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### THE ARTERIAL VASCULARIZATION OF THE AMYGDALA IN DOGS

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*We investigated the vascularization of the amygdala of the dog using various contrast agents injected into the internal carotid and vertebral artery. Subsequent clearance of the tissues gives us an opportunity to study the finest capillary network in the amygdala.*

*The corticomedial part of the amygdala has a good vascularization given by the rostral and caudal choroid arteries. The rostral choroid gives off a rich capillary net to the nucleus centralis (NCE), its pars lateralis (NCE<sub>L</sub>) and pars medialis (NCE<sub>M</sub>). Nucleus medialis (NM) is poorly vascularized compared to the central nuclei. The massa intercalata (MI) also has a delicate artery around which there is a fine net of capillaries. Nucleus corticalis (NCO) and area enthorhinalis (AER) are better vascularized by the subpial blood vessel. The caudal choroid, a branch of the caudal cerebral artery is a source of subpial capillaries.*

*The basolateral part of the amygdala is very well vascularized by the rostral choroid artery, a branch of the middle cerebral, especially the nucleus basolateralis (NBL) and nucleus lateralis posterior (NLP). The nucleus basomedialis (NBM) is poorly vascularize by the rostral choroid artery .*

*The amygdala, is served by the middle cerebral artery, via its branch, the rostral choroid, and the caudal cerebral artery via its branch the caudal choroid artery.*

*Every amygdala nucleus has its own central artery around which is formed a rich net of capillaries.*

*Key words: dog, amygdala, arteries, nuclei of the amygdala, massa intercalata, area enthorhinalis.*

### INTRODUCTION

There is a variety of methods used for examining the vascularization of organs and other parts of the body. They include, in the first place, injecting of blood vessels with different substances and then monitoring their course by

making specimens, tissue clearing, X-raying, making casts, etc. (Swindle, 1956; Carleton, *et al.*, 1957; Hughes, 1965; Hildebrand, 1968; Popovic and Champo, *et al.*, 1974; Popovic, 1977; Drekić, 1979).

The arterial system of animals has been studied by many authors. This includes the circulatory system of domestic mammals (Miller *et al.*, 1964;; Ellenberger and Baum 1974; Sisson *et al.*, 1975; Nickel *et al.*, 1981; Janković *et al.*, 1988), the arteries of the small green monkey (Blagojević *et al.*, 1989, 1999, 1998, 2004; Nikolić *et al.*, 2003.), the subclavian artery and its branches in the ground squirrel (Nikolić *et al.*, 2004), the arterial circulation of the limbic structures in the guinea pig (Librizzi *et al.*, 1999) and morphology of the amygdala nuclei of the rat (Drekić *et al.*, 1995a, 1995b, 2002).

The available literature offers very little information on the arteries which supply the amygdala of the dog. Librizzi *et al.* (1999) investigated the vascular territories of the cerebral arteries that originate from the Willis circle to supply limbic structures in the guinea pig brain. Drekić *et al.* (1995a; 1995b) have studied different neuronal cell groups in corticomedial and basolateral amygdala in male and female rats, but did not investigate the vascularization of these parts of the amygdala. This was the reason we undertook these studies of the amygdala nuclei and its arteries in the dog which were then compared with the corresponding arteries in other domestic mammals (Sisson *et al.*, 1975; Janković *et al.*, 1988), small green monkey (Blagojević *et al.*, 2004; Blagojević *et al.*, 1999; Blagojević *et al.*, 1998; Teofilovski, 1982; Stanojević *et al.*, 1983) , guinea pig (Librizzi *et al.*, 1999) and ground squirrel (Nikolić *et al.*, 2004).

#### MATERIALS AND METHODS

A study of the vascularisation of the amygdala in the dog was carried out on 10 adult animals, of both sexes. Two methods were implemented: injection of various contrast agents and Spalteholtz's method. The most frequently used substances for injection into the internal carotid artery, were gelatin-ink, gelatin-minium and latex- ink. Spalteholtz method used for clearing the tissue gives us an opportunity to study a fine net of capillaries in the amygdala.

In order to study the three-dimensional aspect of the amygdala of blood vessels, as well as its relationship to various structures visible on the preparation, specimens about 50  $\mu\text{m}$  thick were made.

The blood vessels of the amygdala were observed in depth by raising and lowering the microscope tube.

A stereo-microscope SM XX and Universal Research Microscope NU 2 Carl Zeiss Jena were used for observing and photographing the specimens.

#### RESULTS AND DISCUSSION

The tissues become fully transparent during the clearing process and their structure is impossible to be recognized. Only the outlines of the preparation, taken as a whole, can be seen. The blood vessels can be examined in depth to a

great extent by raising or lowering the microscope tube (Swindle, 1956; Carleton *et al.*, 1957; Hughes, 1965; Hildebrand, 1968; Champo *et al.*, 1974; Popovic, 1977; Popovic *et al.*, 1979).

The internal carotid and vertebral arteries in the dog, as well as in other domestic mammals (Sisson *et al.*, 1975; Jankovic *et al.*, 1988; Nickel *et al.*, 1981; Baum, 1974), small green monkey (Blagojevic *et al.*, 2004) guinea pig (Popesko, 1992; Librizzi *et al.*, 1999) are the largest sources of blood to the brain of these animals. However, the arterial blood of the ground squirrel (Nikolic *et al.*, 2004) is brought to the brain exclusively by the vertebral artery.

Both internal carotid (*A. carotis interna sinistra et A. carotis interna dextra*) of the dog runs through the pharyngeal space towards the cranial cavity, which they enter after passing through the carotid canals (*canales carotici*) in the petrosa of the temporal bone into the cavernous sinus, where both internal carotides are joined via the *A. intercarotica caudalis*. Inside the cavernous sinus the *Aa. carotides internae* run rostrally, lateral to the hypophysis and then perforate the dura mater as the cerebral carotid artery (*a. carotis cerebralis*). On entering in the subarachnoidal space the vessel trifurcates, as the middle cerebral, rostral cerebral and caudal communicating arteries in the dog, and as opthalmic, middle cerebral and caudal communicating arteries in the small green monkey (Blagojevic, 2004; Teofilovski *et al.*, 1984).

The caudal communicating artery (*a. communicans caudalis*) is the weakest branch of the internal carotid artery in the dog and small green monkey (Blagojevic *et al.*, 2004). It runs aborally and connects the internal carotid and basilar arteries and forms the caudolateral quadrant of the *Circulus arteriosus cerebri* (Nomina Anatomica, 1973). The caudal cerebral artery is the terminal branch of the basilar artery in the dog or one of the branch of the caudal communicating (Jankovic *et al.*, 1991). It runs to the cerebral peduncle crossing its ventral side and than reverses to the dorsal side of the peduncle. It gives off the caudal choroid artery which runs along the choroid plexus towards the interventricular foramen. A rich capillary network is found in the ventral part of the hippocampus, entorhinal area and cortical nuclei (Figure 2A).

The middle cerebral artery (*a. cerebri media*, Figure 2A) is the largest branch of the internal carotid artery which supplies the brain of the dog and other domestic mammals (Miller *et al.*, 1964; Jankovic *et al.*, 1988), rodents (Popesko *et al.*, 1992; Librizzi *et al.*, 1999; Nikolic *et al.*, 2004) and small green monkey (Blagojevic *et al.*, 2004). However, Jankovic, *et al.*, 1991. have described the middle cerebral artery as the branch of the cerebral carotid or communication arteries. The middle cerebral artery in the dog leaves the internal carotid as a terminal branch, about 1mm from the origin of the caudal communication. At its beginning the middle cerebral artery gives off the rostral choroid artery similar to other domestic animals (Sisson *et Grossman*, 1975) and than it lies on the anterior perforated substance. The rostral choroid runs along the optic tract, penetrating the lateral ventricle and supplies its walls and vessels of the chorioid plexus (Figure 1B). The rostral choroid sends branches to the amygdala, gives off central arteries which enter through the nuclei of the amygdala, branching into a fine net of capillaries around the nuclei of the corticomedia and basolateral part of the

amygdala (Figure 2, 3). There is a rich net of capillaries of the corticomедial part of the amygdala about the central nuclei (NCE) and its medial and lateral part (NCE<sub>M</sub> and NCE<sub>L</sub>, Figure 2A) similarly to the guinea pig (Librizzi *et al.*, 1999). The *Massa intercalata* (MI) has a good vascularization, as well (Figure 3B). The medial nuclei (NM) is poorly vascularized, compared to the central nuclei (Figure 2A). The cortical nuclei (NCO) and enthorhinal area (AER) are vascularised by the subpial blood vessels (figure 2A). The caudal choroid artery is a source of subpial capillaries. Basolateral part of the amygdala is very well vascularized, especially basolateral (NBL), posterior lateral (NLP) and basomedial nuclei (NBM, Figure 2A).

The middle cerebral artery then divides into the cortical and central branches. The terminal cortical branches (rami corticales), in the form of minute twigs, enter the cortex of the lateral surface of the cerebral hemisphere and richly supply it. The central branches (rami centrales) leave the middle cerebral artery near its origin in the form of a large number of small branches which pass through the "perforate substantia" and supply the basal nuclei and adjacent tracts. The amygdaloid body is customarily listed with the basal nuclei for morphological reasons. On the basis of its connections it may be included among the structures of the rhinencephalon (Drekic, 1983).

The caudal cerebral artery of the dog is the terminal branch of the basilar artery, but in some cases it is a branch of the caudal communication (Jankovic *et al.*, 1991). It runs to the cerebral peduncle crossing its ventral side and then reverses to the dorsal side of the peduncle. In this spot it gives off the caudal choroid artery (*a. choroidea caudalis*) which runs along the choroid plexus towards the interventricular foramen. A rich capillary network is found in the ventral part of the hippocampus, enthorhinal area and cortical nuclei (Figure 2A). Ventral, temporal, part of the hippocampus in the guinea pig (Librizzi *et al.*, 1999) is served exclusively by the rostral posterior cerebral artery similarly to other rodents (Popesko, 1992).

According to our investigations, as well as results of other authors (Librizzi *et al.*, 1999; Drekic *et al.*, 2002; Nikolic *et al.*, 2004) the main suppliers of blood to the basolateral and corticomедial part of the amygdala, except NCO, in the dog is the rostral choroid artery and the branches of the middle cerebral artery. However, the cortical nuclei, hippocampus and enthorhinal area are served exclusively by the caudal choroid, a branch of the caudal cerebral artery.

The rostral cerebral artery (*a. cerebri rostralis*) originates directly from the internal carotid artery in the dog and other domestic mammals (Nomina Anatomica Veterinaria, 1973), but in the small green monkey it represents the continuation of the internal carotid artery (Blagojevic *et al.*, 2004). It arises lateral to the optic nerve and runs dorsal to the optic nerve. On reaching the longitudinal fissure it unites with its fellow, after which the two vessels separate. In some specimens the rostral communicating artery (*a. communicans rostralis*) connects both rostral cerebral arteries (*a. cerebri rostralis dextra et sinistra*) just rostral to the optic chiasma (Nomina Anatomica Veterinaria, 1973). The rostral cerebral artery forms the rostral quadrant of the *Circulus arteriosus cerebri* (Miller *et al.*, 1964). The rostral cerebral artery sends also central branches (*rami centrales*) into

the medullary substance of the hemisphere in the dog, mammals (Nomina Anatomica Veterinaria, 1973) and small green monkey (Blagojević *et al.*, 2004).

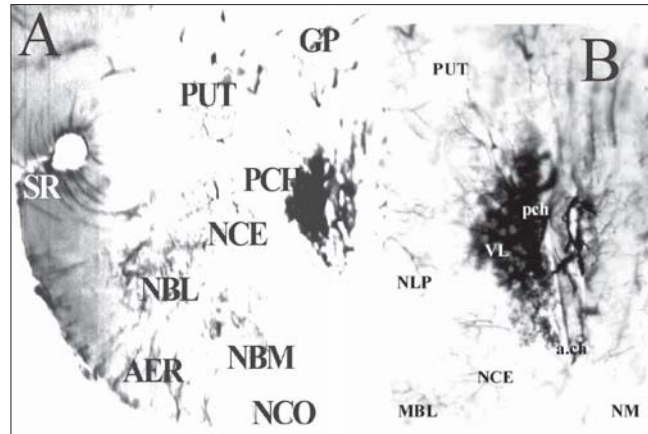


Figure 1. Rostral part of the amygdala nuclei of the dog investigated with gelatin-ink method

Panel A. NCO (cortical nuclei), NBM (basomedial nuclei), NBL (basolateral nuclei), NCE (central nuclei), AER (enthorhinal area), pch (choroid plexus), PUT (putamen), GP (palidal globus), SR (rhinal sulcus) X 120

Panel B. NM (medial nuclei), NLP (lateral posterior nuclei), a. ch (choroide artery), VL (lateral ventricul) X 300

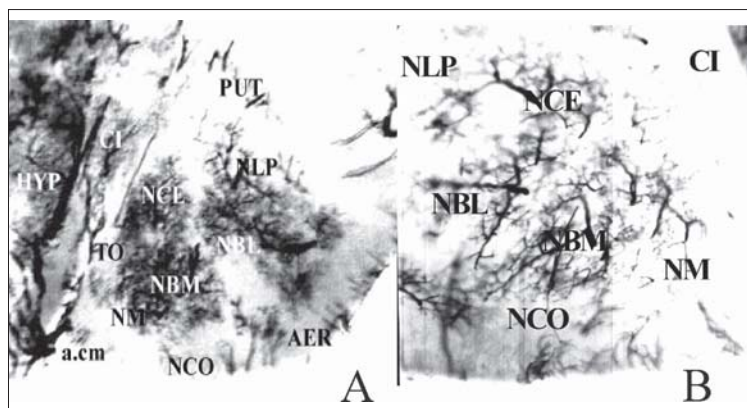


Figure 2. Coronal section of the amygdala dog investigated with gelatin-ink method

Panel A. a. cm (the middle cerebral artery), TO (optic tract), CI (internal capsula), HYP (hypothalamus), NCO (cortical nuclei), NBM (basomedial nuclei), NBL (basolateral nuclei), NCE (central nuclei), AER (enthorhinal area), PUT (putamen) X 300

Panel B. NCO (cortical nuclei), NBM (basomedial nuclei), NBL (basolateral nuclei), NCE (central nuclei)

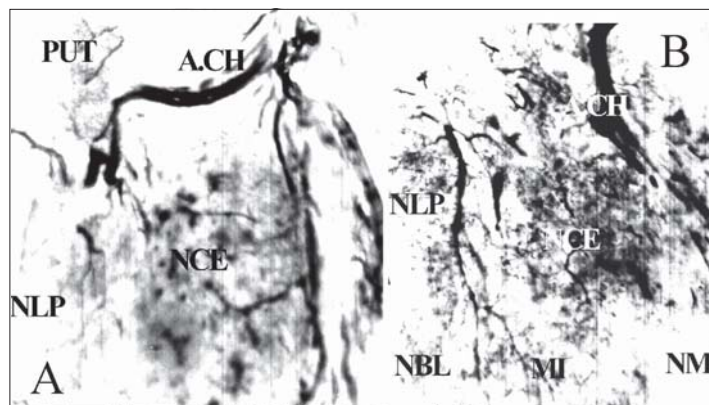


Figure 3. Coronal section of the central nuclei of the amygdala of the dog investigated with gelatin-ink method  
Panel A. NCE (central nuclei), NLP (lateral posterior nuclei), PUT (putamen), A.CH (rostral choroid artery), X 300  
Panel B. NCE (central nuclei), NLP (lateral posterior nuclei), A. CH (rostral choroid artery) X 300

The internal ophthalmic and internal ethmoidal arteries are smaller arteries which leave the rostral cerebral artery in the dog and other domestic mammals (Nomina Anatomica Veterinaria 1973). However, Jankovic *et al.* (1991) have designated the rostral choroid and internal ophthalmic arteries as the branches of the rostral cerebral artery in the dog and other domestic mammals.

The amygdaloid body of the rat was previously referred to in the description of the basal nuclei (Drekic, 1983). Several nuclei of the amygdaloid complex of the rat receive fibers from the olfactory bulbs and the anterior commissure (Drekic, 1983). There are also connections with the septal area by way of the diagonal band. The main efferent pathway is a clearly defined fiber bundle, the stria terminalis.

The stria terminalis in the dog and rat (Drekic, 1983) arises from the rostral part of the amygdaloid body and runs dorsal and caudal along the optic tract to the tail of the caudate nucleus. From there the stria terminalis runs rostrally and ventrally in a groove between the caudate nucleus and the thalamus and ends at the base of the brain in the preoptic and hypothalamic region. These regions are supplied with blood by the middle cerebral artery.

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**ARTERIJSKA VASKULARIZACIJA AMIGDALE PSA**

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## SADRŽAJ

Proučavanje vaskularizacije amigdale psa vršeno je upotrebom različitih kontrastnih sredstava injiciranih u *a. carotis interna* i *a. vertebralis* a prosvetljavanje tkiva daje mogućnost da se prouči i najfinija vaskularna mreža u amigdali.

Kortikomedijalni deo amigdale vaskularišu *a. choroidea rostralis* i *a. choroidea caudalis*. *A. choroidea rostralis*, daje bogatu mrežu kapilara u NCE (*nucleus centralis*) i njegovom lateralnom i medijalnom delu (NCE<sub>L</sub> i NCE<sub>M</sub>). *Nucleus medialis* (NME) je slabije vaskularisan u poredjenju sa vaskularizacijom NCE. *Massa intercalata* (MI) je takodje snabdevena bogatom mrežom kapilara. *A. choroidea caudalis*, kao subpialna arterija, vaskulariše NCO (*nucleus corticalis*) i AER (*area enthorhinalis*).

Bazolateralni deo amigdale vaskulariše samo *a. choroid rostralis*. Bogata mreža kapilara uočava se u NBL (*nucleus basolateralis*) i NLP (*nucleus lateralis posterior*) a nešto siromašnija u NBM (*nucleus basomedialis*).

Krv u amigdalu dovode dve arterije: *a. choroidea rostralis*, grana od *a. cerebri media*, i *a. choroidea caudalis*, grana od *a. cerebri caudalis*.

Svaki nukleus amigdale ima svoju centralnu arteriju i oko nje jako razvijenu mrežu kapilara.