Histological Atlas and Morphological Features by Nissl Staining in the Amygdaloid Complex of the Horse, Cow and Pig

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There are no reports about the amygdaloid complex in horses, cows and pigs. The amygdaloid complex is divided into many subnuclei from the point of histology, and the purpose of this study was to clarify its features by comparison among horses, cows and pigs. Using the Nissl staining method, we examined coronal serial sections of brains from the three species. The division of the amygdaloid subnuclei was performed on the magnified photos and three-dimensional analysis was also performed for the three species. The shape of the amygdala in the horse and cow was an anterior ellipsoid, long and slender in the dorso-ventral direction, posteriorly. In the pig, it was long and slender in the dorso-ventral direction as a whole. The volume of the amygdala in the horse as a percentage of cerebral volume was the biggest among the three species. We divided the amygdala into 27 subnuclei and these subnuclei were classified into 11 subnuclei groups. Features of some subnuclei resembled those of other species, such as rabbit, cat and monkey. Taking into consideration brain development, it was clear that the shapes of the subnuclei were fairly reflected by the brain development within the skull. From the results of three-dimensional analysis, the amygdala was mainly occupied by the lateral and basal nuclei, each occupying about 30% of the amygdala. Features of each species were that the relative size of the posterior cortical nucleus was high in the horse and cow, and the relative size of the amygdalohippocampal area was high in the pig. We concluded that the features of the subnuclei may be related to the functional development.

Key words: amygdaloid complex, histological division, subnucleus, three-dimensional analysis

The amygdaloid complex in primates occupies a central position in the anterior temporal lobe, lying posterior to the temporal pole, anterior to the hippocampal formation and entorhinal cortex, and inferior to the striatum at the level of the decussation of the anterior commissure [26]. A part of the limbic system, it exerts an integratory and regulatory influence on certain basic functions of the organism such as food intake, sexual reactions, defense against danger and emotional expression [18, 19, 27]. Physiological studies in experimental animals suggest that the amygdala integrates sensory stimuli from the environment to modulate autonomic [14, 16] or neuro-endocrine [11, 32] functions, and emotional [33], social [20] and feeding behaviors [6]. The amygdaloid complex also plays an important role in modulating the secretion of gonadotropins [36].

The amygdala has common amygdaloid subnuclei among marsupials, rodents, insectivores, carnivores and primates. Uchida [34, 35] reported on the dividing of the amygdaloid complex into numerous small subdivisions in the mouse, rat, rabbit and pig. Comparative and neuropathological studies of the amygdaloid complex are hampered by the lack of a consensus on the terminology used to describe its subdivisions, both in humans and other animal species [28]. Most anatomical classifications are based upon

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cytoarchitectural differences as seen in Nissl stains [31], since Nissl staining shows Nissl body, nucleus, nuclear membrane and nucleolus. The Nissl method is frequently utilized in various studies that investigate the morphology of neurons, and their number and distribution. Crosby and Humphrey [9] presented a simpler scheme based largely on Johnston’s early classification, which was derived from comparative studies in a variety of species.

Anatomically, the amygdala is a complex and heterogeneous structure. Based on its fiber connections, the basolateral amygdaloid complex is directly connected with many cortical structures and receives visual, auditory, somatosensory and gustatory input. Because it also has input from the limbic system, it is related to the synthesis of learning and memory. The central nucleus receives the largest intra-amygdaloid projection, mainly from the lateral, basal and accessory basal nuclei [3]. Sensory stimuli from the environment reach the centromedial amygdala. The cortical nucleus receive primary afferents from the olfactory bulb, and the central nucleus have hypothalamic input with relay to autonomic and endocrine centers in the hypothalamus and brainstem [4, 28]. It is reported that the lateral nucleus, which is larger than the medial nucleus in higher mammals, appears later in development with the capacity to receive this information directly [29].

Many studies of the amygdaloid subnuclei have been reported for marsupials, rodents, insectivores, chiroptera, carnivores and primates. Although it is clear that the amygdaloid complex is a center for the emotional behavior in the horse, there are few reports on the amygdala in ungulates. A comparative anatomical study through observation of various animals provides the difference existing in the construction of the nuclear groups. The purpose of the present study was to divide the amygdaloid complex of the horse, cow and pig, and to compare the sizes of these subnuclei against the whole amygdaloid complex among the three species. It is possible that characterizations of these species’ amygdaloid subnuclei may help to further understanding of the function of the amygdala in each of these three animals.

Materials and Methods

Animals
Two Thoroughbred horses (2-year-old female, body weight, 450 kg; and a 7-year-old female, body weight, 480 kg) were obtained from the Equine Research Institute, Japan Racing Association (JRA). Two Holstein cows (adult female, body weight, 120 kg) were obtained from the farm attached to Utsunomiya University and the National Institute of Livestock and Grassland Science. Two Landrace pigs (adult female, body weight, 100 kg) were obtained from the Tochigi Prefectural Institute of Animal Industry.

Nissl preparation
The animals were deeply anesthetized with sodium pentobarbital (30 mg/kg) and exsanguinated by section of the arteriae carotis communis. Then, the brain and the attached structures were carefully removed from the skull. Brains were fixed in 10% formaldehyde for about one month and then meninges and blood vessels were peeled off. The right hemisphere was preserved in 10% formaldehyde containing 10% sucrose for 1 week. It was then cut into 100 µm serial coronal sections with a large-sized freezing microtome (YAMATO Co.). The cutting line was the line crossing the ventral aspect of brain at right angles. Sections were put on gelatin coated large glass slides (size: 90 mm × 65 mm), and after fat was removed by the chloroform, tissues were stained for Nissl substance with 1% cresyl violet, differentiated in water, dehydrated, and finally coverslipped (cover glass size: 80 mm × 55 mm).

Observation
Histological observation was done under a light microscope. However, the amygdaloid complex of these animals is too big to observe under a standard microscope. So we had to take photos of all specimens directly to globally observe the amygdaloid complex.

Division of the amygdaloid subnuclei and data analysis
Tracing paper was put upon the photo of the amygdaloid complex, and the outline of the amygdaloid complex was traced on the tracing paper. The division of the amygdaloid subnuclei was based on size, density, and characterization of staining of neurons constituting the amygdaloid complex. The amygdaloid complex had many subnuclei. Many
subnuclei were allocated to some subnuclei groups, based on their location and densely. The divisions and names of the amygdaloid subnuclei were made according to the rat atlas on which advanced studies have already taken place [13]. The boundaries of these subnuclei and the amygdala were input into three-dimensional analysis software (TRI system: RATOC Systems) and analyzed statistically. Unpaired $t$-tests were used to compare the sizes of the subnuclei against the whole amygdala in each animal. The total shape of the amygdaloid complex, the volumes of the amygdaloid subnuclei and their locations relative to the structure of the cerebrum were analyzed in each animal by the reconstruction system.

**Results**

*Morphological features of the amygdala by three-dimensional analysis*

The amygdala was located from the middle part of the brain to the anterior part of it, the rostro-cordal distance of the amygdala was 14.8 mm in the horse, 14.0 mm in the cow and 12.8 mm in the pig. The dorso-ventral distance of the amygdala was 20.0 mm in the horse, 19.8 mm in the cow and 15.5 mm in the pig. The shape of the amygdala of the horse and cow was ellipsoid in the anterior amygdala and was long and slender in the dorso-ventral direction in the posterior amygdala (Fig. 1e, 1f). The amygdala of the pig was generally long and slender in the dorso-ventral direction (Fig. 1g). The results of three-dimensional analysis showed that the volume of the horse amygdala was $5.69 \times 10^2$ mm$^3$, that of the cow was $3.96 \times 10^2$ mm$^3$ and that of the pig was $1.35 \times 10^2$ mm$^3$. These volumes of the amygdala expressed as percentages of the whole brain volume were 0.020% in the horse, 0.016% in the cow and 0.015% in the pig (Table 1).

*Division of the amygdaloid subnuclei*

On the division of the amygdaloid subnuclei, there was no remarkable difference among the horse, cow and pig. There were 27 subnuclei in common among these three species. Eleven subnuclei groups were classified as follows according to the location and densely of the subnuclei: anterior amygdaloid area (AA), intercalated nuclei amygdala (I), amygdalopiriform transition area (APir), amygdalostriatal transition area (ASTr), bed nucleus, striatum, intraamygdaloid nucleus, division (BSTIA), basal amygdaloid nuclei (B), central amygdaloid nuclei (Ce), lateral amygdaloid nuclei (La), medial amygdaloid nuclei (Me), posterior cortical amygdaloid nuclei (P) and amygdalohippocampal areas (AHi) (Table 2).

*Histological features of amygdaloid subnuclei*

The histological atlas of the horse, cow and pig amygdala is shown in Figs. 2, 3 and 4. They show the relationships among nuclei observed in the amygdaloid subnuclei groups and histological features of the subnuclei. In Fig. 5, a comparison of the relative size of each subnucleus group compared to the total amygdala is shown for the horse, cow and pig.

1. The Anterior Amygdaloid area (AA) was located at the rostral edge of the amygdala. It was attached on the dorsal side to the cortex medialis lobus piriform and attached on the ventral side to the nucleus anterocortex. The boundary of this subnucleus was slightly indistinct and it was observed that the size of it was larger toward the cordal side (data not shown). AA of the horse and cow were about 2.5% of total amygdala volume and about 1.4% for the pig (Fig. 5).

2. The rostral edge of the Intercalated Nuclei Amygdala (I) was located at the cordal part of the anterior amygdaloid area (AA) and also at the border with the capsula externa. The dorsal part of this subnucleus was surrounded by central (Ce) and basal nuclei (B) (Figs. 2b, 3b, 4a). This subnucleus was located in the center of the amygdala and was composed of dense, small neurons (data not shown). This subnucleus occupied less than 1% of the total amygdala volume in all three species. This subnucleus was the smallest of the subnuclei groups (Fig. 5).

3. The Amygdalopiriform Transition Area (APir) was located along the rostro-coronal direction at the dorsal part of the amygdala. It was attached to lateral nuclei (La) on the ventro-lateral side and to central nuclei (Ce) on the ventro-medial side. The boundary of the ventral part of this subnucleus was distinct (Figs. 2d, 3c,
Reconstruction of the amygdala by three-dimensional analysis. (a) The slice line in the horse, is at right angles to orbita-auris line. The orbita-auris line connect the margo infraorbitals and porus auris externa, and the zero-point shows the porus auris externa. (b–d) Nissl-stained micrographs at the mid-points of the amygdala in the horse (b), cow (c) and pig (d). Scale: bar=1 cm. (e–g) Three-dimensional figure of the outward appearance of the amygdala from all the sections made in this study constructed by three-dimensional analysis software. The areas enclosed by rectangles in the figures shows the image of one section. Amygdala of the horse (e) and cow (f) diverge in two directions in the cordal region; one is dorsal part and the other is ventral part. The amygdala of the pig does not separate in the cordal region (g). Scale: bar=5 mm.
4c). It was attached to the medio-cordal part of the putamen on the dorsal side, the boundary of this side was not distinct (Figs. 3d, 4c). This subnucleus occupied 4.3% of the total amygdala volume in the horse. In the cow and pig, it occupied around 1.5% of the amygdala volume. The relative size of this subnucleus in the horse was larger than that in the cow and pig (Fig. 5).

5. The Bed Nucleus Striatum, Intra-Amygdaloid Nucleus, Division (BSTIA) was located inside the amygdala from the anterior to medial part. It was attached to central nuclei (Ce) on the dorsal side, and to basal nuclei (B) on the ventro-lateral side and to medial nuclei (Me) on the ventro-medial side (Figs. 2d, 4c). Although the relative size of this subnucleus in the horse and pig was 2.3% and 3.8%, it was much smaller in the cow, less than 1% (Fig. 5).

6. The Basal Amygdaloid Nuclei (B) were widely located from the anterior to posterior of the medial part of the amygdala. As it contains the most dense neurons and the largest neurons of all the subnuclei, it was easy to identify this subnucleus and the boundary of it (data not shown). It was attached to lateral nuclei (La) on the dorsal side (Figs. 2d, 3c, 4c), to postero-cortical nuclei on the rostro-ventral side and to amygdalohippocampal area (AHi) on the cordal-ventral side (Figs. 2d, 2e, 3b, 3c, 4c, 4d). It also was attached to the piriform cortex on the lateral side and to medial nuclei (Me) on the medial side. This subnucleus could be subdivided into two groups, basolateral (BL) and basomedial nuclei (BM), and the former could be further subdivided into three parts, the anterior (BLA), posterior (BLP) and ventral nuclei (BLV), and the latter into three parts, i.e. (BM), the anterior (BMA) and posterior nuclei (BMP) (Table 2). The cell density of the lateral side of BLA or BLP was higher than the medial area in the cow and pig (data not shown). In the horse, the cell density of this subnucleus was almost uniform (data not shown). This subnucleus of the horse and pig occupied around 30% of the amygdala volume, making it the largest of the amygdaloid subnucleus. In the cow, it was 26% of the amygdala volume, making it the largest subnucleus after the lateral nuclei (La) (Fig. 5).

7. The Central Amygdaloid Nuclei (Ce) were located on the dorsal side, from the anterior to the medial part of the amygdala. It was attached to amygdalostral transitional area (ASTr) on the dorsal side, to basal nuclei (B) on the ventral side, to lateral nuclei (La) on the lateral side and to nucleus caudatus on the medial side. This subnucleus could be further subdivided into

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<th>Table 1. Average volumes of the amygdala and their proportion as a percentage of the cerebrum</th>
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<th>Table 2. Proposed nomenclature for the horse, cow and pig amygdaloid nuclei</th>
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<td>BSTIA bed nucleus striatum, intraamygdaloid nucleus, division</td>
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Fig. 3. Higher magnified photographs outlined in Fig. 1c and schema of the cow amygdala. (a–f) show the amygdala from the anterior to posterior sections. The distances in the schema show the distance from the front point of the amygdala. For abbreviations, see Table 2. Scale: (a) bar=5 mm.

Fig. 2. Higher magnified photographs outlined in Fig. 1b and schema of the horse amygdala. (a–h) show the amygdala from the anterior to posterior sections. The distances in the schema show the distance from the front point of the amygdala. For abbreviations, see Table 2. Scale: (a) bar=5 mm. (i) Nuclei of the area around the amygdaloid complex of (f). cc: cerebral cortex, CPu: caudate putamen, ec: external capsule, Hip: hippocampus, ic: internal capsule, opt: optic tract, Pir: piriform cortex, Thal: thalamus.
two divisions: lateral (CeL) and medial nuclei (CeM). CeL could be further subdivided into a capsular region (CeLC) and a central region (CeLCn) (Table 2). Although it had a relative size of 4 to 5% in the horse and cow, it was smaller in the pig, 2.7% (Fig. 5).

8. The Lateral Amygdaloid Nuclei (La) were located on the dorsal side, from the anterior to the posterior part of the amygdala. It was attached to the cordolateral part of the putamen on the dorsal side, to basal nuclei (B) on the ventral side, to piriform cortex on the

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**Fig. 4.** Higher magnified photographs outlined in Fig. 1d and schema of the pig amygdala. (a–f) show the amygdala from the anterior to posterior sections. The distances in the schema show the distance from the front point of the amygdala. For abbreviations, see Table 2. Scale: (a) bar=5 mm.
lateral side, to central nuclei (Ce) on the rostro-medial side and to the ventriculus lateralis on the cordal side (Figs. 2d, 2h, 3c, 3e, 4c, 4e). This subnucleus could be subdivided into lateral (LaDL) and ventral nuclei (LaVM) (Table 2). The relative size of this subnucleus in the horse was around 34% making it the largest of all the subnuclei in this animal. The relative size in the cow and pig was around 23%, smaller than the horse.

9. The Medial Amygdaloid Nuclei (Me) were located on the medio-ventral side of the amygdala from the anterior to medial part. This subnucleus was attached to bed nucleus striatum, intra-amgdaloid nucleus, division (BSTIA) on the dorsal side, to postero-cortical nuclei (P) on the ventral side, to basal nuclei (B) on the lateral side and to the optic tructus on the medial side (Figs. 2d, 3c, 4c). This subnucleus could be subdivided into two nuclei, anterior dorsal (MeAD) and posterior nuclei (MeP). The latter could also be subdivided into posteroventral nuclei (MePV) (Table 2). Although the relative size of this subnucleus in the horse and cow was 3.8 and 4.0% of the total amygdala, its relative size in the pig was 7.0%, around 1.8 times larger than the other two species (Fig. 5).

10. The Posterior Cortical Amygdaloid Nuclei (P) were located at the bottom of the amygdala from the medial to posterior part. This subnucleus was attached to basal nuclei (B) on the dorsal side of the anterior part and to the amygdalohippocampal areas (AHi) on the posterior side (Figs. 2d, 2g, 3b, 3e, 4c, 4e). This subnucleus could be subdivided into posterolateral (PLCo) and posteromedial nuclei (PMCo) (Table 2). Although the relative size of this subnucleus in the horse and cow was around 10% of the total amygdala, its relative size in the pig was 6.5% (Fig. 5).

11. The Amygdalohippocampal Areas (AHi) were located on the ventral side of the amygdala from the medial to posterior part. This subnucleus was attached to basal nuclei (B) on the dorsal side, to postero-cortical nuclei (P) on the ventral side and to piriform cortex on the lateral side (Figs. 2e, 3d, 4d). This subnucleus could be subdivided into anterolateral (AHiAL) and posteromedial nuclei (AHiPM) (Table 2). Although the relative size of this subnucleus in the horse was only 5% of the total amygdala, its relative size in the cow and pig was around 12% (Fig. 5).

Three-dimensional analysis of amygdaloid subnuclei

Figure 5 shows the relative sizes of the 11 subnuclei as percentages of the total amygdala in the horse, cow and pig, as computed by three-dimensional analysis. Lateral (La) and basal nuclei (B) were the largest subnuclei in all three species. As for other features, the anterior amygdaloid area (AA) and amygdalostriatial transition area (ASTr) in the horse was comparatively large. ASTR of the horse was significantly larger than those of the cow and pig (p<0.05). The relative sizes of central nuclei (Ce), posterior cortical nuclei (P) and the amygdalohippocampal areas (AHi) in the cow, and medial nuclei (Me), amygdalopiriform transition area (APir) and amygdalohippocampal area (AHi) in the pig were large compared to the horse. AHi of the cow and pig were significantly larger than that of the horse (p<0.05). APir in the pig was significantly large compared to the horse, and BSTIA in the pig was...
significantly large compared to the cow (p<0.05).

Discussion

In this study, the amygdala of the horse, cow and pig was divided into 27 subnuclei and classified into 11 amygdaloid groups. The functions of some subnuclei have already been reported but those of others are still not known. In the 1950’s, Papez [27] and MacLean [19] defined the emotional circle, the amygdaloid complex has intervened in neocortex or lower nucleus of cortex by anatomically and functionally. It is reported that the amygdaloid complex, which is a part of limbic system, has an important role in the synthesis and regulation of the basical functions of numerous adaptive behaviors, such as feeding behavior, sexual desire, avoidance and the defense reaction. These results defined the amygdala can be divided into two parts, the dorso-medial and baso-lateral parts [12]. The former includes the anterior amygdaloid area (AA), central (Ce) and medial nuclei (Me), and the latter includes lateral (La), basal (B) and posterior cortical nuclei (P). Crosby and Humphrey [10] showed that the general pattern of nuclear organization of the amygdaloid complex is more similar among the various mammalian groups than the various nomenclatures would suggest. It is clear from many studies that this general classification is common from lower to higher animals, and it also corresponds to the phylogeny [12, 17, 26, 29, 30]. We classified many subnuclei, which constitute the amygdala, according to their histological characterization and their correlation with subnuclei groups and their functions.

**Basal Amygdaloid Nuclei (B).** These nuclei are distinct in the horse, cow and pig, well developed, as well as the lateral nuclei (La). B have been divided, in many mammals, into a large-celled lateral part lying close to lateral nuclei (La) and a small-celled part which is frequently in the medial position [10, 30]. The features of B were common to the three species and the basis of the division of the amygdala. Further, amygdalo-striate projections originate in these nuclei, and it plays a role in making up behaviors in accordance with the state of emotion or motivation [8]. In the horse and pig, this nuclei is the biggest subnuclei, therefore, it may have an important role in the behavior of these species and the activity by way of lateral nuclei (La).

**Central Amygdaloid Nuclei (Ce).** These nuclei of the three species were adjacent to the putamen and lateral nuclei (La). The parcellation of Ce into large cells at the medial portion and small cells at the lateral portion was observed. In the monkey, the medial subdivision projects to the ventromedial and caudal lateral nuclei of the hypothalamus, whereas the lateral subdivision projects to the lateral mammillary nucleus [4, 30]. Furthermore, Ce in the rat, makes a gradual transition to bed nucleus of stria terminals and the anterior amygdaloid area [7]. Ce is embryologically the oldest of the nuclear groups of the amygdala, and it is related to some neurotransmitters and hormones [20]. Therefore, the similarity of its formation will be common in higher animal including the animals that we observed.

**Lateral Amygdaloid Nuclei (La).** These nuclei in the three species were positioned medial to the external capsule and claustrum, lateral to the putamen and central nuclei (Ce), and more posteriorly to the ventricle, makes them easy to identify. These features resemble those reported in the rat [7]. In addition, it has also been reported that La can be identified as dorsal and ventral parts, which extend further backwards in the rabbit as well as in the rat [7]. La is a major site of sensory convergence in the amygdala, as it receive widespread afferents from the sensory association cortex, and it also projects back into the cortex [2, 24]. Further, La has connections with many limbic structures via the other nuclei of the amygdala [28]. In the present study, the relative size of La was not different among the three species, therefore, the role of La will be common.

**Medial Amygdaloid Nuclei (Me).** In the horse, cow and pig, the anterior extremity is indistinctly outlined, since its cells gradually fuse with those of the anterior amygdaloid area (AA), and the posterior is elongated in the posterior direction, establishing contact with the subiculum. These features resemble those reported in the rat [7]. Me is older phylogenetically, and is related to sense of smell. With a decrease in the size of the olfactory system in primates, Me has become less well differentiated and, although present, is not conspicuous in humans [10]. Generally, domestic animals as such as the cow, pig and horse have well developed olfaction. There is a considerable relationship of the olfactory function in these animals with Me.

**Posterior Cortical Amygdaloid Nuclei (P).** In the three species, P is on the side of the basal forebrain which is surrounded by the lateral piriform cortex, hippocampus and medial nuclei (Me). It is reported
that P is well developed in all mammals. It disappears anteriorly at the level of P in the lateral olfactory tract. Posteriorly it enlarges and its most posterior part is found as a rounded [7].

P of almost mammals resembles the cortex in its cellular structure [1, 28, 30]. It could be divided two subnuclei, lateral (PLCo) and medial (PMCo), in the three species of the present study. Detailed cytoarchitectural reports in lower mammals suggest partition into the anterior cortical nucleus, periamygdaloid cortex and the posterior cortical nucleus. However, the change in positional terminology among species reflects an increased periaxial rotation of the amygdaloid complex in human. The lack of complexity in human cortical nuclei may reflect a relative decrease in the functional importance of olfaction and a consequent simplification of this nuclear group in higher mammals [30]. Crosby and Humphrey [10] reported that P appears to have a subcortical position, but subcortical portions are not present in carnivores. Secondary subdivision occurs in the caudal two-thirds of the cortical nucleus in humans. Taken together, these reports support the idea that P of the three species divide into two subnuclei, since they are higher mammals and herbivores or omnivorous animals. The development of the olfactory function in the pig may lead to enrich the piriform cortex, that is observed as a part of cortical nucleus in the horse and cow. Therefore, it may show that the relative size of P is large in the horse and cow, but small in the pig.

Amygdalo-hippocampal Areas (AHi). The amygdala plays important roles in motivation, memory and visual recognition, and also play a role in the processing of the hippocampal formation. The amygdaloid complex appears to be particularly important for learning associations of stimuli in different modalities [21, 25]. There are few reports about AHi. In the present study, the relative size of AHi in the horse was lower than those of the cow and pig suggesting that the horse hippocampus may be well developed and that its function may be higher and more independent than those of the cow and pig. Therefore, the relative size of AHi in the horse might be decreased due to hippocampal development.

The relation of evolution and function between amygdaloid subnuclei.

As for the distribution of the amygdaloid subnuclei, it is considered that the shape of the amygdala is also important. The olfactory, especially the lateral olfactory tract, is well developed in the pig compared with the horse and cow, and the relative size of the medial nuclei (Me) was two times larger than the other species. It is reported that the efferent fibers from the lateral olfactory tract to the amygdala terminate in the medial nuclei (Me) [8], so there may be a close relation between sense of smell and the medial nuclei (Me). In the present study, cortical nuclei (P) were located in the ventral and the ventromedial parts of the amygdala, and lateral nuclei (La) and basal nuclei (B) were the biggest subnucleus in the amygdala. These results correspond to the reported evolution followed by primates.

Relationship between the development of the amygdala and its functions.

There are many experimental reports about the functions of the amygdaloid subnuclei, especially the larger subnuclei, such as the basal (B), medial (Me) and lateral nuclei (La). There are also experimental reports that cortico-medial nuclei are closely related to the autonomic or endocrine centers in the hypothalamus and brainstem [11, 36]. Baso-lateral nuclei appeared later in development and have an important role in the creation of emotions from sensory and limbic inputs [5, 23]. In the present study, basal (B) and lateral nuclei (La) were the biggest of the amygdaloid subnuclei. When these results are considered together with previous reports, basal (B) and lateral nuclei (La) may have developed to adjust the capacity on the purpose of unify learning and memory, such as activation on the level of the neocortex.

The amygdala may play an important role in the expression of emotion by the emotional memory as well
as function in learning and memory in the hippocampus, by way of complicated neural networks among many areas such as the cerebral cortex or limbic system. Experimental reports on the role of amygdaloid subnuclei are many, and the function of human amygdala has been revealed by clinical reports. However, it is difficult in practice to perform experimental stimuli or damage in large-sized animals. In this study, the amygdaloid subnuclei have been identified in the horse, cow and pig, and it is clear that the features of these species’ amygdaloid subnuclei are similar to those reported for various species. In future, it is expected that the functional roles of the amygdaloid subnuclei will become apparent from clinical research and their typical roles will be elucidated by comparison with various species.

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