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FREQUENCY ANALYSIS OF OLFACTORY SYSTEM EEG IN CAT, RABBIT, AND RAT

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Introduction

The olfactory bulb (OB), anterior olfactory nucleus (AON), prepyriform cortex (PC), and amygdaloid nucleus generate repeated bursts of sinusoidal oscillation which are highly conspicuous in the EEG. A principal means of describing this activity has been through spectral analysis, through which it has been observed that the EEGs of different parts of the olfactory system are often highly correlated (Boudreau 1964). The purpose of this report is to present measurements of EEG frequencies from 3 species of mammals, and to compare the results with values reported in the literature for other species. Data are presented on the distribution of burst frequencies from OB, AON, and PC of cat and rabbit, and from OB and PC of the rat. The span of frequencies observed in these species was from 35 to 85 c/sec.

Methods

EEGs were recorded from chronically implanted tungsten microelectrodes in the OB and PC of 3 rats. In 6 cats and 12 rabbits, this same electrode type was implanted in the AON and PC. The prepyriform cortex was considered to be the termination area of lateral olfactory tract (LOT) axons, lying on the part of the forebrain from the entorhinal sulcus medially to the rhinal fissure laterally. Bulbar recording in cat and rabbit was from stainless steel wires (250 µm in diameter) embedded in dental acrylic at intervals of 500 µm, forming an 8 X 8 array which was placed on the lateral surface of the OB. Depth electrodes were positioned to maximize the amplitude of the antidromic bulbar and orthodromic cortical field potential evoked by repetitive electrical stimulation of the LOT. Since the LOT stimulating electrode was positioned in the retrobulbar area, it was used for chronic recording from the AON. All sites in the bulbar array were monitored, and the electrode location with maximal amplitude bursts was chosen for polygraph recording. Bursts from depth and surface sites were selected from polygraph records taken at least 7 days postoperatively from waking, motivated animals. Recordings of OB and PC (and AON in cat and rabbit) EEGs were made simultaneously on adjacent polygraph channels.

Bursts were sampled from polygraph records lasting several seconds during motivated states when burst amplitudes were high. A random sample of bursts was formed from several records of each animal, taken over a number of recording sessions. No specific sensory stimuli were employed. Frequencies were determined by counting the number of zero crossings in bursts within a given time period. This procedure was validated by use in conjunction with power spectral analysis (Boudreau and Freeman 1963).

Results

Bursts were observed in two circumstances. The first was when a burst occurred in one structure (OB, PC, AON) and was accompanied by a burst in the other two (Fig. 1). Table I lists the resulting mean and standard deviation for the distribution of frequencies from each region in the cat, rabbit, and rat.

The second circumstance was when a burst was observed in one structure without a concurrent burst in the other structures. These isolated bursts occurred far less commonly than concurrent bursts. The means and standard deviations of their frequency distributions are tabulated in Table II.

The most striking result in Tables I and II and Fig. 1 was a species difference. Frequencies in the cat's olfactory system had lower mean values and had smaller variances in all 3 regions than in either the rabbit or rat. The frequency distribution for concurrent and isolated bursts from the OB had a lower mean and smaller variance in the rat than in the rabbit. The corresponding values for the PC were also lower in the rat for isolated bursts, but were nearly the same in rat and rabbit for concurrent bursts.

An *F* test was applied to the data in Tables I and II to determine whether the frequency distributions differed significantly between structures for the concurrent and isolated burst cases. The hypothesis employed was that for each species the mean frequencies of the structures were equal. By the *F* test, the hypothesis was accepted for the concurrent case in cat and rat, and was rejected for the rabbit. The rabbit data were inspected for differences between pairs of means by the Q method (based on the Studentized Range). The only significant difference at the 95% confidence level between pairs of structures was between the OB and PC. Therefore, except for the difference between OB and PC in rabbit, concurrent burst frequencies were not significantly different at the 95% level between structures in cat, rabbit and rat. For isolated bursts, the mean frequencies were significantly different at the 99% level between all pairs of structures in all 3 animals, except between OB and PC in cat.

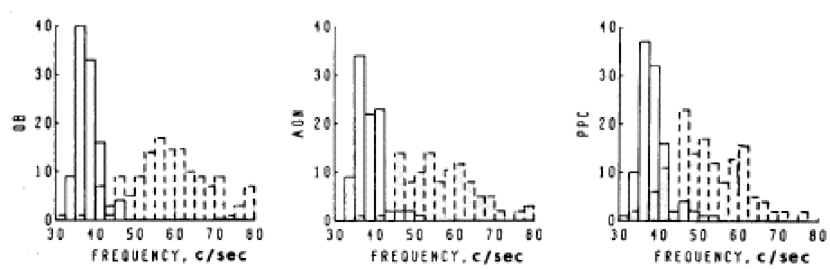

Fig. 1. Histograms are shown of EEG bursts recorded simultaneously from the olfactory bulb (OB), anterior olfactory nucleus (AON), and prepyriform cortex (PPC) in waking cats (open bars) and rabbits (stippled bars). The measurements were made from zero crossings in polygraph traces at 100 mm/sec

paper speed, and were pooled from 12 rabbits and 6 cats.

TABLE I

Concurrent bursts: sample size, mean frequency, and standard deviation are listed according to species and structure.

		Frequency (c/sec)		
		Sample size	Mean	Standard deviation
Cat	OB	104	37.8	2.6
	AON	96	38.2	3.3
	PC	106	38.3	3.7
Rabbit	OB	140	58.6	10.6
	AON	103	56.3	10.7
	PC	140	52.2	8.8
Rat	OB	327	52.6	7.6
	PC	272	52.5	9.3

TABLE I Concurrent bursts:sample size, mean frequency, and standard deviation are listed according to species and structure.

TABLE II

Isolated bursts: sample size, mean frequency, and standard deviation are listed according to species and structure.

		Frequency (c/sec)		
		Sample size	Mean	Standard deviation
Cat	OB	30	37.1	2.1
	AON	28	40.8	4.8
	PC	22	37.3	2.0
Rabbit	OB	42	55.5	7.5
	AON	15	65.6	11.3
	PC	48	46.0	7.2
Rat	OB	143	51.0	5.5
	PC	21	43.4	4.5

TABLE II Isolated burst: sample size, mean frequency, and standard deviation are listed according to species and structure.

TABLE III

Comparison of frequencies of concurrent and isolated bursts with pooled two-tailed t test.

		Difference (isolated − concurrent means)	t	P
Cat	OB	37.1 − 37.8 = −0.7	−1.35	0.018
	AON	40.8 − 38.2 = +2.6	3.29	0.002
	PC	37.3 − 38.3 = −1.0	−1.23	0.200
Rabbit	OB	55.5 − 58.6 = −3.1	−1.77	0.081
	AON	65.6 − 56.3 = +9.3	3.12	0.003
	PC	46.0 − 52.2 = −6.2	−4.40	0.001
Rat	OB	51.0 − 52.6 = −1.6	−2.28	0.022
	PC	43.4 − 52.5 = −9.1	−4.43	0.001

TABLE III Comparison of frequencies of concurrent and isolated bursts with pooled two–tailed t test.

TABLE IV

Rat OB concurrent bursts: sample size, mean frequency, and standard deviation are listed for 3 test animals.

Subject	Frequency (c/sec)		
	Sample size	Mean	Standard deviation
1	98	45.6	5.5
2	116	52.9	8.1
3	113	59.3	9.2

TABLE IV Rat OB concurrent bursts: sample size, mean frequency, and standard deviation are listed for 3 test animals.

In which structures were the frequency differences between concurrent and isolated bursts predominant? Table III shows that on the average over species the frequency of isolated bursts from the OB was 3.4% lower than the frequency of concurrent bursts ($P < 0.05$ for cat and rat). The mean AON frequency was 11.7% higher for isolated than concurrent bursts ($P < 0.01$ for cat and rabbit), whereas the mean PC frequency was 10.6% lower for isolated than concurrent bursts ($P < 0.001$ for rabbit and rat).

Discussion

These results imply that burst frequencies in the AON and PC were dependent on the frequency of concurrent bursts in the OB, whereas the OB frequency was relatively independent of concurrent bursts in the AON or PC. This is predictable from the facts that the main anatomical pathway in the olfactory system is from the OB to the AON and PC, whereas feedback pathways from the latter two structures to the OB are secondary. That is, each of the 3 structures may be capable of generating oscillatory bursts at its characteristic frequency, but the bulb is equipped with a powerful axonal projection pathway (the lateral olfactory tract) to drive the other structures at the bulbar transmission frequency.

Evoked potential studies have shown that the AON and PC respond to impulse input in the manner of a tuned bandpass filter (Freeman 1975). The present EEG results suggest that these parts of the olfactory projection area may be tuned to different characteristic frequencies. This might enable each part to respond preferentially to a selected range of bulbar transmission frequencies.

Over pools of measurements the variance of frequency was positively correlated with the mean. Thus in the cat, the mean frequencies in all 3 olfactory regions were the lowest of the 3 species, and the variance was the smallest. In the rat, the mean and variance were intermediary, and in the rabbit both the mean and variance were greatest. Another example of this relation was found in comparing the individual data for concurrent OB bursts from the 3 rats used in this study (Table IV). (Cat and rabbit data were not tabulated on an individual basis.) A positive correlation was found between the frequency mean and variance. These results suggest that there are circumstances in which frequencies in the olfactory system cluster at the low end of the observed 35–85 c/sec range with a relatively small variance compared to other conditions in which higher variance accompanies higher frequencies.

Mathematical studies of the neural feedback mechanisms of the bulb and cortex by piece–wise linear analysis (Freeman 1975) and by simulation with non–linear integrodifferential equations (Freeman 1979) have demonstrated a mechanism for frequency convergence within a narrow range near 40 c/sec. The sets of equations sufficed to generate simulated EEG bursts over a frequency range of 35–85 c/sec, but within the limits of stability the maximal power of the system, with its accompanying capacity for entrainment, occurred at 40 c/sec.

Sinusoidal bursts from the olfactory bulb have been subjected to frequency analysis in a large number of species, both poikilothermic and homeothermic. For poikilotherms, frequencies from the OB have been measured in rainbow trout from 5 to 15 c/sec (Hara et al. 1973); himé salmon from 5 to 8 c/sec (Satou 1974); chum salmon at 5 c/sec (Kaji et al. 1975); char from 4 to 12 c/sec (Døving and Belgaugh 1977); frog from 3 to 16 c/sec (Ottoson 1959; Takagi and Shibuya 1960; Hobson 1967); toad from 5 to 19 c/sec (Takagi and Shibuya 1960; Segura and De Juan 1966; Graystone et al. 1970); iguana from 10 to 20 c/sec (Graystone et al. 1970); caiman from 5 to 14 c/see (Verlander and Huggins 1977); coachwhip snake from 22 to 32 c/sec (Graystone et al. 1970); and bullsnake from 12 to 24 c/sec (Graystone et al. 1970). In avian species, OB frequencies have been reported in multiple ranges: in albatross at 10–12, 20–25 and 39 c/sec (Wenzel and Sieck 1972); in duck at 1–5, 15–25 and 38 c/sec (Wenzel and Sieck 1972); in shearwater at 40–45 and 56 c/sec (Wenzel and Sieck 1972); in vulture at 1–5, 20–25 and 18 c/sec (Wenzel and Sieck 1972); and in pigeon at 1–6, 15–25 and 30–45 c/sec (Sieck and Wenzel 1969). In mammals, frequencies have been measured in rat PC at 51 c/sec (Woolley and Timiras 1965); rabbit OB from 37 to 75 c/sec (Hughes and Hendrix 1967; Moulton 1963); hedgehog OB from 30 to 40 c/sec (Adrian 1942); cat OB from 30 to 35 c/sec (Gault and Leaton 1963) and from 38 to 58 in PC (Freeman 1959); dog OB from 40 to 46 c/sec (Domino and Ueki 1960); monkey OB from 32 to 50 c/sec (Domino and Ueki 1960; Hughes and Mazurowski 1962); and human OB from 32 to 44 c/sec (Hughes et al. 1969).

The olfactory bulb of the homeotherm sustains oscillations in a higher frequency range than the poikilotherm. Takagi and Shibuya (1960) suggested that if Q_{10} for this mechanism were assumed to be 2, then the frequency differences between mammals and amphibians could be explained on the basis of temperature. Graystone et al. (1970) in amphibians, Dupé and Godet (1969) in lungfish, Døving and Belgaugh (1977) in char, Huggins et al. (1968) in caiman, and Putkonen and Sarajas (1968) in hypothermic rabbits have demonstrated intraspecific linear dependence of frequency on temperature. The Q_{10}s were approximately 2 in all these studies. Given the similarity of neuronal cell type and connectivity among species, it appears that the frequency difference between homeotherms and poikilotherms is in fact dependent on temperature. However, this does not hold for the smaller range of interspecific differences among mammals.

EEG activity in the 35–85 c/sec range has been recorded from the hippocampus (Stumpf 1965), medial geniculate (Rowland 1958), striaturn (Galambos 1958), pallidurn (Galambos 1958), amygdaloid (Lesse 1960; Killam and Killam 1967), and frontal, parietal, temporal, and occipital cortex in cat, dog, monkey, and man (Sheer and Grandstaff 1970; Sheer 1976). The paucity of systematic analysis of this activity in comparison to studies of the theta, alpha, and beta ranges may be attributed to its inaccessibility. It is usually low in amplitude, is smoothed by the inertia of pen recorders, and is readily obscured by the scalp EMG. The tendency for smoothing by latency dispersion in spatially distributed generators is much greater than for lower EEG frequencies.

Summary

EEG activity in the 35–85 c/sec range has been reported from the rhinencephalon of numerous species of homeotherms, and from numerous parts of the forebrain in carnivores and primates, including man. Unimodal distributions of frequencies within this range for each of 3 species (cat, rat and rabbit) are reported here with proportionality between means and variances. Theoretical analysis has shown that the basis for this oscillatory activity lies in the feedback synaptic interactions of assemblies of excitatory and inhibitory neurons, and that there is a neural basis for frequency convergence and high amplitude near 40 c/sec. Given the synaptic mechanisms, the unimodal sprectal distributions, and the widespread occurrence of EEG activity in this range, it clearly represents a unique and identifiable form of brain activity.

Résumé

Analyse de fréquence de l'EEG du systéme olfactif chez le chat, le lapin et le rat

Une activité EEG dans la bande de 35–85 c/sec a été décrite au niveau du rhinencéphale de plusieurs espéces d'animaux homéothermes et dans plusieurs parties du tronc cérébral antérieur chez les carnivores et les primates, incluant I'homme. Des distributions unimodales de fréquences A l'intérieur de cette bande sont rapportés ici pour chacune de 3 espèces (chat, rat et lapin), avec la proportionnalité entre les moyennes et les variances. L'analyse théorique a montré que la base de cette activité oscillatoire siége dans des interactions synaptiques avec feed–back de groupes de neurones excitateurs et inhibiteurs et qu'il y a une base neuronique A la convergence de fréquence et a l'amplitude élevée voisine de 40 c/sec. Etant donné ces mécanismes synaptiques, les distributions spectrales unimodales, et la survenue trés largement répartie de l'activité EEG dans cette bande de fréquence, celle–ci représente clairement une forme unique et identifiable d'activité cérébrale.

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