama-rama’. Sebaliknya, corak kawasan paling dalam seperti ‘kubah’ yang menunjuk arah dorsal diamati pada keratan rentas serebelum tilapia. Keratan-keratan memanjang serebelum yang diwarnai dengan pewarnaan Thionin menunjukkan gentian dalam jirim putih yang wujud di bahagian tengah kawasan paling dalam. Berkemungkinan gentian-gentian ini adalah *mossy fiber* dan *climbing fiber*, seperti yang telah dilaporkan dalam vertebrat yang lebih tinggi. Berkemungkinan juga serebelum ikan mewakili satu lingkaran primitif / folium serebelum vertebrat yang lebih tinggi. Ciri-ciri 3D bentuk unik lapisan neuron granular dan kaitannya dengan fungsi fisiologi serebelum perlu dikaji dengan lebih lanjut.

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF APPENDICES

LIST OF ABBREVIATIONS

Brain structures

Cer	-	Cerebellum
CCe	-	Corpus cerebellum
EG	-	Eminentia granularis
FL	-	Flocculonodular lobe
GL	-	Granular layer
II	-	Optic nerves
IL	-	Inferior lobe
MO	-	Medulla oblongata
Nflm	-	Nucleus of medial longitudinal fascicles
OpL	-	Optic lobe
P	-	Pituitary
SC	-	Spinal cord
Tel	-	Telencephalon
V3	-	3rd ventricle
V4	-	4th ventricle
VCe	-	Valvula cerebellum

Unit

cm	-	centimeter
g	-	gram
mm	-	millimeter
mL	-	milliliter
µm	-	micrometer

Reagents

H&E	-	Haematoxylin and Eosin
HCl	-	Hydrochloric acid
MS222	-	Tricaine Methanesulfonate
$NaHCO_3$	-	Sodium carbonate
DPX	-	Distrene-80

Others

PASUM	-	Pusat Asasi Sains Universiti Malaya
ISB	-	Institute of Biological Sciences