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AN INVESTIGATION OF THE NATURE OF THE ANATOMICAL CONNECTIVITY BETWEEN MEDIAL PREFRONTAL CORTEX AND CAUDATE-PUTAMEN REWARD SITES IN THE RAT

Monika Maria Trzcińska

A thesis submitted to the School of Graduate Studies of the University of Ottawa as partial fulfilment of the requirements for the degree of Doctor of Philosophy

May 1995

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Finances 0277
Formation des enseignants 0530
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Langues et littérature 0279
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- Comparée 0295
- Médiévale 0297
- Moderne 0298
- Africaine 0316
- Américaine 0591
- Anglaise 0593
- Asiatique 0305
- Canadienne (Anglaise) 0352
- Canadienne (Française) 0355
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Droit 0398
Économie
- Généralités 0501
- Commerce-Affaires 0505
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- Finances 0508
- Histoire 0509
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Études canadiennes 0385
Études féministes 0453
Folklore 0358
Géographie 0366
Gérontologie 0351
Gestion des affaires
- Généralités 0310
- Administration 0454
- Banques 0770
- Comptabilité 0272
- Marketing 0338

Histoire
- Histoire générale 0578
- Ancienne 0579
- Médiévale 0581
- Moderne 0582
- Histoire des noirs 0328
- Africaine 0331
- Canadienne 0334
- États-Unis 0337
- Européenne 0335
- Moyen-orientale 0333
- Latino-américaine 0336
- Asie, Australie et Océanie 0332

Histoire des sciences 0585
Loisirs 0814
Planification urbaine et régionale 0999
Science politique
- Généralités 0615
- Administration publique 0617
- Droit et relations internationales 0616

Sociologie
- Généralités 0626
- Aide et bien-être social 0630
- Criminologie et établissements pénitentiaires 0627
- Démographie 0938
- Études de l' individu et de la famille 0628
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Transports 0709
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Agriculture
- Généralités 0473
- Agronomie. 0285
- Alimentation et technologie alimentaire 0359
- Culture 0479
- Élevage et alimentation 0475
- Exploitation des pâturages 0777
- Pathologie animale 0476
- Pathologie végétale 0480
- Physiologie végétale 0817
- Sylviculture et faune 0478
- Technologie du bois 0746

Biologie
- Généralités 0306
- Anatomie 0287
- Biologie (Statistiques) 0308
- Biologie moléculaire 0307
- Botanique 0309
- Cellule 0379
- Écologie 0329
- Entomologie 0353
- Génétique 0369
- Limnologie 0793
- Microbiologie 0410
- Neurologie 0317
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- Radiation 0821
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Biophysique
- Généralités 0786
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Paléozoologie 0985
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- Alimentation et nutrition 0570
- Audiologie 0300
- Chimiothérapie 0992
- Dentisterie 0567
- Développement humain 0758
- Enseignement 0350
- Immunologie 0982
- Loisirs 0575
- Médecine du travail et thérapie 0354
- Médecine et chirurgie 0564
- Obstétrique et gynécologie 0380
- Ophtalmologie 0381
- Orthophonie 0460
- Pathologie 0571
- Pharmacie 0572
- Pharmacologie 0419
- Physiothérapie 0382
- Radiologie 0574
- Santé mentale 0347
- Santé publique 0573
- Soins infirmiers 0569
- Toxicologie 0383

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Sciences Pures
Chimie
- Généralités 0485
- Biochimie 487
- Chimie agricole 0749
- Chimie analytique 0486
- Chimie minérale 0488
- Chimie nucléaire 0738
- Chimie organique 0490
- Chimie pharmaceutique 0491
- Physique 0494
- PolymÇres 0495
- Radiation 0754

Mathématiques 0405
Physique
- Généralités 0605
- Acoustique 0986
- Astronomie et astrophysique 0606
- Électronique et électricité 0607
- Fluides et plasma 0759
- Météorologie 0608
- Optique 0752
- Particules (Physique nucléaire) 0798
- Physique atomique 0748
- Physique de l'état solide 0611
- Physique moléculaire 0609
- Physique nucléaire 0610
- Radiation 0756

Statistiques 0463

Sciences Appliqués Et Technologie
Informatique 0984
Ingénierie
- Généralités 0537
- Agricole 0539
- Automobile 0540
- Biomédicale 0541
- Chaleur et thermodynamique 0348
- Conditionnement (Emballage) 0549
- Génie aérospatial 0538
- Génie chimique 0542
- Génie civil 0543
- Génie électronique et électrique 0544
- Génie industriel 0546
- Génie mécanique 0548
- Génie nucléaire 0552
- Ingénierie des systèmes 0790
- Mécanique navale 0547
- Métallurgie 0743
- Science des matériaux 0794
- Technique du pétrole 0765
- Technique minière 0551
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- Technologie hydraulique 0545

Mécanique appliquée 0346
Géotechnologie 0428
Matières plastiques (Technologie) 0795
Recherche opérationnelle 0796
Textiles et tissus (Technologie) 0794

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Généralités 0621
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Psychobiologie 0349
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Psychologie sociale 0451
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Dedication

This thesis is dedicated to my parents, Urszula Trzcińska - Master of Science in Mathematics and Kazimierz Trzciński - Master of Arts in German Philology, who truly deserve an honourary doctorate for seeing me through this program and much more...

"One never notices what has been done, one can only see what remains to be done."

Maria Skłodowska-Curie

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My dearest Matthew, now I am ready for our Bostonian adventure!

Abstract

Research on the mechanisms by which the brain codes, understands, and remembers reward has focused on sites lying along the medial forebrain bundle (MFB). Regions more anterior to the MFB, such as the medial prefrontal cortex (MPFC) and caudate-putamen (CPu), have received little attention in this regard.

Consequently, the first experiment was to determine the distribution of sites in the MPFC and CPu that support intracranial self-stimulation. Overall, 255 MPFC and 187 CPu individual sites were evaluated in 67 animals using moveable electrodes; only 11% of the examined areas showed reliable self-stimulation, which was in most cases accompanied by overt seizures. Most positive sites were clustered in the ventromedial aspects of the MPFC and CPu. Charge values obtained for both regions were widely distributed and ranged from 0.68 to 1.63 μC across sites, values in line with those reported for MFB sites.

One of the problems encountered in this study was the slow acquisition of MPFC self-stimulation. In order to overcome this obstacle, some subjects were implanted with two electrodes, one aimed at the MPFC, and the other at the CPu, ventral tegmental area, or lateral hypothalamus. Once stable thresholds were obtained at the extra MPFC sites, self-stimulation was then evaluated at the MPFC site. Only animals with CPu placements showed transference of this behaviour to the MPFC, suggesting

that these two regions might form part of the same reward substrate, a view that has anatomical and electrophysiological support.

The next step was to estimate the excitability cycles that characterize MPFC and CPu reward fibers. Using the behavioural adaptation of the refractory period test, the range of recovery from refractoriness was determined to span from 0.8 to 5.4 ms for the CPu and 1.4 to 7.9 ms for the MPFC. These values tend to be longer than the ones typically obtained at MFB self-stimulation sites. One interpretation of the overlap in these estimates is that these regions constitute two separate locations along the same axonal bundle.

This hypothesis was investigated in the third experiment using the behavioural collision technique. Nine ipsilateral pairs of CPu and MPFC sites were evaluated for axonal connectivity, four of which showed a pattern consistent with the interpretation that at least a subset of fibers course between the MPFC and CPu. The conduction velocity estimates of these were calculated to be between 0.2 and 1.8 m/s, values that are much less than the estimates reported for the MFB and consistent with the activation of thin, unmyelinated axons.

The anatomical relationship between the MPFC and CPu is characterized by direct corticostriatal projections, some of which use glutamate as a neurotransmitter. There is also some evidence that dopaminergic fibers make direct contact with these descending neurons. The behavioural estimates of refractoriness

and the conduction velocities obtained here closely match those found electrophysiologically for dopaminergic and glutamatergic fibers. Given this background, a model of a possible neuronal interaction between the MPFC and CPu reward regions is proposed to account for the findings reported here, suggesting the involvement of these neurotransmitter systems in mediating the rewarding effects of MPFC and CPu stimulation.

Table of contents

List of figures

List of tables

INTRODUCTION

Historical background

Forty years ago Olds and Milner (1954) discovered that electrical stimulation of certain brain structures in the rat could induce the strong pursuit of a stimulus. This phenomenon, termed brain stimulation reward, was the start of a new era in the study of the physiological mechanisms underlying reward and motivation (Milner, 1989). Up to that moment experimental evidence available at the time appeared to indicate that stimulation of the brain did not produce any emotional experience (Valenstein, 1973). Since then, brain stimulation reward has been observed in a number of species, from invertebrates, such as snail (Balaban and Chase, 1989), vertebrates, including birds, (Goodman and Brown, 1966), and fish (Boyd and Gardner, 1962), to mammals, including mouse (Zacharko, Bowers, Kokkinidis, and Anisman, 1983), rabbit (Bruner, 1966), cat (Justesen, Sharp, and Porter, 1963), dog (Stark and Boyd, 1961), as well as monkeys (Brodie, Moreno, Malis, and Boren, 1960) and humans (Bishop, Elder, and Heath, 1963; Heath, 1972), although it has been most studied in the laboratory rat. The fact that brain stimulation reward can be obtained from the same anatomical regions across species suggests that reward plays a fundamental role in the organism's behavioural repertoire, irrespective of its position along the evolutionary scale (Valenstein, 1973).

The study of brain stimulation reward in humans was short-

lived, primarily because of moral and ethical issues centered around such invasive procedures as implantation of depth electrodes in the brain, and also because of a fear of using the technique as a means of dominating individuals, either by controlling their movement or by evoking extreme and "addictive" pleasurable sensations (Delgado, 1973; Valenstein, 1973).

In rats, brain stimulation reward has been used as a tool to study the circuitry underlying appetitive behaviours. Typically, the animal is trained on a continuous reinforcement schedule to perform an instrumental task, such as lever pressing, in order to stimulate neurons at the tip of an electrode chronically implanted in the brain. It learns quickly to engage in persistent, vigorous behaviour, sustaining high response rates over several hours without any apparent loss of interest or fatigue. Once the stimulation is turned off, the behaviour extinguishes rapidly (Howarth and Deutsch, 1962). It is important to note that it is the animal that controls the rate of stimulation delivery. Indeed, it has been found that rats will escape experimenter-imposed stimulation, when presented at the same rate and pattern as their self-stimulation trials (Steiner, Beer and Shafter, 1969). Furthermore, when allowed to choose between self-stimulation and non-contingently delivered stimulation, rats prefer the former (Ettenberg, Laferrière, Milner, and White, 1981).

These animals can be trained to perform a variety of behaviours in order to obtain electrical stimulation of the

brain; for example it has been shown that under restricted experimental conditions, they will run a maze, forgo food, water, sleep, a mate, and tolerate intense painful stimuli for access to rewarding brain stimulation (Gallistel, 1975; Olds, 1958; Routtenberg and Lindy, 1965; Stellar, Brooks, and Mills, 1979; Valenstein, 1973).

Another reason why brain stimulation reward is viewed as a model for the study of naturally occurring reinforcers is that stimulation of reward-relevant structures also supports feeding (Hoebel and Teitelbaum, 1962), drinking (Mogenson and Stevenson, 1966), copulation (Cagguila and Hoebel, 1966), analgesia (Rose, 1974), exploration (Rompré and Miliaressis 1980), locomotion (Christopher and Butter, 1968; Rolls and Kelly, 1972), sniffing (Rossi III and Panksepp, 1992), startle response (Yeomans, Hempel, and Chapman, 1993a), and aversion, as demonstrated by stimulation-induced escape (Bielajew and Shizgal., 1980; Bower and Miller, 1958). It is thus believed that the neural elements triggered as a consequence of the task are at least a subset of the ones that are active when an organism is faced with a naturally occurring reinforcer (Conover and Shizgal, 1994; Conover, Woodside, and Shizgal, 1994). The similarities observed between natural feeding, stimulation-induced feeding, intraoral sucrose infusion, sodium balance, and self-stimulation support this notion (Conover and Shizgal, 1994; Conover et al., 1994; Valenstein, Cox, and Kakolewski, 1968). One interpretation is that electrical self-stimulation of the brain represents a

behavioural response to a reinforcing stimulus that is not actually consumed (Lenzer, 1971). In fact, when intermittent (vs. continuous) reinforcement schedules are used, or when the intensity and quality of a reinforcer, as well as the duration of the deprivation period are properly adjusted, central and conventional reinforcers become equally effective in maintaining an operant behaviour, such as bar pressing (Beninger, Laferrière, and Milner, 1978; Mogenson and Cioe, 1977).

In this respect, there has been much attention paid to the anatomical structures that underlie brain stimulation reward and other stimulation-evoked behaviours. Of the latter the one that has raised much interest is stimulation-induced feeding. There is some evidence that the substrates for brain stimulation reward and stimulation-induced feeding are indistinguishable (Gratton and Wise, 1988a), at least at the level of the medial forebrain bundle (MFB). However, recent studies suggest that the parametric and pharmacological profiles of these two behaviours are different (Harris, Bielajew, and Merali, 1991; Waraczynski and Kaplan, 1990), and that their circuitry deviates in the more anterior regions, such as the sulcal prefrontal cortex (Bielajew and Trzcińska, 1994). For example, recently it was demonstrated that stimulation-induced feeding can be obtained in the absence of self-stimulation in this structure (Bielajew and Trzcińska, 1994). Other studies have shown that circling, motoric effects, and aversion aren't subserved by the same neurons that mediate brain stimulation reward (Bielajew and Shizgal, 1980; Durivage

and Miliaressis, 1987; Miliaressis and Rompré, 1980). In summary, the resemblance between the effects of rewarding brain stimulation, other stimulation-evoked behaviours, and natural reinforcers may not be spurious, but may indeed reflect the activation of the circuitry underlying natural appetitive behaviours.

Most of what is known about the reward substrate to date has come from studies of two MFB regions - the lateral hypothalamus and the ventral tegmental area (for reviews, see Milner, 1991; Shizgal, 1989; Stellar and Stellar, 1985; Yeomans, 1990). In addition, brain stimulation reward from activation of such structures as the cerebellum (Corbett, Fox, and Milner, 1982a), central gray (Cooper and Taylor, 1967), periaqueductal gray (Bielajew, Jordan, Ferme-Enright, and Shizgal, 1981), raphe nuclei (Miliaressis, Bouchard, and Jacobowitz, 1975; Rompré and Miliaressis, 1985), pontine tegmentum (Rompré and Boye, 1989), hippocampus (Ursin, Ursin, and Olds, 1966), amygdala (Kane, Coulombe, and Miliaressis, 1991; Prado-Alcala and Wise, 1984), dorsal diencephalon (Clavier and Gerfen, 1979; Vachon and Miliaressis, 1992), mediodorsal thalamus (Huston and Borbely, 1974; Bielajew & Fouriezos, 1985), habenular complex (Sutherland and Nakajima, 1981), lateral preoptic area (Fouriezos, Walker, Rick, and Bielajew, 1987), nucleus accumbens (Phillips and Fibiger, 1978; Prado-Alcala, Kent, and Reid, 1975), substantia nigra (Zacharko, Kasian, Irwin, Zalcman, Lalonde, MacNeil, and Anisman, 1983), caudate-putamen (CPu) (Justesen et al.,1963), red

nucleus (Zacharko et al., 1990), medial prefrontal cortex (MPFC) (Schenk, Prince, and Shizgal, 1985), and sulcal prefrontal cortex (Corbett, Laferrière, and Milner, 1982c; Robertson, Laferrière, and Milner, 1982a) has been reported [for a more detailed review see Phillips, 1984; Phillips and Fibiger, 1989].

In order to systematically study brain stimulation reward, researchers had to agree on certain assumptions about the neural population being stimulated (Ranck, 1975), as well as what constitutes adequate stimulation parameters. In addition, it is elemental in the studies of brain stimulation reward to distinguish between reward neurons and other cells activated by stimulation at reward loci. The psychophysical approach which was developed to deal with these issues is the strategy behind the studies in this dissertation.

Psychophysical approach

According to the four-stage minimal model (Gallistel, Shizgal, and Yeomans, 1981), the neurons directly under the electrode tip constitute the first-stage neurons, but they are not the ones responsible for the experience of reward. These cells are analogous to "transducer cells" in any of the five sense modalities. They convert a physical stimulus, such as pressure, light, sound, etc., to the language that the central nervous system can understand - an action potential. In the case of the reward system, these cells translate a physical stimulus - current, into a psychological effect - reward. The signal

propagated by the action potentials generated in neurons directly activated at the tip of the electrode has to be integrated over many synapses (stage 2), both temporally (frequency of stimulation) and spatially (current) before producing the rewarding effect. Once the computation is accomplished, the conversion process (stage 3) gives rise to a memory (stage 4) of the rewarding effect of electrical stimulation, as demonstrated by stable and enduring self-stimulation.

In order for stage 4 (output) to reflect what has occurred in stage 1 (input) the assumption of monotonicity has to be met. The integrator of spatiotemporal information acts as a monotonic counter, as it can record the same effect from different patterns of stimulation (Gallistel, 1978; Gallistel and Leon, 1991; Gallistel et al., 1981). For example, let's say that at the level of the first-stage neurons, 100 neurons fire 10 times; this will be equivalent to 10 neurons firing 100 times (Gallistel 1974). If every output of each stage stems from one single input then the input/output relationship of all stages is monotonic. If the monotonicity argument is violated at any one of the four stages, then it cannot be corrected at the subsequent stage, and as a consequence the behavioural output fails to reflect the initial input (Gallistel and Leon, 1991; Gallistel et al., 1981). In other words, a monotonic relationship should exist between inputs and outputs at any of the four stages between the directly stimulated neurons and the behavioural outcome of this stimulation, in order for the output to accurately reflect what

has occurred in the input stage (for additional elaboration of the model, see Gallistel et al., 1981). However, the violation of the monotonicity assumption does not seriously affect the measurement of the behavioural output, as long as the properties of the first stage neurons are inferred from the ascending portion of the rate-frequency curve. Further, the magnitude of the experienced reward is determined by the net rate of firing in the population, and to the integrator it is irrelevant how this net rate is obtained (by increasing the current and decreasing the frequency, or vice versa). However, recent evidence points to the fact that there may be more than one integrator operating to combine the signals produced in different reward-relevant axons within the MFB (Leon and Gallistel, 1992). The magnitude of the spatio-temporal integration of signals in the reward substrate, as measured by a two-lever choice paradigm with variable interval schedule, depends on the contribution of different subsets of reward-relevant fibers excited by the stimulation.

Although the input (stage 1) can be controlled and its effects on the output (stage 4) observed, not much has been known about what occurs at stages 2 and 3. However, recent studies using a preference paradigm suggest that the magnitude of reward can summate, as indicated by the preference of the animal for one of two rewards (Anderson and Miliaressis, 1994; Malette and Miliaressis, 1990; Miliaressis and Malette, 1987), thus suggesting what may occur at stages 2 and 3. Those same tests

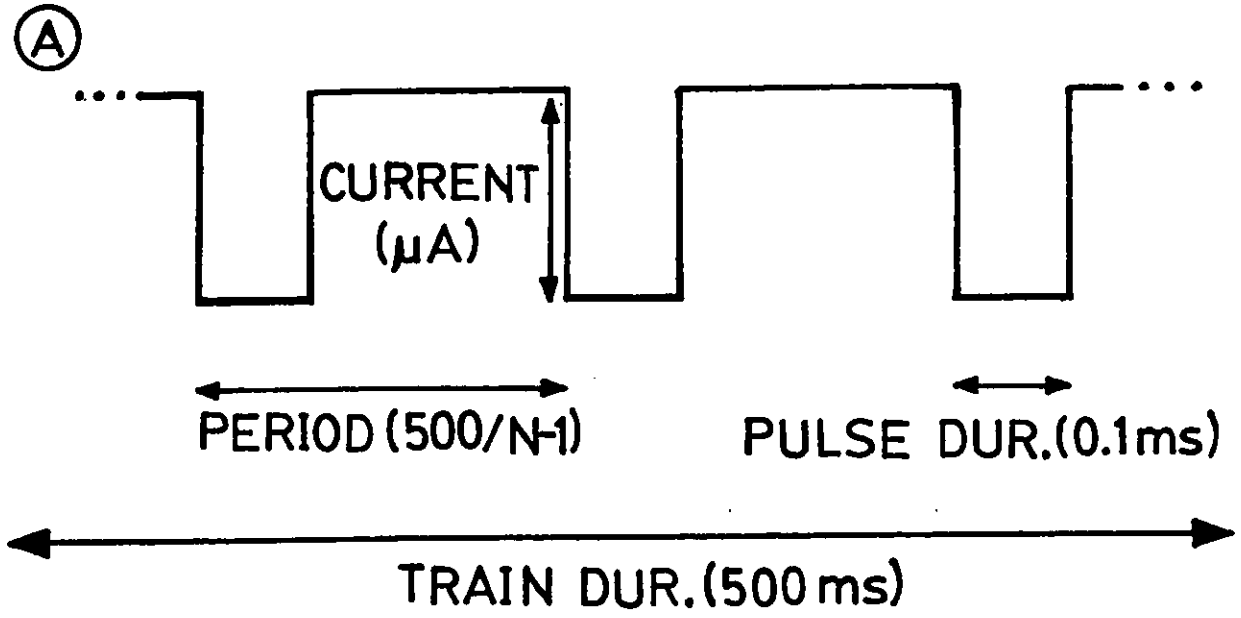
have also provided evidence that the MFB is not a homogenous reward pathway (Anderson and Miliaressis, 1992), as often implied in the literature.

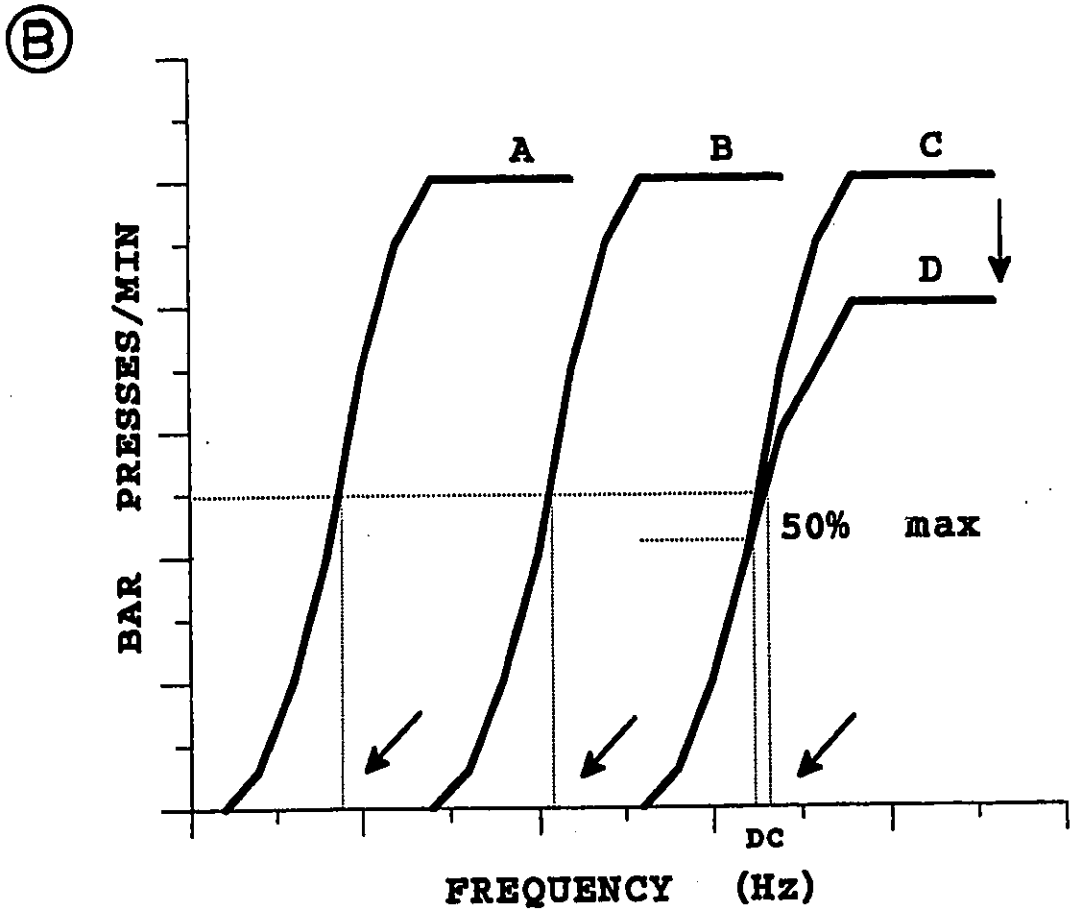
Typically, the simplest method for characterizing first stage neurons is to analyze a family of rate-frequency curves, similar to the one shown in Figure 1B, obtained by keeping one parameter constant and by systematically varying another one, until some behavioural criterion is reached (Gallistel, 1978; Miliaressis, Rompré, Laviolette, Phillippe, and Coulombe, 1986b). The parameters used to collect such a curve are illustrated in Figure 1A. Below a certain frequency, very few or no responses are observed. Once the minimal frequency necessary to evoke behaviour is obtained, the rate of bar pressing dramatically increases, and approaches an asymptote, or maximum level of performance. At that point, a further increase in the frequency often produces aversive or motoric reactions, and consequently may result in lower rates of bar pressing. Historically, changes in rate have been used as an index of reward. However, long ago Valenstein (1964) pointed out that rates did not constitute a sensible measure of reward. Since then studies have demonstrated that thresholds are a more reliable index of the change in reward magnitude. A decrease in the maximum rate does not indicate a decrease in the rewarding value of the stimulation, but may instead reflect the inability of the animal to perform, due to factors unrelated to reward, such as motoric effects, seizures, health of the animal, etc., (Gallistel et al., 1981;

Figure 1. A) This figure illustrates the relationship between the parameters of square wave cathodal stimulation. Train duration (500 ms) and pulse duration (0.1 ms) are typically set at a constant value, while the frequency, or number of pulses (N) per train, and the current are varied. Period is the reciprocal of the frequency.

B) The series of rate-frequency curves demonstrates how reward and performance factors (motoric effects, seizures) are distinguished using psychophysical techniques. The threshold values (index of reward) are indicated by arrows pointing to the abscissa, and defined as the frequency corresponding to half the maximum rate of responding. If one considers curve B as the baseline, then curve A has shifted to the left, with respect to curve B, with no change in its asymptotic level and thus interpreted as an increase in the rewarding value of the stimulation (lower threshold). Curve C, on the other hand, has shifted to the right, again with no change in the asymptote, and therefore interpreted as a decrease in the rewarding value of the stimulation (higher threshold). Curve D has also shifted to the right in comparison to curve B; in this case however, there is a concomitant decrease in the asymptote, hence indicating not only a drop in the rewarding value of the stimulation, but also one in the level of performance. The functions also illustrate the problem encountered when rate changes are used to measure reward. Consider curves C and D alone. If only the two asymptotic levels are compared, the conclusion is that the experimental

manipulation has decreased the rewarding value of stimulation in curve D, compared to curve C. However, comparison of the frequency thresholds of the same pair of curves suggests that the manipulation has in fact given rise to a slight increase in the rewarding value of the stimulation; curve D has a lower threshold than that of curve C. The curve-shift changes in thresholds at various currents have been borne out from studies of drug effects on brain stimulation reward (Edmonds and Gallistel, 1974).


A
CURRENT
(μA)
PERIOD (500/N-1)
PULSE DUR.(0.1ms)
TRAIN DUR.(500 ms)

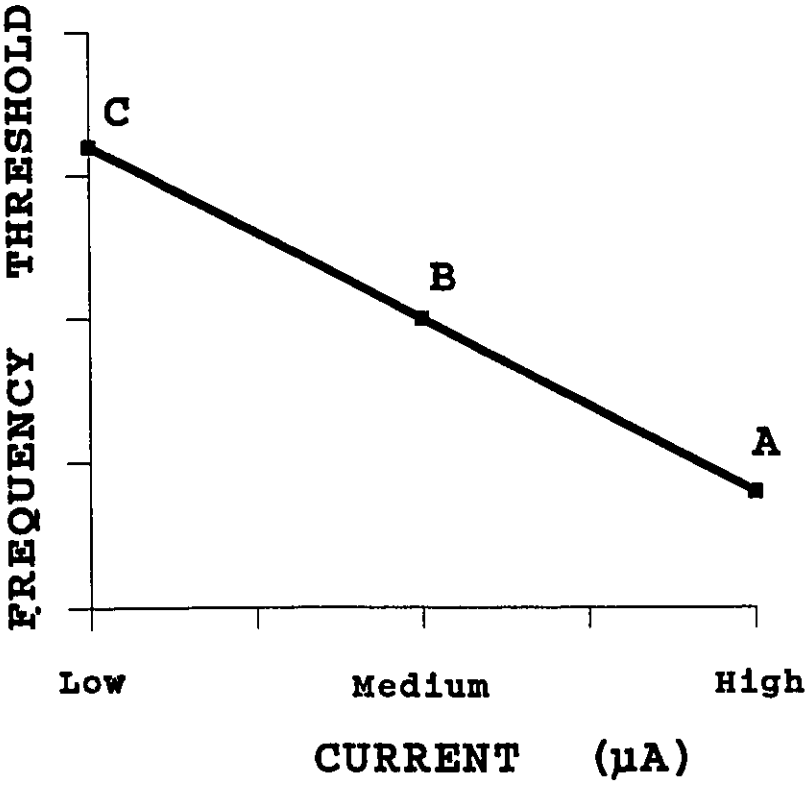

B
A
B
C
D
50%
max
BAR PRESSES/MIN
DC
FREQUENCY (Hz)

Miliaressis, Rompré, and Durivage 1982; Miliaressis and Rompré, 1987; Miliaressis, Malette, and Coulombe, 1986a). In addition, response rate seems to depend not only on the strength of the rewarding stimulation, but also on the reinforcement schedule (Druhan, Levy, and Shizgal, 1993) and the performance requirements (Edmonds and Gallistel, 1974; Fouriezos, Bielajew, and Pagotto, 1990; Frank and Williams, 1985; Hamilton, Stellar, and Hart, 1985). For that reason most investigators in this field today do not use rate of bar pressing to assess reward level. Instead, they choose a threshold as their measure, often defined as the frequency corresponding to half of the maximum response rate.

The frequency thresholds obtained from a family of rate-frequency curves can be plotted as a function of current, as shown in Figure 2. A decrease in the stimulation current results in a shift towards higher frequency thresholds. Because reducing the current decreases the size of the effective stimulation field, more pulses have to be added per stimulation train in order to maintain a constant behavioural output. Conversely, when the stimulation current is augmented, a shift towards lower frequency thresholds is observed.

This relationship between current and frequency provides information concerning the distribution of reward-relevant fibers relative to the stimulating electrode (Gallistel et al., 1981). In a homogenous population of neurons, increasing current would enlarge the size of the stimulation field and augmenting the

Figure 2. This figure illustrates the trade-off function between current and frequency thresholds, generated from the rate-frequency curves shown in figure 1B that were collected at different currents. The pattern suggests that the relationship between current and frequency is linear when plotted on a log-log scale, and indicates the activation of a homogenous population of reward neurons.


FREQUENCY THRESHOLD
C
B
A
Low
Medium
High
CURRENT (µA)

frequency of electrical impulses would increase the number of action potentials per neuron (Gallistel, 1974, 1978; Gallistel and Leon, 1991; Gallistel et al., 1981). It is thus assumed that current and frequency are inversely proportional. Hence, in order to keep the behavioural output constant, when one parameter increases in magnitude, the other one has to decrease in order to compensate. However, there are instances where the radius of effective stimulation field encompasses all of the relevant reward fibers surrounding the electrode tip. Thus, additional increases in current will fail to recruit more reward-relevant fibers. In such case augmenting the current will fail to increase the number of action potentials in the directly stimulated axons, and consequently a decrease in the number of required pulses will not be observed. Similarly, when small currents are employed, very high frequencies have to be applied in order to maintain the inverse proportionality between current and frequency. In such instances the intense level of stimulation may hinder the output of the reward relevant axons, and hence restrict the number of action potentials integrated.

The trade-off functions, such as those described above, are useful in evaluating various brain areas because similar changes in threshold in two different loci are assumed to reflect similar changes in neural density. A basic assumption, stemming from Gallistel's minimal model, states that when neurons with different thresholds are randomly distributed within the stimulation field, the slopes of the trade-off functions reflect

changes in neural density (Miliaressis et al., 1982; Yeomans, 1990a). As the stimulation field exceeds the boundary of effective neurons, the slope of the trade-off function becomes shallower. Hence the trade-off functions permit evaluation of the spatial limits of the reward substrate, and have been used to characterize several stimulation evoked behaviours, such as feeding, self-stimulation (Bielajew and Trzcińska, 1994; Harris and Bielajew, 1988), and circling (Malette and Miliaressis, 1990; Miliaressis and Phillippe, 1984; Miliaressis and Rompré, 1980; Yeomans, Pearce, Wen, and Hawkins, 1984).

A trade-off experiment thus assesses the relationship between two stimulus parameters that produce equivalent behavioural responses. The advantage of using this approach lies in avoiding floor and ceiling effects and the influence of performance variables, such as the animal's state of health. In this manner, trade-off functions have been obtained between current and pulse duration, current and pulse number within a train of fixed duration, and strength, or total charge as a function of train duration (Bielajew, Jurgens, and Fouriezos, 1987a; Edmonds, Stellar, and Gallistel, 1974; Gallistel, 1974; Gallistel et al., 1981; Malette and Miliaressis, 1990; Matthews, 1978; Miliaressis et al., 1986b). In addition, single and double electrode experiments have been used to determine the trade-off functions between current and the number of pulses as a function of pulse-pair interval. In single electrode designs, trade-off functions have been designed to estimate recovery from refractoriness of a

variety of reward structures (Bielajew and Shizgal, 1980; Rompré and Miliaressis, 1980; Yeomans, 1975). Two electrode experiments, on the other hand, have enabled investigators of brain stimulation reward to assess whether two areas that support self-stimulation, for example the lateral hypothalamus and ventral tegmental area, are linked by the same fiber bundle (Shizgal, Bielajew, Corbett, Skelton, and Yeomans, 1980). Ultimately the information obtained from refractory period and collision studies can be used to estimate conduction velocity (Bielajew and Shizgal, 1982; Shizgal et al., 1980). By evaluating self-stimulation in this fashion, some of the electrophysiological properties of the first stage neurons have been identified; based on the refractory period and conduction velocity estimates it appears that these neurons are long and thinly myelinated axons (Gallistel et al., 1981; Shizgal and Murray, 1989; Yeomans, 1988).

Neuropharmacology of the first-stage neurons

Among the many neurotransmitters that could potentially be involved in mediating the response of the first-stage neurons, dopamine has gained the greatest attention. There is some evidence that this neurotransmitter underlies some of the rewarding effects of MFB stimulation (Phillips & Fibiger, 1978; Wise, 1978, 1981; Wise and Hoffman, 1992; Yeomans, Maidment, and Bunney, 1988). This may be true, especially when small electrode tips, or long pulse durations and high currents are employed

(Bielajew, 1991; Bielajew and Fouriezos, 1985; Fouriezos and Wise, 1984; Rolls, 1974; Yeomans et al., 1988). Further, dopamine appears to modulate the impact of such rewarding stimuli as food (Evans and Eikelboom, 1987), water (Heyman, Kinzie, and Seiden, 1986) and brain stimulation reward (Arbuthnott, Crow, Fuxe, Olson, and Ungerstedt, 1970; Stellar and Corbett, 1989). In addition, pharmacological agents that block dopamine release also inhibit self-stimulation in most areas that support this behaviour (Gallistel, 1986; Wise, 1981; Wise and Rompré, 1989; Yeomans, 1989). Despite the evidence for the involvement of dopamine in reward, it appears that many self-stimulation areas, such as those in the cerebellum, dorsal pons, and caudal midbrain are not located near dopaminergic axons (Yeomans, 1990a). In addition, the unmyelinated dopaminergic fibers recover more slowly from refractoriness (1.2 to 2.5 ms) and conduct at slower speeds (0.3-1.5 m/s) than the first stage reward fibers in the MFB (Bielajew and Shizgal, 1986; Gallistel et al., 1981; Shizgal et al., 1980; Yeomans et al., 1988; Yim and Mogenson, 1980), placing suspicion on the idea that they form part of the directly stimulated substrate. One hypothesis that has been proposed suggests that self-stimulation electrodes might excite afferents to the dopamine cells that relay information to forebrain or midbrain structures. However, stimulation parameters usually employed in brain stimulation reward studies, 0.1 ms rectangular pulses at 100-200 Hz (Gallistel et al., 1981), have little effect on unmyelinated dopaminergic fibers (Szabo and Milner, 1973;

Yeomans, 1989). It is possible that dopamine might play a tonic role in brain stimulation reward by modulating the activity of the reward-related neurons beyond the directly stimulated first stage (Gallistel et al., 1981; Gratton, Hoffer, and Gerhardt, 1988, 1988). Also, more recent studies (Gratton et al., 1988; Miliaressis, Emond, and Merali, 1991) report that the region in which dopaminergic neurons can be excited around the electrode tip decreases as pulse duration increases, at least at the level of the MFB. Indeed, some of the dopaminergic cells can be fired transynaptically by the typical self-stimulation parameters (Gratton et al., 1988; Yeomans, et al., 1988). In addition, it appears that equirewarding self-stimulation parameters fail to maintain constant dopamine output, which indicates that the reward signal cannot be transmitted or relayed through the dopaminergic system at any stage of the circuitry (Miliaressis et al., 1991).

Recent results further complicate the picture by indicating that dopaminergic neurons might not be the only ones contributing to the rewarding effect of electrical stimulation. Gratton and Wise (1985) suggested that the MFB reward substrate comprises more than one population of reward fibers, with one of the neurotransmitters being acetylcholine. More recently, Yeomans, Mathur, and Tampakeras (1993b) found that the cholinergic neurons of the pedunculopontine nucleus synapse on ventral tegmental dopamine neurons and appear to be critical for the rewarding effects of lateral hypothalamic stimulation.

Although the search for a neurochemical substrate underlying brain stimulation reward has proven to be elusive, the trade-off functions provide important electrophysiological characteristics of the directly stimulated reward neurons.

One example of a trade-off function is the comparison of frequency thresholds for a train of single-pulses to the frequency threshold for a train of double-pulses. When the double pulses are applied to a single electrode, a refractory period test is conducted, and when applied to two separate electrodes, a test of collision is performed. Both techniques are described in the following section.

Refractory period technique

The refractory period test, as used here, is designed to investigate the post-stimulation excitability characteristics of the neurons that underlie self-stimulation in a given region, and to determine whether this area of interest shares the same characteristics as that of the MFB reward substrate. The first experiments to study the phenomenon of neural refractoriness focused on the behaviour of a frog muscle to infer the recovery properties of a peripheral nerve attached to that muscle (Lucas, 1917); stimulation of the nerve resulted in a muscle twitch. The amplitude of that response was used as an indicator of the nerve excitability. If another stimulation was applied immediately following the first one, no increase in the amplitude of the muscle twitch was observed until a minimum of 1.6 ms interval had

elapsed between stimulations. Because a pair of stimuli delivered within 1.6 ms produced the same response as a single stimulus, 1.6 ms was determined to be the absolute refractory period of that peripheral nerve.

The principles derived from peripheral nervous system studies appear to explain the mechanisms underlying the excitability cycles of central nervous system neurons. Deutsch (1964) was the first to study recovery from refractoriness in self-stimulation studies. He believed that one could indirectly derive the excitability characteristics of cells activated by rewarding brain stimulation from changes in the animal's response rate, at various currents, prompted by varying the interval between two consecutive pulses applied through a single electrode. By convention, the first pulse was called the conditioning, or C pulse, and the second, the test, or T pulse. The latter was called the T pulse because it tested the excitability of the neuronal population following delivery of the C pulse. The interval between the two pulses was termed the C-T interval. In the Deutsch experiment it was assumed that if the two sets of pulses were delivered at a C-T interval exceeding the refractory period of the directly stimulated neurons, then both sets of pulses would produce neural firings, which would subsequently be reflected in the animal's higher rate of responding. If, however, the two sets of pulses were delivered at a C-T interval arriving within the refractory period of the stimulated neurons, then these neurons would respond as if stimulated by the C pulses

only and lower response rates would be observed. By progressively varying the C-T interval, a range of response rates would be produced, from minimum to maximum, depending on whether the second pulse fell within or outside of the refractory period of the directly stimulated neuronal bundle. Some years later, Yeomans (1975) argued that evaluating refractoriness on the basis of response rates biased the results, because it required the unwarranted assumption that a linear relationship between response rate and neural excitability existed. He showed that the estimates of refractoriness based on rate varied as a consequence of the stimulation parameters: different frequencies gave rise to different recovery estimates. Interestingly, the highest and the lowest frequencies produced no change in rate as a function of C-T interval, yielding a flat function due to the ceiling and floor effects on performance. As mentioned earlier, the relationship between the rate of responding and the rate of neuronal excitation has to be monotonic in order to adequately reflect the behavioural outcome of stimulation (stage 4) as a function of the original level of stimulation (stage 1).

The parameters used to estimate behaviourally derived refractory periods and a representative refractory period curve are illustrated in Figure 3B. A series of rate-frequency curves are first obtained at different C-T intervals. The threshold for trains of single pulses (C pulses only) is then compared to the threshold for trains of double pulses (C and T pulses). When the delay between the C and T pulses is greater than the refractory

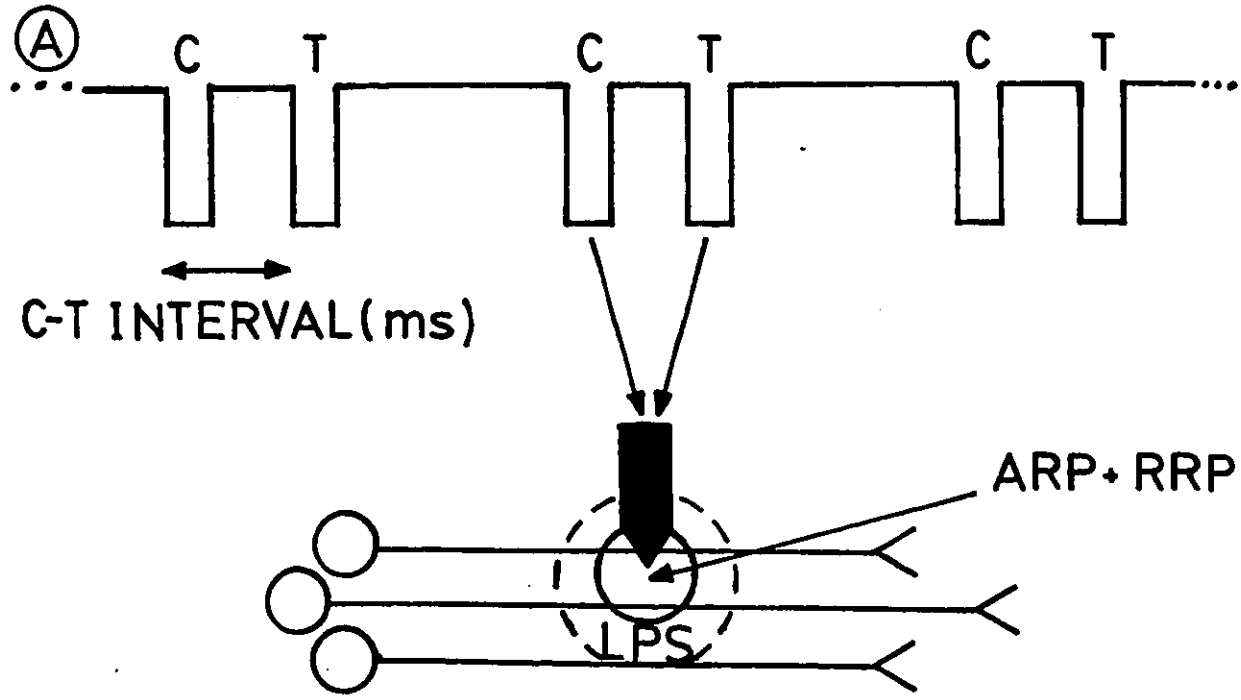
Figure 3. A) This figure shows the configuration of double-pulse stimulation in the refractory period test. Both C and T pulses are applied to the same electrode. Each C pulse is followed by a T pulse of the same amplitude. By varying the interval between C and T pulses and subsequently comparing the frequency threshold required for trains of double pulses to that for single pulses (T pulses absent), one can infer the range of recovery from refractoriness. In this design, because the C and T pulses are of the same amplitude, one cannot distinguish the different contributions of the absolute and the relative refractory periods. When the fibers at the fringe of the stimulation field are stimulated by trains of pairs of double pulses in close succession, local potential summation is observed.

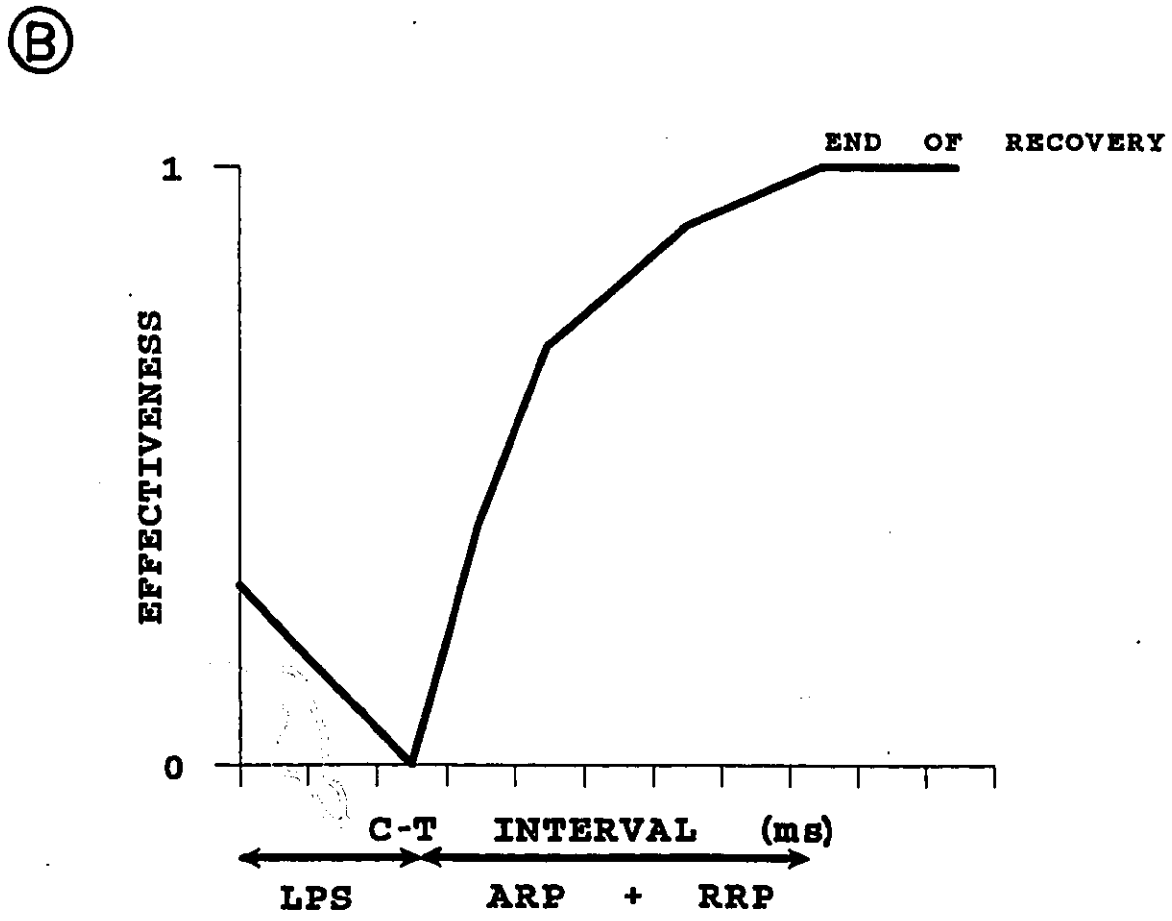
B) This figure illustrates a theoretical refractory period curve. The initial rise in effectiveness at lower C-T intervals reflects the contribution of local potential summation, and the gradual rising portion of the curve, the combination of neurons in the absolute refractory period and those in the relative refractory period. Recovery is believed to be completed when effectiveness values of nearly 1 are obtained, which occurs at the longer C-T intervals.

LPS: Local potential summation

ARP: Absolute refractory period

RRP: Relative refractory period


A
C
T
C
T
C
T
C-T INTERVAL(ms)
ARP+RRP
LPS


B
END OF RECOVERY
1
EFFECTIVENESS
0
C-T INTERVAL (ms)
LPS
ARP + RRP

period of the stimulated neurons, each T pulse is just as effective as each C pulse. Thus, the number of pulse pairs required to support criterion responding should be half the number of single pulses, and the ratio between the two should result in an effectiveness value of 1. When the C-T interval is less than the refractory period, additional pulse-pairs have to be added in order to maintain criterion responding. In that case the T pulses are ineffective, so that the number of pulse-pairs should be equal to the number of single pulses, resulting in an effectiveness value of 0. At intermediate C-T intervals, the proportional effects of the T pulses relative to the C pulses are seen, resulting in effectiveness values that fall between 0 and 1. According to the minimal model (Gallistel et al., 1981), the same integrator output is maintained at all C-T intervals by modifying the number of pulse-pairs. As the C-T interval decreases, additional pulse-pairs are added to replace firings lost due to refractoriness.

By modifying the above paradigm, other components of neural excitability can be detected, such as local potential summation, absolute refractory period, relative refractory period, as well as the supernormal and subnormal periods. The first of these phenomena, local potential summation, is seen in most refractory period curves. It occurs as a result of the summation of two subthreshold excitations produced by the C and T pulses in the subliminal fringe of the effective stimulation field. This takes place when the T pulses are delivered before the membrane

potential has had a chance to return to its resting value, a phenomenon that is only observed at short C-T interval (Yeomans, 1979; 1990a). The activation of fibers immediately outside the effective stimulation field gives rise to effectiveness values above 0. In addition, the increase in effectiveness values at low C-T intervals may be due to recovery of neurons within the effective stimulation field that are characterized by very short absolute refractory periods. These may be in their relative refractory period phase when the T pulses arrive immediately after the effects of the C pulses have dissipated (Yeomans, 1975; 1979). These two phenomena, local potential summation and short absolute refractory periods, may contribute to the elevated effectiveness values observed at short C-T intervals.

It has been hypothesized that the magnitude of the local potential summation depends on the location of the electrode tip within the reward substrate and the current intensity (Yeomans, Mercouris, and Ellard, 1985). In a homogenous population, when the electrode is located at the fringe of densely packed reward-relevant fibers, local potential summation effects will be larger than if the electrode is located more centrally in a bundle of less densely packed fibers. Similarly, increasing the current appears to diminish local potential summation effects. This is because the increase in current enlarges the effective stimulation field, thus recruiting those neurons which were previously in the subliminal fringe of the stimulation field. At the level of the MFB, local potential summation effects are seen

at or below C-T intervals of 0.4 ms (Yeomans, Matthews, Hawkins, Bellman, and Doppelt, 1979).

When local potential summation effects have dissipated, the absolute refractory period effects become apparent. During this period the neurons stimulated by the C pulses are completely unresponsive to any additional stimulation, which is reflected by effectiveness values of 0. Ideally, the absolute refractory period estimate corresponds to the longest C-T interval at which the effectiveness value is equal to 0. In most studies, however, the decay of local potential summation and the absolute refractory period overlap, preventing precise estimation of the absolute refractory period.

In the relative refractory period, the neurons stimulated by the C pulses will fire only if the T pulse current is of sufficient strength. Those neurons closest to the electrode tip will fire again as soon as the T pulses fall within their relative refractory period; those further away will fire later at the end of their relative refractory period. A refractory period curve demonstrating a gradual increase in recovery (the slope of the rising portion of the curve), might indicate that neurons with long refractory periods are being stimulated. Alternatively, it may also suggest that a heterogenous population of fibers with a wide range of absolute refractory periods is being excited.

Two other effects, the supernormal period, representing hyperexcitability of the stimulated neurons, and the subnormal

period, representing hypoexcitability of these fibers, might contribute to the effectiveness values at long C-T intervals, when the effects of the relative refractory period have dissipated. Although little is known about these phenomena, it is believed that the occurrence of these two periods depends on the repetitiveness of the stimulation, as well as the size of the neurons being stimulated (Bielajew and Fouriezos, 1985; Yeomans, 1990a).

In refractory period tests employing C and T pulses of equal current, the contribution of absolute and relative refractory periods cannot be distinguished. If, however, smaller current C pulses are used in combination with higher T pulse current, the effects of local potential summation and absolute refractory period can be observed. By comparing the results obtained for the unequal pulse condition to that of equal current C and T pulses, absolute refractory period effects can thus be determined. If larger current C than T pulses are employed, the effects of local potential summation are diminished thus allowing a better determination of the absolute refractory periods (Miliaressis, 1981). In addition, the contribution of relative refractory period as well as the supernormal period can be observed. Once again, by comparing the unequal pulse condition, this time one in which the C pulse current is higher, to the equal pulse condition, the effects of the supernormal period can be determined (Bielajew and Fouriezos, 1985; Miliaressis, 1981b; Miliaressis and Rompré, 1980; Yeomans, 1990b). The results of

such studies suggest that the relative refractory period does not appear to make a significant contribution to the excitability cycle, at least at the level of the MFB (Bielajew, Lapointe, Kiss, and Shizgal, 1982; Yeomans, 1979). It is also important to mention that the estimates of refractory periods are related to fiber diameter and the degree of myelination. Large, myelinated axons have shorter refractory periods (Swadlow and Waxman, 1978; Szabo, Lenard, and Kosaras, 1974; Waxman and Bennett, 1972) than smaller unmyelinated ones.

The behaviourally derived refractory period range is estimated by examining the interval at which the curve just begins to rise (beginning of recovery) and the interval at which the curve levels off (end of recovery). For example, it has been found that the refractory periods for stimulation-induced feeding (Gratton and Wise, 1988a; Hawkins, Puerto, Roll, and Yeomans, 1983), exploration (Rompré and Miliaressis, 1980), stimulation-induced escape (Skelton and Shizgal, 1980), and self-stimulation at the level of the lateral hypothalamus, ventral midbrain, periaqueductal gray, and ventral tegmentum (Bielajew and Shizgal, 1982; Bielajew et al., 1981; MacMillan, Simantirakis, and Shizgal, 1985; Miliaressis, 1981b), have similar refractory periods, ranging from 0.4 to 2.0 ms. In contrast, there is less overlap in the absolute refractory periods obtained for circling and self-stimulation at the level of the lateral hypothalamus and median raphe (Miliaressis and Rompré, 1980).

The behavioural estimates of refractoriness are strengthened

by the information provided from electrophysiological studies aimed at measuring the refractoriness of individual cells. For example, it has been determined that a population of descending MFB neurons, arising in the septum and diagonal band of Broca, have estimates similar to those usually found for MFB reward neurons using behavioural methods (Rompré and Shizgal., 1986; Shizgal, Schindler, and Rompré, 1989).

Longer estimates of recovery have been obtained for self-stimulation sites in the central gray (Bielajew et al., 1981), dorsal diencephalon (Bielajew and Fouriezos 1985; Vachon and Miliaressis, 1994), prefrontal cortex (Schenk and Shizgal, 1982; Trzcińska and Bielajew, 1992), CPu (Trzcińska and Bielajew, 1992), ventral pallidum (Panagis, Spyraki, and Miliaressis, 1995b), and anterior basal forebrain (Fouriezos et al., 1987).

In summary, similar refractory periods indicate that similar size axons are stimulated, but reveal nothing about their trajectories. In order to assess whether any two sites with overlapping refractory period ranges share the same fiber bundle, the collision test must be employed.

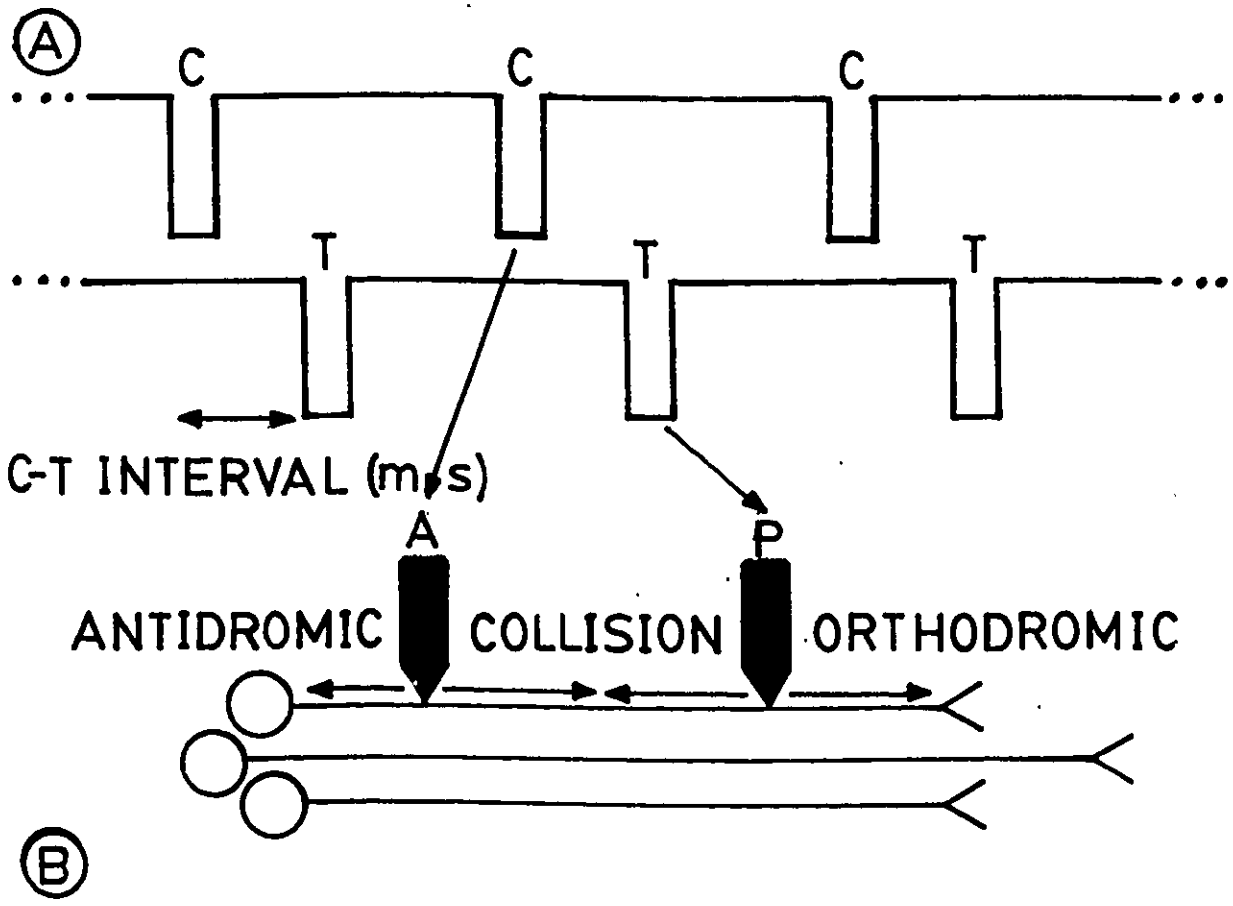
Collision technique

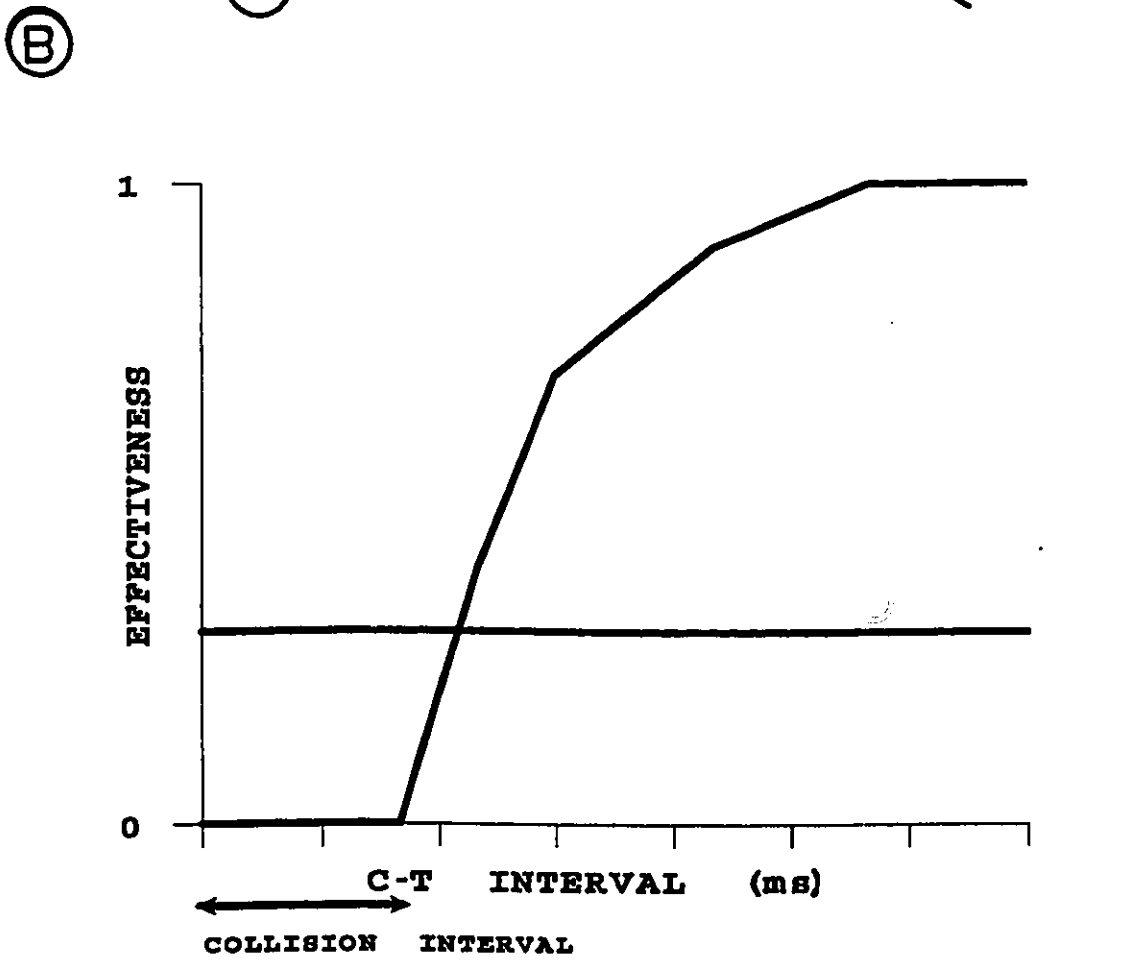
When the range of refractory periods of two separate regions that support self-stimulation overlap, one simple interpretation is that the two areas have common fibers. One way to assess this can be accomplished by using the collision technique, which is analogous to the refractory period method, except that the pairs

Figure 4. A) This figure shows the configuration of double-pulse stimulation used in the collision test. The trains of C and T pulses are applied to two different electrodes. In the anteroposterior (AP) condition, the C pulses are delivered to the anterior electrode and the T pulses are delivered to the posterior electrode. In the posteroanterior (PA) condition, the reverse occurs. Collision will occur if the antidromic action potential generated at one electrode collides with the orthodromic action potential generated at the second electrode, provided that the C-T interval is less than the sum of the inter-electrode conduction time and the underlying refractory period.

B) This part illustrates a theoretical collision curve. The flat portion of the curve overlapping the abscissa at the shortest C-T intervals is defined as the collision interval. During this phase, double-pulse stimulation is no more effective than single pulse stimulation, resulting in effectiveness values of 0. The rise in effectiveness, at progressively longer C-T intervals, indicates that double pulse stimulation is more effective than single pulse stimulation, resulting in effectiveness values between 0 and 1 (See the text for further detail).

The flat line, representing no change in effectiveness value as a function of C-T interval, is interpreted as the absence of a collision effect. However, because effectiveness values are greater than 0, it suggests that the signals generated at the two electrodes summate to some degree.


A
C
C
C
T
T
T
C-T INTERVAL (ms)
A
P
ANTIDROMIC
COLLISION
ORTHODROMIC


B
1
0
EFFECTIVENESS
C-T INTERVAL (ms)
COLLISION INTERVAL

of pulses are applied to two different electrodes instead of a single one. This double pulse technique was first employed by Lucas (1913) to study the effects of alcohol on the conduction velocity of the nerve-muscle preparation. The logic behind this method is based on the bidirectional nature of axonal propagation (refer to Figure 4A). Collision is inferred when action potentials conducting towards each other along the same fiber bundle collide, so that only the orthodromic action potentials reach the relevant terminals. This occurs when the interval between stimulation at each site (C-T interval) is less than the sum of the inter-electrode conduction time and the refractory period of the stimulated axonal bundle. This phenomenon is interpreted from an increase in the effectiveness of double-pulse stimulation at longer C-T intervals. Because axonal propagation is bidirectional, the shape of the plot that describes the effectiveness of double-pulse stimulation as a function of C-T interval should the same regardless of which electrode receives the C pulses. The longest inter-pulse interval at which collision occurs is defined as the collision interval.

A typical collision curve is illustrated in Figure 4B. An effectiveness of 0 indicates that double-pulse stimulation is no more effective than trains of single pulses, so that the frequency threshold of double pulses (C and T pulses) is identical to the frequency threshold of single pulses (T pulses absent). An effectiveness of 1 indicates that double-pulse stimulation is just as effective as trains of single pulses;

thus, the frequency threshold of double pulses is equal to half of the frequency threshold of single pulses, assuming that the two sets of pulses stimulate the same fiber bundle. By subtracting the refractory period estimate from the collision interval one obtains a measure of conduction time. Dividing the conduction time by the interelectrode distance gives an estimate of conduction velocity; no allowance is provided for the fact that the two regions may not be connected by a straight fiber bundle. Conduction velocity is known to be correlated with fiber diameter (Swadlow and Waxman, 1978). Large fibers conduct faster than smaller ones. Hence the calibre of first-stage neurons can be estimated. However, the shape of the collision profile provides no information about the direction of conduction, because as stated earlier, the fact that conduction is bidirectional gives rise to the same pattern of effectiveness values regardless of which electrode receives the C pulses. The collision method has unequivocally demonstrated direct axonal linkage between self-stimulation sites in the lateral hypothalamic and ventral tegmental areas (Bielajew, 1983; Bielajew and Shizgal, 1982; Durivage and Miliaressis, 1987; Gratton and Wise, 1988b; Malette and Miliaressis, 1995; Shizgal et al., 1980), and also between the lateral hypothalamic and lateral preoptic area (Bielajew, Thrasher, and Fouriezos, 1987b).

In order to assess whether ascending or descending signals participate in the collision-like effect obtained from concurrent lateral hypothalamic and ventral tegmental area stimulation,

Bielajew and Shizgal (1986) measured the effects of an anodal hyperpolarization block at each of the sites. Their results indicate that the potentials propagated along the fibers underlying MFB self-stimulation, at least at the level of the lateral hypothalamus and ventral tegmental area, are descending.

The collision method has also been used to characterize the substrates for other stimulation-evoked behaviours, such as escape (Bielajew and Shizgal, 1980), turning (Miliaressis and Philippe, 1983; Miliaressis and Rompré, 1980; Tehovnik and Yeomans, 1986, 1988; Yeomans and Buckenham, 1992; Yeomans and Tehovnik, 1988), startle (Frankland, Scott, and Yeomans, 1995; Yeomans and Cochrane, 1993; Yeomans et al., 1993a), feeding (Gratton and Wise, 1988b), and exploration (Durivage and Miliaressis, 1987). For example, when the lateral hypothalamus and ventral tegmental area electrodes were tested for self-stimulation and stimulation-escape, different patterns of double-pulse stimulation were obtained. These patterns were consistent with the notion that the directly stimulated substrates for aversion and reward are anatomically distinct (Bielajew and Shizgal, 1980). Likewise, different fibers appear to mediate self-stimulation and exploration. For example, collision was found for self-stimulation, but not for general motor activity at the same lateral hypothalamic and ventral tegmental sites (Durivage and Miliaressis, 1987). Similar collision effects have been found for stimulation-induced feeding and self-stimulation elicited from the lateral hypothalamus and ventral tegmental

area, suggesting that the directly stimulated fibers underlying both behaviours are the same (Gratton and Wise, 1988b). However, the size of the collision effect, as well as the length of the collision intervals differed for the two behaviours, casting some doubt as to the accuracy of their conclusions.

The absence of collision is harder to interpret: it indicates that either the two stimulated areas do not share the same fibers of passage, or that the electrodes are misaligned and hence activate different neurons along the same fiber bundle. When the collision test fails to produce a typical step-like profile, the effectiveness values observed at different C-T intervals are almost always of similar values, but greater than zero. This phenomenon is often referred to as summation of the rewarding effects obtained at two different loci. It suggests that the signals produced by two electrodes converge at some later stage in the reward circuity. For example, Bielajew et al. (1981) found summation between the lateral hypothalamus and central gray, but reported no evidence of direct axonal linkage between these two sites. Similarly, varying summation levels were found for the lateral hypothalamus and MPFC (Schenk and Shizgal, 1982), as well as for the MPFC and the anterior cingulate cortex (Silva, Vogel, and Corbett, 1982) self-stimulation sites.

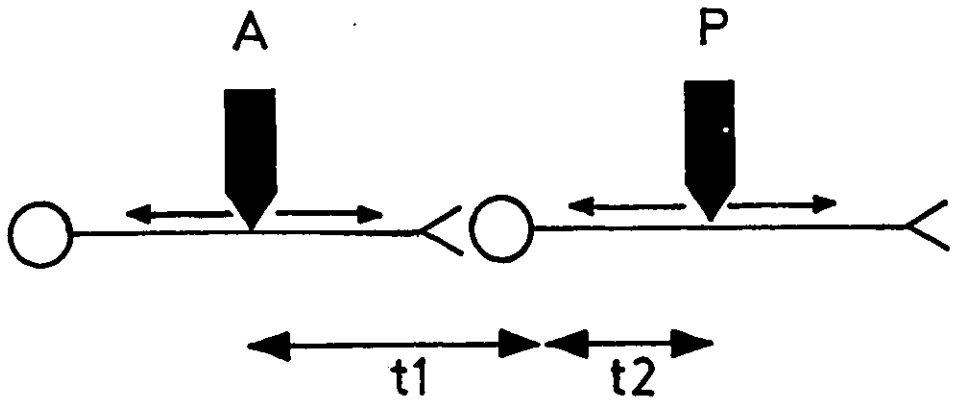
Transynaptic collision technique

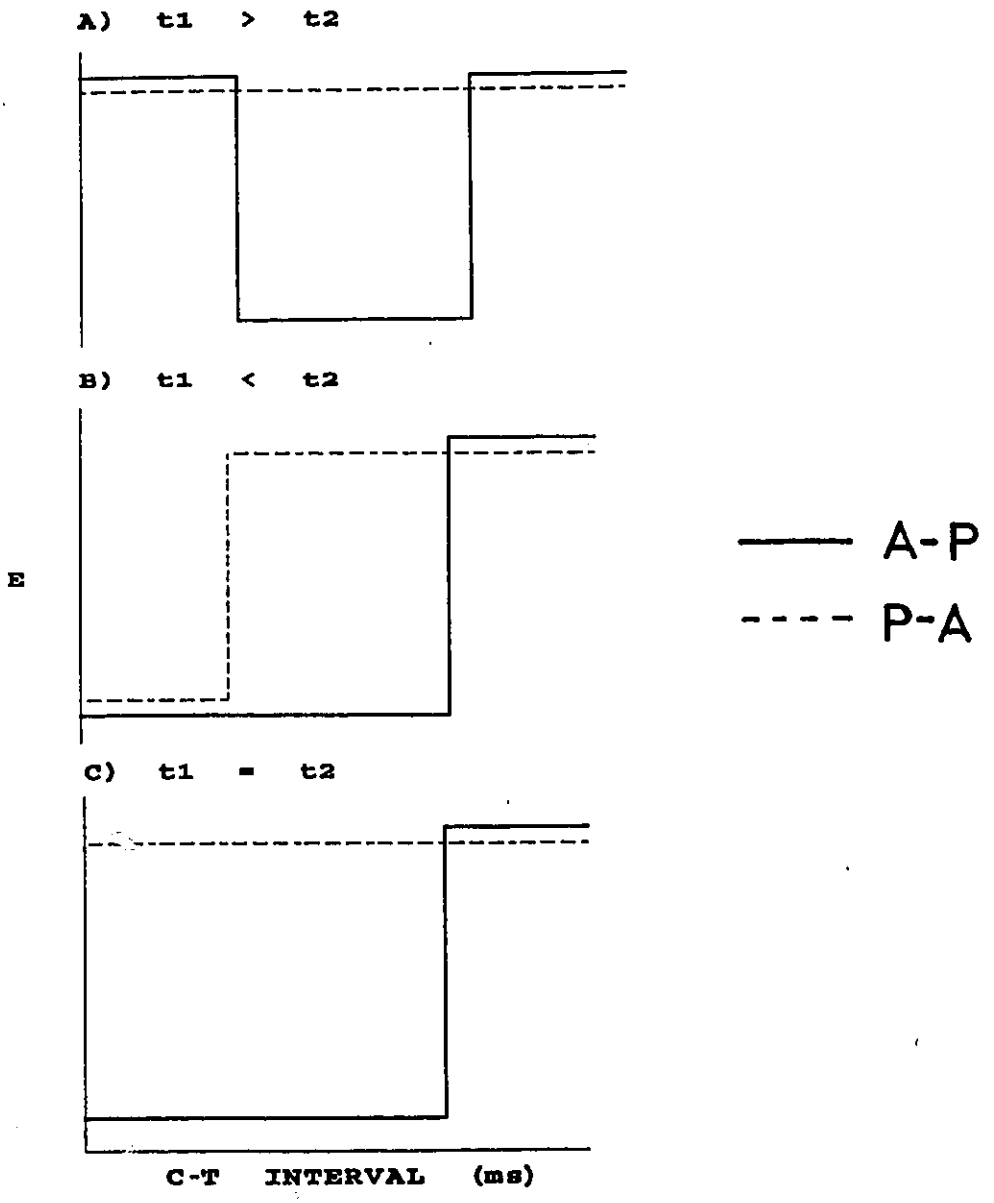
Occasionally, collision data do not match the models that account for axonal collision. To evaluate other patterns that

frequently arise from concurrent stimulation of two sites, a model of transynaptic collision was proposed by Yeomans (1990a; 1993a). This model was developed to explain a transynaptic anatomical arrangement, that is when two electrodes are located on two different fiber bundles that are synaptically linked. The result of such an electrode arrangement gives rise to asymmetric collision effects, since impulse conduction across synapses is unidirectional.

This two-neuron model of transynaptic collision is illustrated in Figure 5. In the first scenario (Figure 5A), when t1 > t2, the C pulses are applied to electrode A; collision will occur when the following three conditions are met - 1) the C-T interval exceeds the difference between t1 and t2 (pre- and post-synaptic conduction times), 2) the C-T interval is less than the time needed for the C pulses to travel past electrode P, and 3) the C-T interval is less than the refractory period under electrode P. Such conditions would result in maximum effectiveness value before and after the collision interval. Collision will not occur when the C pulses are applied to electrode P, since the T pulses sent to electrode A cannot be delivered at short enough C-T interval to stimulate the post-synaptic neuron before the antidromic impulse generated at electrode P has dissipated. In such cases, effectiveness values will always be at their maximum. In the second scenario (Figure 5B), when t1 < t2, the C pulses are applied to electrode A; collision will occur when the following two conditions are met; 1) the time for the orthodromic

Figure 5. This figure shows the three collision profiles predicted by the transynaptic collision model. The top of the figure shows the arrangement where a synapse is positioned between the two electrodes (A and P); t1 (pre-synaptic conduction time) represents the time for the orthodromic signal to travel from A to the initial segment of the post-synaptic neuron; t2 (post-synaptic conduction time) represents the time for the antidromic signal to travel from P to the cell body of the first neuron. The three different profiles are obtained by changing the timing relationship between t1 and t2.


A
P
t1
t2


A) t1 > t2
B) t1 < t2
C) t1 = t2
E
A-P
P-A
C-T INTERVAL (ms)

impulse from electrode A to travel past electrode P exceeds the C-T interval, 2) the membrane under electrode P has sufficient time to recover from refractoriness. In such cases, longer collision interval would be observed than in the first example. When the C pulses are delivered to electrode P, collision can still be observed, but only when the C-T interval is smaller than the difference between the pre and post-synaptic conduction times. In such cases, a shorter collision interval will be observed, resembling an axonal collision effect. In the final scenario (Figure 5C), when t1 = t2, the C pulses are applied to electrode A; collision will occur at every C-T interval that is smaller than the conduction time of the orthodromic signal generated at electrode A to pass electrode P, and when the membrane under electrode P has recovered from refractoriness. When the C pulses are delivered to electrode P, no collision effect will be seen, because the T pulses, sent to electrode A, will never have time to reach the post-synaptic neuron before the effects of the antidromic impulse produced at electrode P have dissipated, regardless of the C-T interval.

Results consistent with the transynaptic collision model have been reported for startle-like responses evoked by electrical stimulation of the reticular formation (Yeomans et al., 1990a), and also for combined lateral preoptic and ventral tegmental area self-stimulation (Bushnik-Harris and Bielajew, 1993).

Other models of collision involving multisynaptic mechanisms have been described for sites along the MFB (Malette and

Miliaressis, 1995) and suggest that many reward neurons contained within the MFB send axon collaterals to the other hemisphere. The collision and transynaptic collision-like profiles indicate that each electrode stimulates a different branch of the same reward neuron and that conduction failure occurs at the point where the two branches meet (For an illustration, refer to Figure 13 of Malette and Miliaressis, 1995).

A glimpse into the MFB reward substrate

The MFB is composed of about 50 different pathways, with some of the densest projections coursing from the olfactory tubercle to the ventromedial part of the mesencephalic tegmentum, traversing the lateral preoptic area and lateral hypothalamus and terminating at the level of the medulla oblongata (Nieuwenhuys, Geeraedts, and Veening, 1982; Olds and Olds, 1963); only some of these trajectories are thought to mediate brain stimulation reward.

From collision studies in the MFB, inferences about the properties of reward sites suggest that the neurons are small (0.5-2.0 μm in diameter), myelinated, have low thresholds of excitability, as reflected in the current-distance constant which is estimated to range between 1000 to 8000 $\mu A/mm^2$ (Fouriezos and Wise, 1984; Milner and Laferrière, 1982); they appear to have absolute refractory periods that range from 0.4 to 1.5 ms (Bielajew et al., 1981; Rompré and Miliaressis, 1980; Szabo et al., 1974; Yeomans, 1979), and their signals conduct rostro-

caudally at velocities between 1 and 8.3 m/s at the level of the lateral hypothalamus and ventral tegmental area (Bielajew, 1983; Bielajew and Shizgal, 1986; Bielajew et al., 1987b; Shizgal et al., 1980), and between 0.24 and 11 m/s more rostrally between the lateral hypothalamic and lateral preoptic areas (Bielajew et al., 1987b).

Other approaches have also proven to be useful in characterizing the MFB reward substrate. For example, single-cell recording studies have shown that there are cells in a variety of self-stimulation loci, particularly in the lateral and medial preoptic areas, ventral pallidum, and septal region that are activated by lateral hypothalamic stimulation. In addition, the refractory periods of these cells have values similar to those which have been determined in behavioural tests in the lateral hypothalamus and ventral tegmental area (Shizgal et al., 1989). From metabolic studies employing [^{14}C]-2-deoxyglucose, it appears that self-stimulation of the anterior ventral tegmental area induces activation of the midhypothalamic regions, vertical limb of the diagonal band of Broca, compartment "c" of the MFB, the bed nucleus of stria terminals, the medial preoptic area, and suppression of activity in the lateral habenula. Similarly, in a study using a different metabolic marker, cytochrome oxidase, stimulation of the lateral hypothalamus with short pulse durations resulted in a marked increase in metabolic activity of the lateral septal nucleus and nucleus accumbens, whereas longer pulse durations produced activation in the dopaminergic

projections that target the frontal cortex, olfactory tubercle, lateral habenula, and septal nucleus (Bielajew, 1991). These results suggest that the mesocorticolimbic regions may be involved in MFB self-stimulation.

Although there has been much progress in delineating the electrophysiological characteristics of the MFB reward substrate, the brain regions anterior to this bundle have been studied in less detail.

EXTRA MFB SITES

Anatomy

Prefrontal cortex

The prefrontal cortex has classically been defined as that area of the frontal lobes which receives afferents from the mediodorsal thalamic nucleus (Beckstead, 1979; Krettek and Price, 1977; Leonard, 1969). In the rat the prefrontal cortex comprises five main regions: the anterior cingulate area (Cg1, Cg2, Cg3), the infralimbic area (IL), the orbital area (LO, VLO VO, MO), the ventral and dorsal agranular insular areas, also called the sulcal cortex (AIV, AID), and the "shoulder" of the precentral cortex (Fr2) (Zilles and Wree, 1985). The area that is often referred to as the MPFC, or the prelimbic cortex, includes the IL and Cg1-3 regions (Robertson 1989).

The prefrontal cortex constitutes a predominant source of topographically organized (in the dorsal-ventral and rostral-caudal planes), ipsilateral and bilateral projections to the

basal ganglia (including the caudate-putamen), (Beckstead 1979; Brutkowski 1965; Divac, Kosmal, Björklund, and Lindvall, 1978; McGeorge and Faull, 1987; Routtenberg, 1971; Sesack, Deutch, Roth, and Bunney, 1989; Wilson, 1987). These corticostriatal projections traverse the striatum in bundles and terminate in the internal capsule (Ferino, Thierry, Saffron, and Glowinski, 1987). Specifically, the dorsal anterior cingulate cortex (Cg1) projects to the dorsal striatum, and the prelimbic cortex (Cg3 and IL regions) projects to the ventromedial striatum (Sesack et al., 1989); however, in contrast to other cortical connections, these do not appear to be reciprocal (Fuster, 1980; Kolb, 1990; McGeorge & Faull, 1989; Nauta & Domesick, 1984; Veening, Cornelissen, and Lieven, 1980), with the exception of some striatocortical fibers, especially in the middle third of the putamen and the ventrolateral caudate putamen (Divac, Mogensen, Marikovic, and Martensson, 1987; Jayaraman, 1980; Purpura, Housepian, and Grundfest, 1958). It appears that these striatocortical fibers are small clustered collaterals of the striatothalamic neurons (Jayaraman, 1980).

Besides the striatal inputs, the prefrontal cortex also sends projections to the mediodorsal thalamic nucleus, nucleus accumbens, amygdala, lateral septum, MFB, pons, and the autonomic regions of the brainstem (Beckstead, 1979; Krettek & Price, 1977; Leonard, 1969). The prefrontal cortex afferents, other than the mediodorsal thalamic nucleus, include the lateral hypothalamus, ventral tegmental area, amygdala, claustrum, substantia nigra and

the hippocampus (see Fuster, 1980 and Kolb and Tees, 1990).

Anatomically distinct regions of the MPFC subserve different functions. For example, areas Cg1 and Cg2 seem to be somatic and motor, whereas the Cg3 area appears to be limbic in function (Zeng and Stuesse, 1991). The limbic system can be defined as a complex system of nuclei and pathways that are involved in the expression of instinct and mood (fear, rage, pleasure), in the activities of the endocrine, autonomic, and motor systems, in all the body's activities related to self-preservation and preservation of the species, and in memory processes (Kandel and Schwartz, 1985). Lesion studies further support the argument that the dorsolateral prefrontal cortex is mainly involved in cognitive (non-limbic) aspects of behaviour. The medioventral prefrontal cortex, on the other hand, seems to be implicated in motivational and affective functions, as well as in the maintenance of balance between internal and external influences on purposive behaviour (see Fuster, 1980).

Caudate-putamen

One of the major components of the basal ganglia - the corpus striatum - comprises both the CPu and the globus pallidus (Heimer, Alheid, and Zaborszky, 1985). The CPu is often referred to as the striatum or neostriatum. In the rat the caudate nucleus and putamen are practically indistinguishable (Heimer et al., 1985). The putamen receives inputs from the sensory-motor cortex, whereas the caudate is more related to the association

cortex, including the prefrontal area (Graybiel, 1984). Some studies argue that the corpus striatum of the rat also includes the nucleus accumbens and olfactory tubercle (Heimer, 1978; Newman and Winans, 1980). Afferents to the CPu are made up of predominantly very small unmyelinated fibers (less than 1 μm in diameter) and derive from practically all cortical areas, the thalamus (particularly the intralaminar nuclei), basolateral amygdala, substantia nigra, globus pallidus, subthalamic nucleus, ventral tegmental area, pedunculopontine tegmental nucleus, and the raphe nuclei (see Heimer et al., 1985; Kitai, Koscis, Preston, and Sugimon, 1976). The striatum projects only to the globus pallidus, entopeduncular nucleus, and substantia nigra par reticulata (Parent, 1990; Staines, Nagy, Vincent, and Fibiger, 1980).

Striatal neurons form a characteristic patch/matrix mosaic. Patches are rich in enkephalin, substance P, and opiate receptors, but poor in acetylcholinesterase (Herkenham and Pert, 1981). The matrix, on the other hand, is acetylcholinesterase rich. The dorsal striatum has a smaller patch/matrix ratio than the ventral striatum (Desban, Kemel, Glowinski, and Gauchy, 1993). The patch/matrix organization has been linked with both limbic and non-limbic functions, based on inputs to the patches from such limbic areas as the amygdala and the prelimbic cortex, and to the matrix from sensorimotor cortical regions (Gerfen, 1992; White, 1989). The patch-matrix organization of the striatum is related to the laminar organization of the cortex

(Gerfen, 1989). Deep layers of the prelimbic cortex project to the patches and the superficial layers project to the matrix (Gerfen, 1984). Furthermore, limbic regions of the MPFC, Cg3, and IL project to the patches, whereas other cortical areas project to the matrix (Gerfen, 1984). The ventral striatum is believed to be part of the limbic, anterior cingular circuit (Alexander, DeLong, and Strick, 1986). For example, electrical stimulation of the CPu in the monkey has been shown to produce analgesia (Lineberry and Vierck, 1975). Also, it appears that ventral striatum mediates opiate-induced feeding (Bakshi and Kelley, 1993). The limbic-ventral striatum circuity is retained in the dorsal striatum, but only in its connections to the patchy compartment (Gerfen, 1992). The dorsal striatum matrix receives input from sensory and motor cortical areas (neocortex) and projects to the globus pallidus, substantia nigra pars reticulata, and the ventral anterior thalamus (Nauta and Domesick, 1984). The dorsal striatum patches receive input from the MPFC and project to the substantia nigra pars compacta (Gerfen, 1984). The ventral striatum patches receives limbic inputs from the amygdala and hippocampus, and project to the ventral pallidum and mediodorsal thalamic nucleus (Gerfen, 1984; Mogenson, Jones, and Yim, 1980).

From the above anatomical description, it appears that the limbic regions of the MPFC and the CPu may form part of the same reward substrate. For example, lesions of the MPFC and CPu produce qualitatively similar deficits (Brutkowski, 1965), and

include motor symptoms, response inhibition, difficulties in temporal ordering, spatial disorientation, social and affective changes, lack of behavioural spontaneity, defective odour discrimination, hyperactivity, impaired habituation of many activities, homeostasis changes, and contralateral neglect (for more details see Kolb, 1984). When hyperactivity is present, the frontal lobectomy also affects the rostral part of the caudate nucleus (Davies, 1958). Generally, the effects of caudate lesions are similar to the consequences following frontal lesions, but are less severe (Brutkowski, 1965). Lesions of the striatum result in changes in species-specific behaviours, activity levels, and appetitive responses to physiological challenges (Iversen and Dunnett, 1990).

Forebrain self-stimulation

Cortical sites which support self-stimulation include the olfactory bulb (Phillips, 1970), prefrontal cortex, also called the posterior medial cingulate cortex (Routtenberg and Sloan, 1972), medial prefrontal cortex (Ferrer, Sanguinetti, Vives, and Mora, 1983; Ferrer, Cobo, and Mora, 1987), sulcal prefrontal cortex (Clavier and Gerfen, 1979; McGregor and Atrens, 1991), pyriform cortex (Prado-Alcala, Streather, and Wise, 1984), entorhinal cortex (Ott, Destrade, and Ruthrich, 1980) and medial entorhinal cortex (Reymann, Mulcko, Ott, and Matthies, 1986). In addition, self-stimulation can be obtained from non-cortical forebrain structures, such as a number of thalamic nuclei

(Bielajew and Fouriezos, 1985; Vachon and Miliaressis, 1992), nucleus accumbens (Fouriezos et al., 1987), CPu (Phillips, Carter, and Fibiger, 1976; Prado-Alcala et al., 1975; Routtenberg, 1971), and ventral pallidum (Panagis, Miliaressis, Anagnostakis, and Spyraki, 1995a).

Electrical stimulation of cortical structures produces a characteristic behavioural profile. Initially, a type of behavioural suppression or "freezing" is observed (Stamm, 1964). As the animal learns to bar press, this characteristic pose eventually subsides (Corbett, Laferrière, and Milner, 1982b; Corbett and Stellar, 1983; Justesen et al., 1963; Phillips et al., 1976; Wilcott, 1979). Some researchers suggest that MPFC stimulation has aversive and motor suppressive effects, which might explain the initial low response rates (McGregor, 1992; Miserendino and Coons, 1989). The arrest phenomenon is most prominent in anterior dorsolateral regions of the MPFC (Spence, Silverman, and Corbett, 1985; Wilcott, Sabol, and Yurcheshen, 1976). Interestingly, the same regions which support MPFC self-stimulation are also the ones that show the strongest inhibitory influences (Wilcott et al., 1976). These arrest reactions are probably indicative of epileptogenic activity, which co-occurs with many cortical self-stimulation sites (Balleine, McGregor, and Atrens, 1989; Zacharko et al., 1990). It appears that kindling-like mechanisms might underlie the development of self-stimulation in the MPFC. As the self-stimulation rates increase, so do the occurrence of seizures (Douglin and Glassman, 1979).

In addition, anticonvulsant agents (diazepam and phenobarbital) delay the development of MPFC self-stimulation, but are ineffective in altering self-stimulation once it has been established (Robertson, Laferrière, and Milner 1982b). Similarly, diazepam appears to alter self-stimulation thresholds by inhibiting competing epileptogenic activity, but has no effect of thresholds obtained from non-seizure sites (Harris and Bielajew, 1991). Further, subconvulsive stimulation retards the acquisition of delayed alteration task, but has no effect on performance of trained animals (Stamm, 1964). Indeed, an attempt to employ brotizolam in the experiments described in this thesis, in the hope of reducing seizures and promoting acquisition of MPFC self-stimulation failed to be effective. However, it has proven to be successful in decreasing seizure activity and severity in MFB self-stimulation sites (Harris and Bielajew, 1990).

The characteristics of forebrain reward sites seem to differ in a number of ways from those of the MFB. For example, the acquisition of cortical self-stimulation is slower and often requires several shaping sessions before it is fully manifested (Corbett et al., 1982b, Corbett, Silva, and Stellar, 1985; Douglin and Glassman, 1979). It can be hastened, however, by the application of either contingent or non-contingent stimulation to the MPFC, although non-contingent stimulation appears to be more efficient (Balleine et al., 1989; Corbett, 1990; Corbett et al., 1982b), especially if a simpler task is required of the animal

(nose-poke vs bar pressing) (Corbett et al., 1985). In addition, this facilitatory effect of non-contingent stimulation on subsequent MPFC self-stimulation acquisition is blocked by ketamine, a NMDA receptor antagonist (Corbett, 1990). Acquisition of MPFC self-stimulation can also be facilitated by prestimulation of the lateral hypothalamus and sulcal prefrontal cortex, but not of the hippocampus or entorhinal cortex (Cobo, Ferrer, and Mora, 1989; Corbett et al., 1985; Robertson et al., 1982a; Robertson, Laferrière, and Milner, 1986). In addition, MPFC self-stimulation is characterized by an absence of the arousal which usually accompanies MFB stimulation, and response rates for MPFC stimulation are typically lower than those observed for MFB stimulation (Schenk and Shizgal, 1982; Yadin, Guarini, and Gallistel, 1983; Zacharko et al., 1990). Furthermore, the currents needed to evoke MPFC self-stimulation are higher, possibly indicating the recruitment of smaller, less excitable, and more sparsely distributed reward-relevant fibers (Nassif, Cardo, and Libersat, 1985; Schenk et al., 1985).

Behaviourally derived refractory periods from forebrain structures seem to be longer than the ones obtained from the MFB (Bielajew and Fouriezos, 1985; Fouriezos et al., 1987; Schenk and Shizgal, 1982; Trzcińska and Bielajew, 1992), and range roughly from 1.0 to 10.0 ms. In addition, collision studies fail to demonstrate evidence of an axonal linkage between the MPFC and the MFB, although some integration of their reward signals has been documented (Schenk and Shizgal, 1982).

The [^{14}C]-2-deoxyglucose autoradiography patterns seem to strengthen the above arguments, and demonstrate that electrodes implanted in the MFB and MPFC activate different neural systems (Yadin et al., 1983). In addition, unilateral electrical stimulation of the MFB, at the parameters that typically produce self-stimulation, fail to induce metabolic changes in both the MPFC and CPu (Gallistel, Gomita, Yadin, and Campbell, 1985; Yadin et al., 1983). Finally, MPFC self-stimulation is not affected by lesions of the MFB, lateral hypothalamus, locus coeruleus, zona incerta, basolateral amygdala, mediodorsal thalamic nucleus, nucleus accumbens, entorhinal cortex, or sulcal prefrontal cortex (Cobo et al., 1989; Corbett et al., 1982c; Ferrer et al., 1983, Ferrer, Myers, and Mora, 1985; Mora and Ferrer, 1986), but is so by destruction of the external capsule, rostral claustrum and lateral neostriatum (Cobo et al., 1989; Vives, Morales and Mora, 1986). Indeed, rats with bilateral lesions of the cortex, hippocampus, striatum, and septum do learn to bar press for lateral hypothalamic stimulation (Huston and Borbely, 1973, 1974). These findings suggest that separate reward systems underlie forebrain and MFB self-stimulation. However, the drawback of these latter studies is that response rates have often been used as a measure of the rewarding effects of stimulation. Corbett et al., (1982c), using threshold as an indication of change in reward magnitude, showed that bilateral knife cuts interconnecting the medial and sulcal cortices eliminated MPFC self-stimulation with no apparent recovery over

21 days. In addition, the results of kainic acid lesions, which destroy cell bodies and spare fibers of passage, indicate that cell bodies within the MPFC seem to be essential in maintaining the rewarding properties of MPFC electrical stimulation (Ferrer, et al., 1985). The same animals showed normal self-stimulation rates in the contralateral MPFC, the hemisphere that was not affected by kainic acid.

The data summarized here offer the possibility that multiple connections between several structures, rather than single reciprocal ones between the MPFC and other areas seem to form the neural network underlying MPFC self-stimulation (Cobo et al., 1989).

The role of the CPu in reward has not been thoroughly investigated. The striatum has been considered to be part of the motor system, because stimulation of this region often produces contralateral head and body movements, full circling, and flopping (German, Dalsass, and Kiser, 1980; Phillips et al., 1976). Electrical stimulation of the CPu occasionally results in the same behavioural suppression as that observed in the MPFC (Justesen et al., 1963). In terms of its role in brain stimulation reward, it has been considered to be neutral (Prado-Alcala et al., 1975), although a few medial anterior sites have been found to support self-stimulation (Phillips et al., 1976; Prado-Alcala et al., 1984; Routtenberg, 1976). One suggestion has been that the ventromedial part of the striatum, including the CPu, is mainly limbic and thus related to reward, while the

dorsolateral part underlies motor and cognitive tasks (McGeorge and Faull, 1989; Nauta and Domesick, 1984; Robbins, Cador, Taylor, and Everitt, 1989). This conclusion is strengthened by the results of electrophysiological recordings of cells in these areas (Schultz, Apicella, Scarnati, and Ljungberg, 1992); for example, cells in the ventral striatum of Macaca monkeys respond to the expectation of reward in a delayed go-no-go task, which is evidenced by a brief burst in striatal activity prior to reward delivery.

A glimpse into the pharmacology of the forebrain

It appears that dopamine, acetylcholine, substance P, but not norepinephrine, serotonin, cholecystokinin, or enkephalins are somehow involved in MPFC self-stimulation (Corbett, 1992; Ferrer, Cobo, and Mora, 1988; Ferron, Thierry, LeDouarin, and Glowinski, 1984; Herkenham, 1987; Mora, 1978; Vives, Gayoso, Osorio, and Mora, 1983; Vives and Mora, 1986). Among these neurotransmitters, the catecholaminergic system shows the most striking anatomical relationship with points of self-stimulation along the MPFC (McGregor, Balleine, and Atrens, 1989). Forebrain estimates of behaviourally derived refractory period (Bielajew and Fouriezos 1985; Fouriezos et al., 1987; Schenk and Shizgal, 1982; Trzcińska and Bielajew, 1992; Vachon and Miliaressis, 1994) seem to match those values obtained electrophysiologically (Deniau, Thierry, and Feger, 1980; German et al., 1980; Lindvall and Björklund, 1974; Moore and Bloom, 1978; Wang 1981; Yim and

Mogenson, 1980), and behaviourally (Yeomans et al., 1988; Yeomans, 1990a) for dopaminergic fibers.

Given ample anatomical evidence for corticostriatal connections, it is not surprising that dopamine appears to be critical in the transmission of information from the cortex to the striatum (Graybiel, 1990). For example, electrical stimulation of the MPFC seems to increase dopamine release in the striatum (Taber and Fibiger, 1993). Dopaminergic innervation is most dense in the motor and anterior cingulate cortex, and shows progressively decreasing density in the rostral to caudal direction (Berger, Gaspar, and Verney, 1991; Sesack and Pickel, 1992). Dopamine neurons can be divided into a dorsal (A10 and dorsal A9), and ventral tier (substantia nigra pars compacta and substantia nigra pars reticulata), (Gerfen, 1989). The MPFC and ventral striatum both receive dopaminergic innervation from the A10 cell group in the mesencephalon, including the ventral tegmental area (Lindvall and Björklund, 1974, 1983). The dorsal striatum receives afferents from the A8 and A9 regions (substantia nigra) (Ungerstedt, 1971). However, 6-OHDA lesions of the A9 and A10 dopaminergic cell group fail to produce a long-lasting loss of MPFC self-stimulation (Corbett et al., 1982b; Ferrer et al., 1983; Gerfen and Clavier, 1981; Phillips and Fibiger, 1978; Simon, Stinus, Tassin, Lavielle, Blanc, Thiery, Glowinski, and LeMoal, 1979; Vives et al., 1986), and only transiently disrupt CPu self-stimulation (Phillips et al., 1976). It has been suggested that the ventral striatum, including the

nucleus accumbens, may form an interface between the limbic system and the extrapyramidal motor system (Voorn, Jorritsma-Byham, Van Dijk, and Buijs, 1986). The hypothesis states that the CPu and the nucleus accumbens act as filters of signals from the limbic part of the cerebral cortex (Mogenson et al., 1980). These filtering mechanisms are influenced by the nigrostriatal and mesolimbic dopamine system. It is generally agreed that the nigrostriatal dopaminergic bundle may be important for motoric functions, whereas the mesolimbic/mesocortical dopamine system may be necessary for the motivational aspects of CPu and forebrain self-stimulation (Zacharko et al., 1990). For example, pharmacological and electrical activation of the mesolimbic dopamine system appears to facilitate feeding and sexual behaviours, and exposure to feeding and sex-related stimuli increases activity within the mesolimbic dopamine system (Joseph and Hodges, 1990; Mitchell and Gratton, 1992). Other studies show that bar pressing for food in food-deprived animals, or simply feeding itself, can enhance dopamine release and turnover in the prefrontal cortex, CPu, and nucleus accumbens (Beninger, D'Amico, and Ranaldi, 1993; Hernandez and Hoebel, 1990). Dopaminergic transmission in the cerebral cortex and ventral striatum appears to be essential for sexual arousal, since dopamine increases are observed in these structures during copulation in hormone-primed rats (Everitt, 1990; Pfaus, Kleopoulos, Mobbs, Gibbs, and Pfaff, 1993).

Additional evidence pointing to the fact that the mesolimbic

dopamine system seems to be important for the motivational aspects of behaviour comes from lesion studies, showing that the thirst-induced motivational and emotional arousal are abolished by 6-OHDA lesions of the mesolimbic but not nigrostriatal dopamine system (Papp and Bal., 1987). Certain environmental stressors, such as footshock (either escapable or inescapable), which activate the mesocortical dopamine system, enhance both MPFC and CPu self-stimulation, while having no effect on lateral hypothalamic self-stimulation (Abercrombie, Keefe, DiFrischia, and Zigmond, 1989; Balleine, 1991; Dunn, 1988; Dunn and File, 1983; Herman, Guillonneau, Dantzer, Scatton, Semerdjian-Rouquier and LeMoal, 1982; McGregor et al., 1989). Moreover, there is a correlation between the location of self-stimulation sites and dopaminergic innervation in forebrain structures (Prado-Alcala and Wise, 1984; Robertson and Mogenson, 1978).

The role of dopamine in reward processes is further demonstrated by the effects of dopaminergic agonists and antagonists on self-stimulation. For example, both cocaine and d-amphetamine enhance the rewarding value of MPFC self-stimulation (Goeders and Smith, 1983; Goeders, Dworkin, and Smith, 1986), as indicated by a decrease in thresholds (Corbett, 1991; Gallistel and Freyd, 1987; Spence et al., 1985), but these changes are apparent only during the acquisition stages (Robertson, Laferrière, and Franklin, 1981; Spence et al., 1985). Other testing paradigms, however, have failed to demonstrate an effect of dopamine agonists on reward. For example, amphetamine

injections into the structures containing dopamine terminals, such as the CPu and MPFC, are not effective in producing conditioned place preference (Carr and White, 1983, 1986). At the level of the MFB, pimozide and clonidine, two dopaminergic antagonists, cause an extinction-like change in runway performance (Gallistel, Boytim, Gomita, and Klebanoff, 1982), and hence is thought to block the rewarding effects of MFB stimulation (Gallistel, 1986). Lesions studies indicate that depleting dopamine levels in the frontal cortex do not abolish self-stimulation in the MPFC (Phillips and Fibiger, 1978).

Hence, although dopamine may not play a direct role in mediating the rewarding effects of MPFC and CPu stimulation, it nonetheless appears to have at least a modulatory function. It has been proposed that it acts transynaptically and may function permissively at the point at which the reward signal is converted to a memory trace (Gallistel and Freyd, 1987; Gallistel et al., 1982; Miliaressis et al., 1991).

RATIONALE

There is good anatomical evidence that the MPFC sends topographically organized projections to corresponding regions of the striatum, including the CPu (Beckstead, 1979; Brutkowski, 1965; Routtenberg, 1971). With respect to brain stimulation reward studies, it has been shown that the estimates of refractoriness for these two reward sites overlap, giving rise to suggestion that a subset of reward neurons are common to both areas (Schenk and Shizgal, 1982; Trzcińska and Bielajew, 1992). Hence, both the anatomical and the behavioural data indicate that the MPFC and CPu might be functionally connected.

Based on the anatomical, electrophysiological, and behavioural body of experimental evidence reviewed above, three main questions were addressed in this dissertation. First, a mapping study of the MPFC and CPu was conducted, in order to distinguish among sites which support self-stimulation from those that do not, and to define the boundaries and densities of the reward substrates using trade-off functions between current and frequency. Moveable electrodes (Kinetrods Reg'd) were employed to investigate consecutive ventral sites (Miliaressis, 1981a), thus allowing for the spatial evaluation of the reward substrate in these areas. One of the methodological constraints of the first experiment was the slow acquisition of MPFC self-stimulation or rates too low and variable to generate rate-frequency functions. It became evident that in those rats which

were first trained to bar press for CPu stimulation, that their subsequent performance for MPFC self-stimulation was characterized by higher and more stable rates, suitable for the determination of frequency thresholds. This observation led to a study designed to investigate whether other self-stimulation sites, such as the lateral hypothalamus and ventral tegmentum, also contributed to the development of MPFC self-stimulation.

Second, refractory period tests were conducted on a subset of sites evaluated in the first study, in order to determine the range of recovery from refractoriness in the MPFC and CPu, and to compare these values to the ones obtained for the MFB.

Last, the nature of the anatomical connectivity of nine ipsilateral and two contralateral CPu and MPFC pairs was evaluated with the behavioural adaptation of the collision test. In those cases where collision effects were observed, conduction velocities between the MPFC and CPu reward neurons were calculated.

GENERAL METHODOLOGY

Unless otherwise indicated, all three experiments were conducted as described below. Treatment of rats was in accordance with the guidelines of the Canadian Council on Animal Care and the university policies pertaining to animal experimentation, approved by the University of Ottawa Animal Care Committee.

Subjects and surgery

Sixty seven Long-Evans male rats (Charles River, Québec, Inc.) were housed either individually or in pairs in shoe-box style cages. They were kept on a 12 h light/12 h dark cycle, with light onset at 7:00 h. Purina rat chow and water were available ad libitum. Their weights ranged between 297 and 365 g at the time of surgery. Stereotaxic surgery was conducted under a combination of sodium pentobarbital (Somnitol 60 mg/kg, i.p.) and Rompun (1.0 mg/ml Xylazine, i.m.) to produce deep and long-lasting anaesthesia. Occasionally, supplemental injections of sodium pentobarbital were required in order to maintain adequate anaesthesia. Twenty minutes before surgery, atropine sulphate (0.6 mg/kg, s.c.) was given to reduce bronchial secretions. In case of respiratory distress, additional injections of atropine sulphate were administered during the surgical procedure itself, and infrequently the animal had to be placed in an oxygen-filled chamber for a few minutes. Each subject was implanted with one

of the following electrode assemblies - a single moveable electrode (Kinetrods Reg'd), a pair of electrodes, one fixed and one moveable, or a pair of moveable ones. The electrodes were made of 0.25 mm stainless-steel wire insulated with Epoxylite to the rounded tips. The stereotaxic coordinates were as follows: MPFC 1.7-5.2 mm anterior to bregma, 0.4-3.0 mm lateral to the midsagittal suture, and 1.0-5.1 mm below dura reading at Bregma; CPu 0.2-1.7 mm anterior to bregma, 1.4-2.6 lateral to the midsagittal suture, and 3.3-5.0 mm below dura; lateral hypothalamus 1.4-2.8 mm posterior to bregma, 1.7-2.0 mm lateral to the midsagittal suture, and 7.7-8.4 mm below dura; ventral tegmental area 4.8-5.3 mm posterior to bregma, 0.7-1.2 mm lateral to the midsagittal suture, and 8.0-8.2 mm below the dura. A stainless-steel wire, soldered to an Amphenol pin and wrapped around 3 or 4 stainless-steel skull screws, served as the current-return. Dental acrylic was used to secure the whole assembly to the skull screws. Once surgery was completed, animals received an injection of sterile saline solution (1.0 ml i.p.), in order to replenish lost fluids.

Apparatus and procedure

About a week after surgery, the animals were screened for self-stimulation on a continuous reinforcement schedule. All testing was conducted in a 27 cm long X 36 cm wide X 51 cm tall wooden and Plexiglas cage, which was fitted with a lever on the right wall, situated 3 cm above the floor of the cage. Each

lever press delivered a 500 ms train of 0.1 ms rectangular cathodal pulses; stimulation was provided by dual constant-current amplifiers (Mundl, 1980) and in-house manufactured pulse generators. In order to prevent polarization at the tip between pulses, the outputs of each channel were shorted to ground via a low resistance path. During stimulation trials, the parameters (see Figure 1A), were monitored on an oscilloscope, by observing the voltage drop across a 1 KΩ resistor in series with the rat. The beginning of each trial and the available stimulation current were signalled by five trains of "priming" pulses, separated from each other by a 1 sec interval. If after several screening sessions bar pressing was not evident, the rat was briefly anaesthetized with an inhalant fluothane (Halothane) and the moveable electrode lowered 0.32 mm using a microdrive (Kinetrods Reg'd); testing resumed 24 h later. This procedure continued until bar pressing was established, according to a minimum criterion of no less than five responses per minute. The currents used to screen for self-stimulation ranged from 200 to 1200 μA and the frequency values from 20 to 200 Hz, across animals and sites. Once bar pressing was reliably observed, a descending order of frequencies was administered, separated from each other by 0.05 $\log_{10}$ steps, starting with a value that yielded maximum responding to one that produced no responses. This procedure was applied at three different currents. The frequency thresholds were then interpolated from each rate-frequency function by finding the frequency corresponding to 50%

of the maximum response rate. The frequency thresholds were considered stable when their values did not vary by more than 0.1 $\log_{10}$ steps, across at least three consecutive sessions. The thresholds were then plotted against current to yield a frequency-current trade-off function. When stimulation-induced seizures occurred, the session was aborted and testing resumed after a 10-30 min interval. Occasionally, following a severe motor seizure, animals ceased responding altogether and testing had to be postponed until the next day.

Histology

When all behavioural tests were completed, the subjects were injected with an overdose of sodium pentobarbital (Somnitol, 60 mg/kg, i.p.) and then perfused intracardially with 0.9% saline, followed by 10% formalin. The brains were immediately removed and placed in the formalin solution for at least two days, after which the tissue was sectioned into 40 µm sections and treated with a Nissl stain to indicate cell bodies. The Paxinos and Watson (1986) atlas was used to determine the coordinates of the electrode tips. If the subject was implanted with a moveable electrode, the individual anatomical sites were estimated from the difference between the consecutive microdrive readings and the last position of the electrode tip. In other words, in order to estimate the depth of an electrode at the time of initial implantation site (site 0), the difference between the last position of that electrode and the number of equidistant 0.32 mm

consecutive moves was calculated.

EXPERIMENT 1

The psychophysical approach has been extensively used to document the characteristics of the reward sites located along the MFB. For example, strength-duration and spatio-temporal integration characteristics, refractory periods and conduction velocity estimates, to name a few, have been identified for MFB reward neurons (for recent reviews see Milner, 1991; Shizgal, 1989; Yeomans, 1990a). However, brain regions lying outside the MFB, such as the MPFC and CPu, have not been systematically studied using this approach. The few experiments that have been conducted suggest that the more anterior brain regions are characteristically different from that of the MFB reward substrate. For example, Schenk and Shizgal (1982) reported that the currents necessary to elicit MPFC self-stimulation are higher than the ones usually employed for MFB stimulation, indicating a less excitable and sparser population of reward fibers than the one underlying MFB self-stimulation. Thus, in order to characterize the spatial characteristics of neurons that support MPFC and CPu self-stimulation, the first aim of this experiment was to document the distribution of reward sites in the MPFC and CPu as reflected in their trade-off relationships between current and frequency.

Another characteristic of MPFC self-stimulation that

distinguishes it from that of the MFB is its gradual acquisition. For example, Corbett et al., (1985) showed that it typically took several sessions for the behaviour to develop, and that it was accelerated by applying non-contingent stimulation to the same site (Corbett at al., 1982b), or by initially shaping the animal to bar press on another self-stimulation site, such as the sulcal cortex, the lateral hypothalamus, or the ventral tegmental area (Cobo et al., 1989; Corbett et al., 1982c, 1985; Robertson et al., 1982a; 1986). Once self-stimulation on the MPFC was established as a consequence of the experience of rewarding stimulation at another site, activation of the training site, either the sulcal cortex, the lateral hypothalamus, or the ventral tegmental area, was no longer necessary to maintain MPFC self-stimulation (Robertson et al., 1986).

Three hypotheses were proposed to explain the mechanism underlying the slower acquisition. First, a kindling-like phenomenon, similar to synaptic potentiation, might facilitate acquisition, since treatment with anticonvulsant agents retards the development of brain stimulation reward in the MPFC (Balleine et al., 1989; Corbett, 1990; Robertson, 1989; Robertson et al., 1982b). Second, a reduction over time in an initial stimulation-induced behavioural inhibition (Corbett et al., 1985) might increase self-stimulation rates. In other words, gradual emergence of MPFC self-stimulation might be explained by an habituation to the disruptive effects of stimulation (Corbett et al., 1982b).

In light of these proposals, the second goal of this experiment was to assess whether MPFC acquisition would be promoted by self-stimulation already established at another site. To that end, the animals were implanted with pairs of electrodes, the first one in the MPFC, and the second in either the CPu, the lateral hypothalamus, or the ventral tegmental area. The latter three electrode placements were defined as the "training sites". After stable frequency thresholds were obtained from one of the training sites, stimulation was applied to the MPFC alone. Note that before data collection commenced at the training site, the MPFC was screened for self-stimulation and shown not to support the behaviour at the parameters tested. If following this manipulation, animals now bar-pressed for MPFC stimulation, the phenomenon in question was called the *transference effect*. Transference was defined as a persistent and consistent maintenance of self-stimulation, long after the effects of training electrode stimulation ceased, that is during the consecutive two to five testing sessions of the MPFC alone. It is important to point out that although many MPFC sites were subsequently tested in the same subject, only the first site was examined for transference, since the behaviour obtained from stimulating subsequent placements might have been confounded by the already established self-stimulation at the initial MPFC site.

According to the studies interested in the generation, pattern, and propagation of seizures in numerous limbic areas, a

transference effect provides evidence that kindling alters the excitability of neurons beyond the actual area of stimulation (Burnham, 1975; Burchfiel, Serpa, and Duffy, 1982; Duchowny and Burchfiel, 1981; Herberg and Rose, 1994). In other words, kindling of one area significantly enhances seizure susceptibility in another area. Interestingly, seizures typically accompany both MPFC and CPu self-stimulation, and it has been suggested that a kindling-like mechanism might be responsible for the gradual acquisition of MPFC self-stimulation (Balleine et al., 1989; Corbett, 1990; Robertson et al., 1982b).

Method

Subjects

Eleven of the 67 animals were used for the transference phase of this experiment.

Behavioural tests

The animals were implanted with a two-electrode assembly comprising one MPFC moveable electrode and either a lateral hypothalamic (3 rats), ventral tegmental area (3 rats), or CPu (5 rats) moveable electrode. Initially the animals were shaped to bar press for MPFC stimulation at currents ranging from 50 to 1000 µA across subjects over five consecutive sessions, adopting the following testing protocol used in other studies of this nature (Corbett and Stellar, 1983). After a minimum of shaping, the stimulation was made available at fixed values for 30 minutes

and the number of bar presses recorded. All of the animals showed only minimal and transient responding at the selected currents; rates of bar pressing ranged from 0 to 6 per 30 minute session. During this period no stimulation was delivered to the training electrode. Once the five-day protocol was completed, stimulation tests began at the training site. Trade-off functions between current and frequency were collected at currents that ranged from 400 to 800 μA for the lateral hypothalamus and 630 to 1000 μA for the ventral tegmental area. Four to six frequency thresholds were collected at three current values - low, medium, and high. For the most part, adjacent current values were in 0.1 $\log_{10}$ unit steps apart. If self-stimulation on the training electrodes was hampered by seizure activity or motoric side-effects, the electrode, if possible, was lowered to the next site. Immediately following the last stable frequency-threshold determination on either the CPu, lateral hypothalamus, or ventral tegmental area electrode, stimulation was applied to the MPFC site, at parameters which produced the lowest frequency threshold at the training site. In some instances, the currents had to be adjusted in order to produce optimal responding and also to eliminate any factor that interfered with bar pressing, such as motor effects and seizures. Once reliable self-stimulation was established, trade-off functions between current and frequency thresholds were collected at the MPFC site. Then the total charge was calculated for each trade-off function between frequency and current, using the

following formula (Gallistel, 1978):

$$Q = INd \quad \text{where}$$

Q = the charge in μC

I = the current in μA

N = the number of pulses in the 500 ms stimulation train

d = the pulse duration in sec (0.0001)

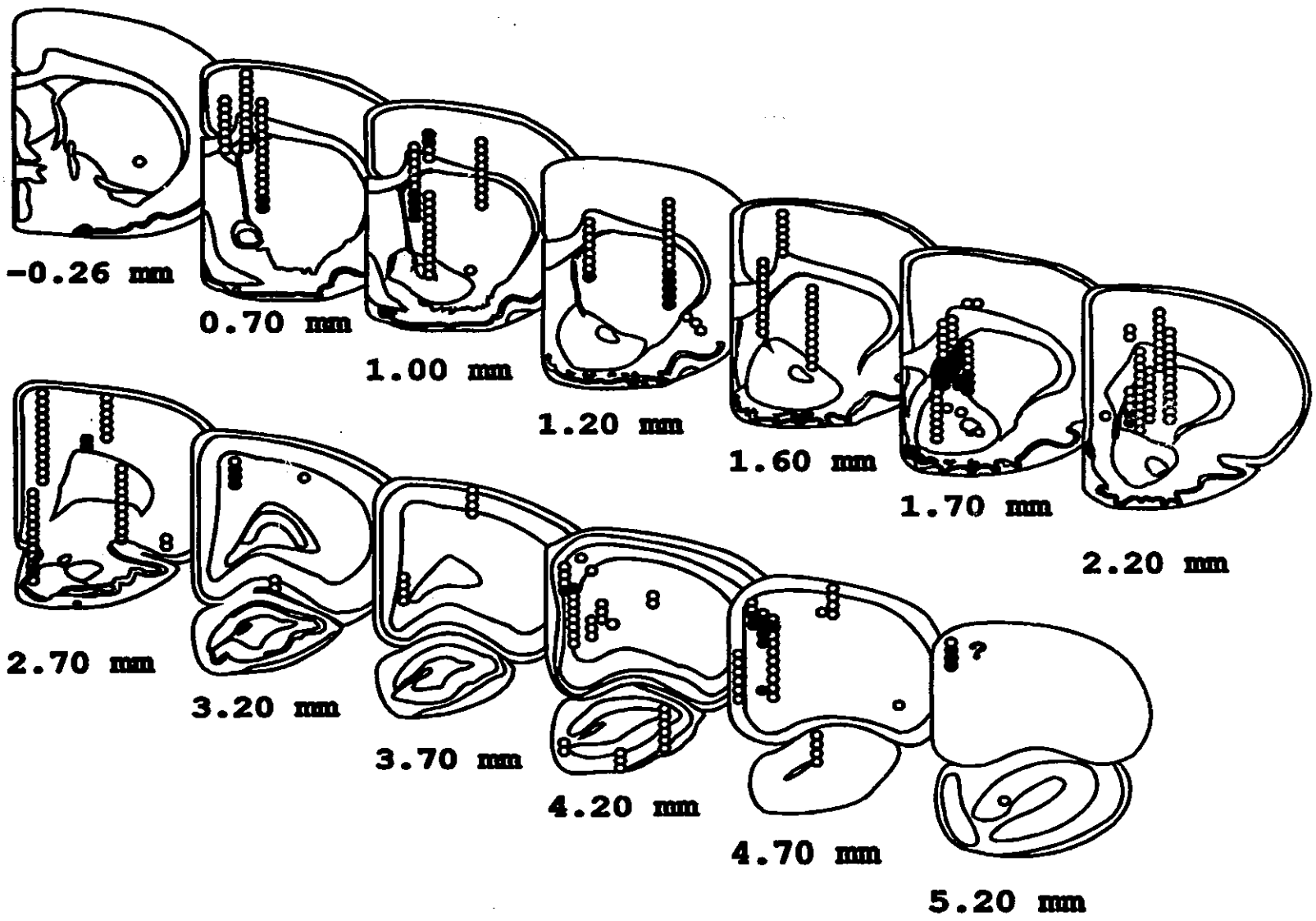
This formula is somewhat simplified as it does not take into account the minimum current needed to evoke self-stimulation; however, it is useful as a comparison to the charge values obtained at other sites which have been evaluated using the same formula. When the reciprocity between frequency threshold and current is equal, the charge values across currents should remain constant, provided that the minimum current is close to zero and the trade-off function between current and frequency is expressed in log values. The advantage of using charge lies in the fact that it combines the different currents and frequencies from the trade-off function into a single representative value for the whole curve. Such manipulation is especially useful when only a few data points are obtained, as was often the case here, which prevents the traditional representation of the changes in frequency thresholds as a function of current.

Results

Histology

Sixty seven animals were screened for self-stimulation on both the MPFC and the CPu. Ten rats, altogether tested at 20 MPFC and 29 CPu sites, demonstrated self-stimulation, ranging from transient behaviour interrupted by epileptogenic activity to consistent bar pressing. Figure 6 shows the 187 and 255 individual sites evaluated in the MPFC and CPu respectively. There was about an 11% success rate of obtaining self-stimulation in both regions. The cluster of positive sites for the MPFC was found in the Cg1, Cg3, Fr1, Fr2, and MO/VLO portions (for the anatomical description see page 44 in the Introduction), and for the CPu, predominantly in the ventromedial region; some positive areas were also found in the ventrolateral CPu. The highest concentration of positive self-stimulation sites in the MPFC was found in the Cg3 region at 4.7 mm in front of bregma, and for the CPu, in the vicinity of the lateral septum at 1.7 mm anterior to bregma, according to the atlas of Paxinos and Watson (1986). Thus, the greatest concentration of positive self-stimulation sites is clustered in the anterior MPFC and CPu regions. Among striatal placements, the nucleus accumbens (core), an inner region of this structure, was found not to support self-stimulation, which is in agreement with previous work that suggests that this area is non-limbic, as opposed to the nucleus accumbens (shell), an outer portion, which is thought to be

Figure 6. This figure illustrates the locations of all the CPu and MPFC sites evaluated for self-stimulation, arranged from the most posterior (upper left) to most anterior (lower right) placements. The circles correspond to the histological verification of the electrode tips; unfilled circles indicate placements from which self-stimulation was not obtained; the filled ones mark positive self-stimulation sites. The clipart sections are from the program NeuroGraphics - The Rat Brain, which is based on the Paxinos and Watson (1986) atlas, with additional plates. The question mark on the coronal section labelled +5.2 reflects uncertainty as to the exact position of that electrode tip.


-0.26 mm
0.70 mm
1.00 mm
1.20 mm
1.60 mm
1.70 mm
2.20 mm
2.70 mm
3.20 mm
3.70 mm
4.20 mm
4.70 mm
5.20 mm

limbic in function (Deutch, Lee and Iadarola, 1992). It appears that positive self-stimulation regions in both the MPFC and CPu overlap with catecholaminergic innervations in these areas (Kolb, 1990; McGregor et al., 1989). For a more detailed discussion on other possible neurotransmitter systems involved in forebrain self-stimulation, refer to page 55 in the Introduction.
Figure 7 shows the histological placements of the training electrodes. The frequency thresholds varied from 26 to 50 Hz for the lateral hypothalamus, 15 to 66 Hz for the ventral tegmental area, and 16 to 84 Hz across the MPFC and CPu. In all but one case (subject CH58), the acquisition of MPFC self-stimulation was not facilitated by prior rewarding MFB stimulation (subjects CV37, CH39, CV44, CV45, CH57, and CH59). However, a transference effect was observed following rewarding stimulation of the perifornical nucleus (subject CH58). Further, rewarding stimulation applied to three out of four training sites in the CPu appeared to facilitate the acquisition of ipsilateral MPFC self-stimulation (subjects CP46, CP64 and CP66, but not CP62), as confirmed by the appearance of reasonable rates of bar pressing. Lastly, no transference of the behaviour to the MPFC was observed following stimulation of a contralateral CPu site (subject CP65).

Charge values

In this study, charge values ranged from 0.81 to 1.63 μC for the MPFC and 0.68 to 1.60 μC for the CPu. The pattern in Figure 8 demonstrates that both MPFC and the CPu show a similar

Figure 7. This figure shows the location of the electrode placements in the transference experiment, assembled from the most posterior (top left) to the most anterior coronal plate (bottom right). Panel A illustrates the sections containing the electrode tips aimed at the lateral hypothalamic and ventral tegmental areas, panel B, the striatal placements, and panel C, the cortical placements. The alphanumeric located beside each site refers to the identity of the subject. The filled circles indicate the sites that gave rise to self-stimulation and the unfilled circles indicate sites where self-stimulation was not obtained.

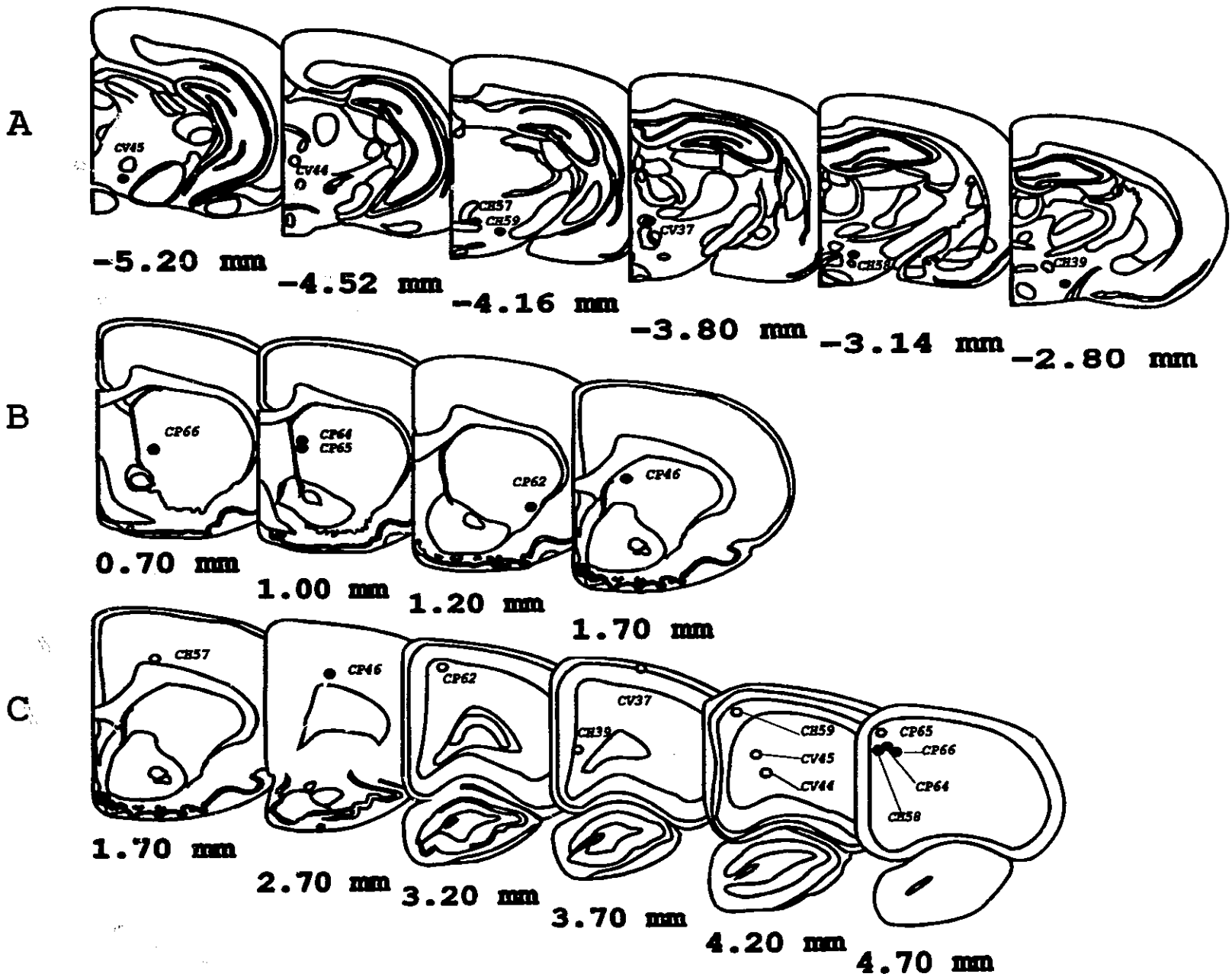
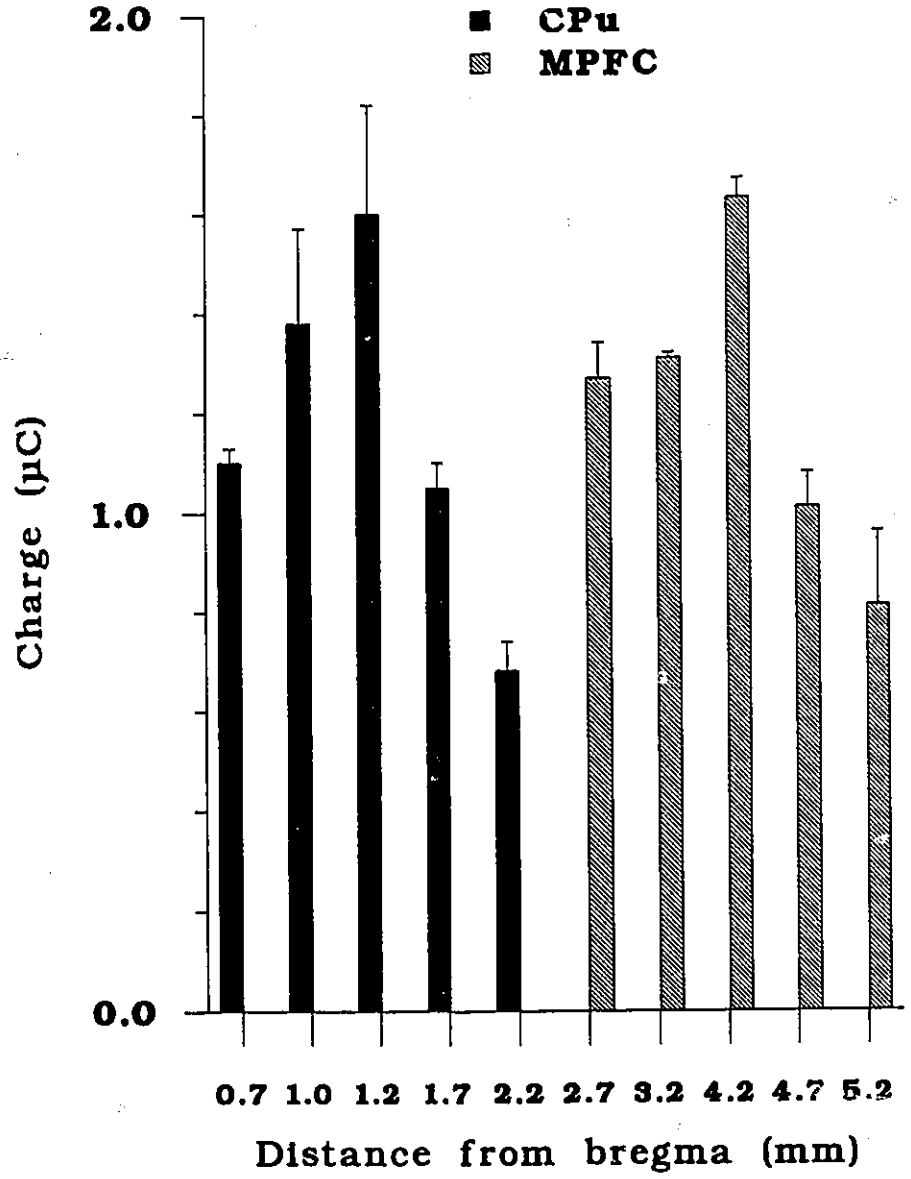

A
CV45
CV44
CB57
CB59
CV37
CB58
CB39
-5.20 mm
-4.52 mm
-4.16 mm
-3.80 mm
-3.14 mm
-2.80 mm
B
CP66
CP64
CP65
CP62
CP46
0.70 mm
1.00 mm
1.20 mm
1.70 mm
C
CB57
CP46
CP62
CV37
CB39
CB59
CV45
CV44
CP65
CP66
CP64
CB58
1.70 mm
2.70 mm
3.20 mm
3.70 mm
4.20 mm
4.70 mm

Figure 8. This graph shows the average charge ± S.E.M. for all CPu and MPFC self-stimulation sites as a function of distance from bregma (0.7-5.2 mm), or anteroposterior coordinate. The charge values associated with each current were averaged to represent the total charge for a particular site. In order to illustrate an overall pattern, the average of all charge values was calculated to yield a single representative value for each coronal section.


2.0
1.0
0.0
CPu
MPFC
Charge (µC)
0.7 1.0 1.2 1.7 2.2 2.7 3.2 4.2 4.7 5.2
Distance from bregma (mm)

distribution of total charge values across the posterior-anterior plane; charge increased progressively from the most posterior MPFC and CPu sites until it reached a peak at roughly 1.? mm in front of bregma for the CPu (the last plate before the appearance of tenia tecta and the first section where nucleus accumbens core is shown in two separate parts), and at 4.2 mm for the MPFC (the first coronal plate where regions Fr1, Fr3, dorsal transition zone, forceps minor, and the corpus callosum are no longer visible); it then decreased gradually to reach the lowest value in the most anterior MPFC and CPu regions. The lowest charge values were found at 2.2 mm (the first coronal section where regions Cg3, IL, VLO, LO and dorsal peduncle appears and where tenia tecta and lateral septal nucleus are no longer visible), and 5.2 mm in front of bregma (the plate where the internal granular layer, internal and external plexiform layers of the olfactory bulb can be seen, and where the regions Cg1 and Cg3 are no longer visible) for the CPu and MPFC respectively (Paxinos and Watson, 1986). A decrease in required charge is interpreted as an increase in the rewarding value of the stimulation and vice versa (Gallistel, 1978; Harris and Bielajew 1991). The charge values are derived from the frequency-current function. The slope of the frequency-current function is believed to reflect the density of the relevant fibers at the site of stimulation, the higher the density the greater the number of relevant fibers recruited, which ultimately is translated into a smaller required current for self-stimulation (Sax and Gallistel, 1991). As an

aside, when the frequency thresholds at a given current are plotted as a function of distance from bregma the same pattern as described above is obtained when charge is graphed as a function of distance from bregma.

It appears that the sites supporting the most reliable self-stimulation are clustered at 4.7 and 1.7 mm in front of bregma respectively, in the ventromedial MPFC and CPu. Interestingly, these sites are also the ones represented by charge values of nearly 1 µC. It was previously reported that charge values obtained for sites along the MFB, at roughly the same parameters as used here, are also around 1 µC (Bielajew et al., 1987a; Gallistel, 1978). In the present study, an average charge of 1.35 µC was obtained for the ventral tegmental area and 1.04 µC for the lateral hypothalamic placements.

Behavioural observations

All animals gradually developed severe motor seizures; however, there were differences in the seizure profile observed at the two sites. The MPFC seizures were generally less frequent, developed more slowly, but tended to be more disruptive to self-stimulation, as reflected in elevated thresholds, or at times complete cessation of self-stimulation. The CPu seizures, on the other hand, appeared abruptly, were more frequent and severe, but less disruptive to thresholds overall. The animals usually recovered quickly and returned to the bar. Reducing the train duration to either 300 or 250 ms, thus shortening the time

of stimulation delivery, has been used in some studies as a strategy to reduce the possibility of seizure occurrence (Balleine et al., 1989; Rick and Fouriezos, 1992). Similarly, applying an anticonvulsant agent, brotizolam (5 mg/ml or 7.5 mg/ml), has been shown to reduce the severity of seizures and the threshold variability at self-stimulation sites in the MFB associated with seizures (Harris and Bielajew, 1991). However, neither of these measures was effective in this study. Interestingly, it has been reported that drugs known to suppress seizure activity also reduce self-stimulation rates (Reid et al., 1964; Weinreich and Clark, 1970). Further, some subjects appear to "titrate" the level of stimulation by decreasing their rate of bar pressing, so as to prevent the seizures from re-occurring. Occasionally, in this study, a decrease in the incidence of seizures throughout the session was also accompanied by progressive elevation in self-stimulation thresholds. It has been pointed out that MPFC stimulation may lower seizure thresholds, which would in turn cause the animal to perform more slowly in order to avoid a seizure (Lenzer, 1971).

The other secondary effects of stimulation included occasional biting of the lever, coprophagia, grooming, yawning, and motoric effects unrelated to seizures, such as circling.

Discussion

It appears that the high required currents, generally low

success rate in establishing reliable self-stimulation, and uneven distribution of charge values obtained at these forebrain self-stimulation sites may indicate a recruitment of smaller, less excitable, or sparsely distributed reward-relevant fibers, which corroborates other studies of this nature (Schenk et al., 1985). It appears that the average minimum current for MPFC self-stimulation is about 300 μA (Schenk et al., 1985), which is significantly higher than the value for most MFB sites - around 100 μA (Gallistel, 1978). It is thus possible that these dissimilarities in current between the MPFC and MFB reflect differences in the density of reward-relevant neurons and possibly in the post-synaptic integration of reward signals in the two substrates (Schenk et al., 1985). A good way to assess this hypothesis would be to conduct an isopreference experiment in which the animal is provided a choice between MPFC and MFB stimulation, a strategy that has already been used to assess anterior and posterior MFB placements (Anderson and Miliaressis, 1994). In their study, in the majority of placements gave rise to an equipreference for frequencies that supported the same proportion of maximum rate; only a few cases showed a preference specifically for one or the other location (Anderson and Miliaressis, 1994).

The fact that transference of lever pressing to the MPFC was not observed following rewarding stimulation of the lateral hypothalamic or ventral tegmental area placements, but did occur following rewarding CPu stimulation under the experimental

protocol used in this study, lends further support to the idea that the MFB and the anterior forebrain regions may constitute separate reward circuits. These conclusions are strengthened by previous research which has shown that behaviourally derived refractory periods associated with cortical sites are longer than the ones usually obtained for the MFB, both beginning and ending later (Trzcińska and Bielajew, 1992), indicating that different populations of fibers underlie self-stimulation at the two sites. Near the end of this study, a pilot test was conducted in which double pulses were delivered concurrently to an MFB electrode that supported self-stimulation and an ipsilateral MPFC site that did not (subject CH39); this strategy did not hasten the acquisition of MPFC self-stimulation. It thus appears that co-stimulation of the MPFC in conjunction with any self-stimulation site is not sufficient to evoke this behaviour during a single shaping session. One possibility is that in order for the transference effect to occur, the two sites must be axonally connected, a hypothesis explored in the third experiment of this thesis, and supported by the observation that rewarding stimulation of a contralateral CPu placement did not give rise to transference of acquisition to the MPFC (subject CP65). It thus appears that in order to observe the acquisition of MPFC self-stimulation, it is not sufficient to simply incorporate bar pressing into the animal's behavioural repertoire. The site at which the animal first encounters rewarding stimulation and its possible anatomical relation to the MPFC could be the determining

factor.

In relation to the MFB placements, it has been shown that non-contingent lateral hypothalamic stimulation does not facilitate MPFC self-stimulation (Robertson et al., 1982a). In another study, Corbett et al. (1985) found that bar pressing for non-contingent lateral hypothalamic stimulation hastened MPFC acquisition. However, they did not assess the MPFC sites prior to the delivery of hypothalamic stimulation, as was done here. It is conceivable that in their study the observed transference was a function of the specific MPFC placements, stimulation of which might have supported self-stimulation anyway, and not related to experience on the lateral hypothalamic site per se. In the present experiment, non-contingent stimulation was not employed to facilitate the acquisition of MPFC self-stimulation, as was done in other studies (Balleine et al., 1989; Corbett, 1990), because rats either find this type of stimulation slightly aversive (Ettenberg et al., 1981; Steiner et al., 1969), or simply prefer to control the rate at which rewarding stimulation is administered (Tsang and Stutz, 1984).

As far as the ventral tegmental area is concerned, studies have found that electrical stimulation of this region and a microiontophoretic application of dopamine into the prefrontal cortex result in an inhibition of spontaneously firing cortical cells (Ferron et al., 1984; Sesack and Bunney, 1989). This inhibitory effect of ventral tegmental area upon the cells in the MPFC may be an explanation for why transference from this region

to the MPFC does not occur.

The results of the present study might also suggest that a kindling-like mechanism through activation of a related structure, such as the CPu, might underlie MPFC self-stimulation (Corbett et al., 1982b; Robertson et al., 1982b). This hypothesis is particularly interesting in light of the fact that self-stimulation of the MPFC and CPu is almost always accompanied by seizures, which become progressively more severe and as a consequence more disruptive to the testing regimen (Herberg and Rose, 1994). There is some evidence that the epileptogenic activity might be crucial to the learning process involved in MPFC self-stimulation, because seizures are often more prominent following and not preceding acquisition of self-stimulation (McGregor, 1992); prior to acquisition, arrest-like behaviour is more prominent than the epileptogenic activity. In addition, experimental evidence suggests that the MPFC has subcortical and limbic cortical connections that may be instrumental in the propagation of seizure activity, which may contribute to the termination of self-stimulation in certain reward areas, as shown by generally low response rates for MPFC self-stimulation (Herberg and Blundell, 1969). Further, it seems that the CPu receives convergent seizure activity from the MPFC, orbital and insular cortices, amygdala, and the hippocampus, the latter two constituting highly epileptogenic structures in the brain (Beckstead, 1979; Parent and Hazrati, 1995a). In addition, midline CPu and MPFC stimulation appears to promote epileptogenic

activity (Handforth and Ackermann, 1993).

Other evidence points to the fact that kindling of one limbic site alone can greatly increase seizure susceptibility of other secondary sites (Burchfiel et al., 1982; Duchowny and Burchfiel, 1981), which suggest that kindling can modify neuronal excitability beyond the actual region of stimulation. Also, if the transfer site (MPFC) is monosynaptically linked to the primary site (CPu) then it might be rapidly or immediately kindled by the input from the primary site and subsequently demonstrate a similar pattern of discharge, or in this case a similar pattern of response acquisition (Spiller and Racine, 1994). In addition, it is not simply striatal stimulation per se which facilitates the acquisition of MPFC self-stimulation; the CPu stimulation must be rewarding. This conclusion is supported by the observation that stimulation of CPu sites which showed high epileptogenic activity but no evidence of self-stimulation failed to facilitate MPFC self-stimulation; this occurred in two animals.

Alternatively, the reason why transference was not observed from stimulation of all but one MFB placement, may be due to the fact that most target cortical areas, except for subjects CH39 and CP65, were located outside the Cg3 region. Anatomical studies suggest that the Cg3 area appears to be limbic in function, while other cortical regions support primarily motor and somatic functions. This may explain why a transference effect was observed following stimulation of the perifornical

hypothalamus in subject CH58; its target cortical site was located in the Cg3 region. McGregor and his associates (1989) found that in some animals the acquisition of MPFC self-stimulation was rapid and seemingly no different from that of the MFB. In others the acquisition may have depended on the dissipation of an initially aversive or motorically suppressive effect induced by MPFC stimulation. However, most studies indicate that stimulation of the MPFC does not seem to produce aversive effects, even at high currents. It may appear that the emergence of rewarding MPFC stimulation depends on the precise locus of stimulation (Robertson, 1989).

In conclusion, the evidence presented here seems to hint at the notion that the MPFC and CPu form part of the same reward substrate and one distinct from that of the MFB. One way in which this hypothesis can be further evaluated is to measure the excitability cycle of the reward neurons contained in these regions using the behavioural adaptation of the refractory period test.

EXPERIMENT 2

There is a growing body of evidence suggesting that the anterior brain regions constitute a different reward substrate from that of the MFB. Much of the support for this notion comes from studies aimed at characterizing the electrophysiological properties of the underlying substrates. For example, it has been found that the behaviourally derived refractory periods underlying self-stimulation in the forebrain tend to be longer than the ones obtained for MFB self-stimulation (Bielajew and Fouriezos, 1985; Fouriezos et al., 1987; Schenk and Shizgal, 1982). In light of the above evidence, the aim of this experiment was to investigate the excitability cycle of the substrate for brain stimulation reward in the MPFC and CPu, using the behavioural adaptation of the refractory period test.

Some of the data presented here have already been published (see Trzcińska and Bielajew, 1992). For a more detailed description of the refractory period method, please refer to pages 21 to 31 in the Introduction.

Method

Subjects

Sixteen of the 67 animals were used for the refractory period experiment.

Behavioural tests

The currents which were associated with the lowest and most stable frequency thresholds, without producing seizures during the training sessions, were selected for the refractory period test. The tests begun and ended with a single pulse threshold determination. These served two functions; first their repetition was used to verify the stability of the measure throughout the session, and second, they provided the average value against which the threshold determinations in the double pulse condition could be evaluated. In the double pulse tests, the C-T interval was varied from 0.2 to 20.0 ms in either a descending or ascending order. Once the testing protocol was completed, usually after four to six test sessions, the electrode was lowered 0.32 mm and the entire stabilization procedure repeated at the new site, as described on page 64, before refractory period tests were initiated. The effectiveness of the T-pulses was assessed by comparing the average threshold for single pulses to each threshold for double-pulse stimulation, using the following formula (Yeomans, 1975):

$$E = (FT_{SP}/FT_{C-T}) - 1 \quad \text{where}$$

E = effectiveness of the T pulses

FT_{SP} = average frequency threshold of single pulses (T pulses absent) required to support criterion performance

FT_{C-T} = frequency threshold of pulse pairs (C and T pulses) at a given C-T interval required to support criterion performance

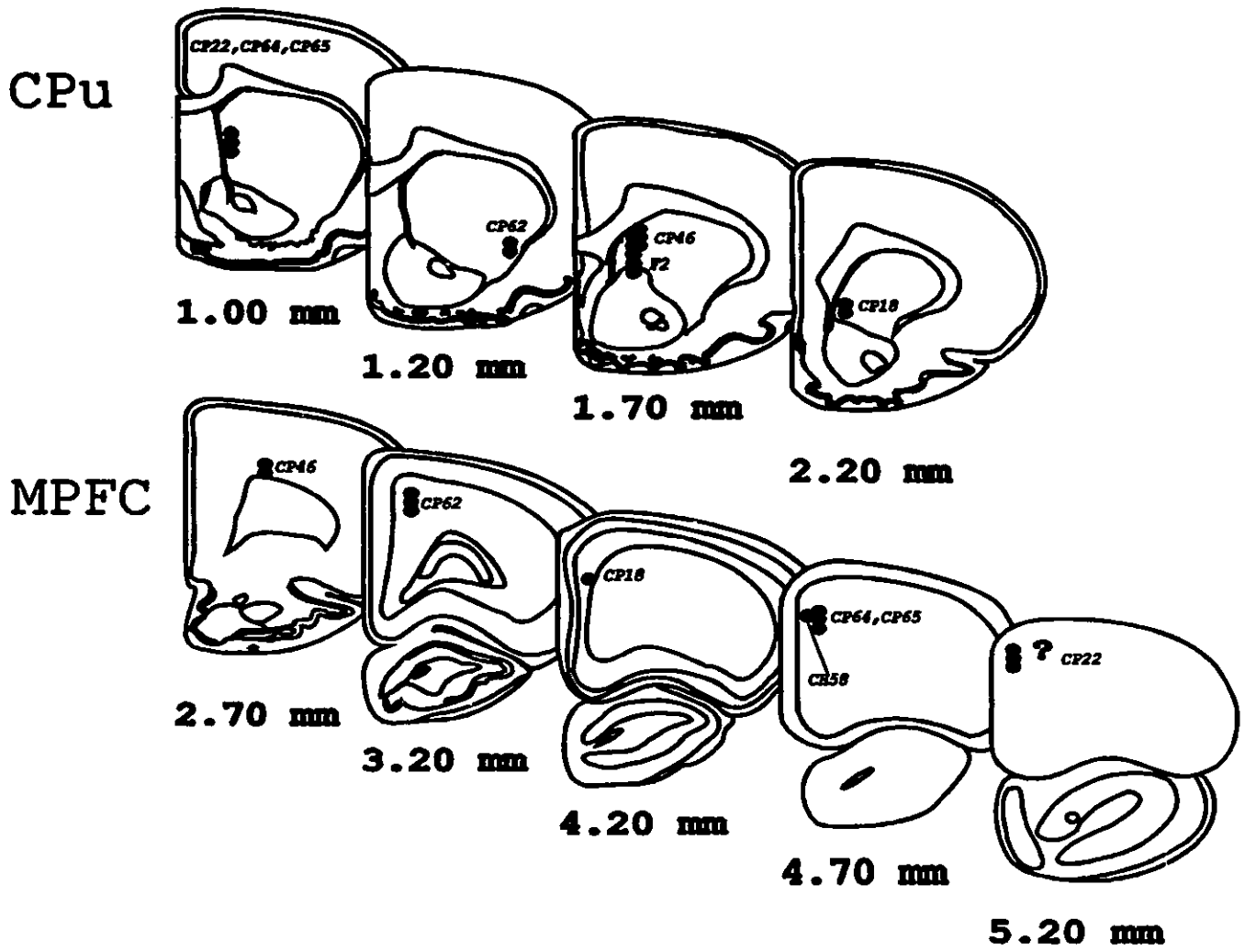
The scaling of effectiveness values as a function of C-T interval produces a characteristic profile. When the T pulses fall within the absolute refractory period, they are completely ineffective; the double pulse threshold is thus equivalent to the single pulse threshold, and the resulting E value is zero. If however, the T pulses fall outside the refractory period range, they are maximally effective; in this case, the double pulse threshold is half of the single pulse threshold and the resulting E value is equal to one. For a more detailed description of the rationale behind the scaling, please refer to pages 23-26.

Results

Histology

Figure 9 summarizes the histologically verified locations of the 11 CPu and 16 MPFC sites in seven animals for which excitability cycles were determined in this experiment. Except for subject CP62, for which the electrode tip was located in the ventrolateral CPu, all striatal sites were found to be in the anterior ventromedial CPu. Most MPFC placements were located in the anterior Cg1 and Cg3 regions, and in one case, in the Fr1/Fr2

Figure 9. This figure illustrates the location of the CPu and MPFC sites, from the most posterior (upper left) to the most anterior (lower right), at which refractory period tests were conducted. The clipart sections are from the program NeuroGraphics. The question mark associated with CP22 (+5.2 mm) reflects uncertainty as to the exact location of that electrode tip. The alphanumeric positioned either directly on the section, or beside a group of sites marked by filled circles, refers to the identity of the subject. In three cases (subjects CP22, CP64, CP65), because of the histological overlap between sites the identity of the subject to its specific site could not be matched.


CPu
CP22,CP64,CP65
CP62
CP46
F2
CP18
1.00 mm
1.20 mm
1.70 mm
2.20 mm
MPFC
CP46
CP62
CP18
CP64,CP65
CP58
? CP22
2.70 mm
3.20 mm
4.20 mm
4.70 mm
5.20 mm

area (subject CP46).

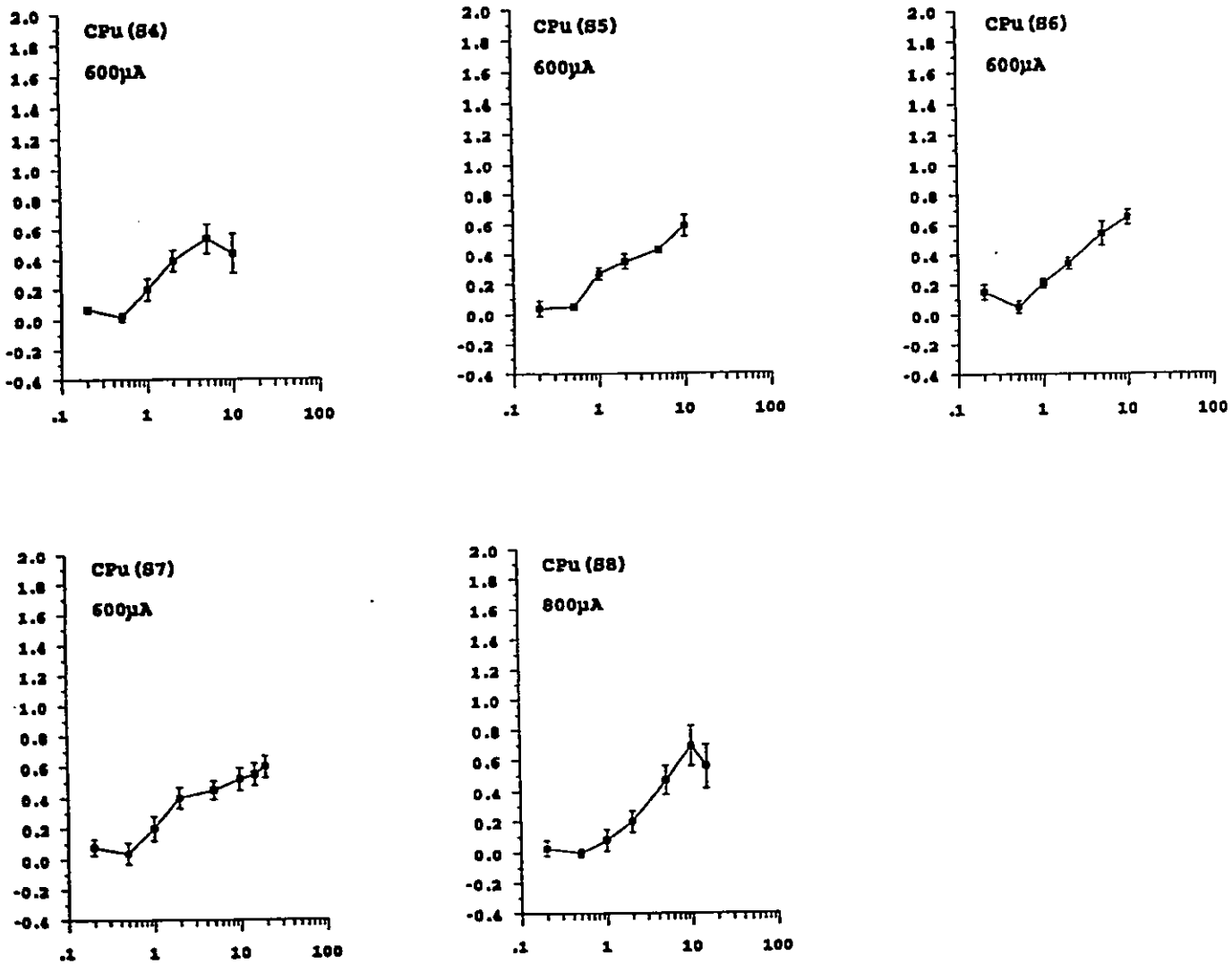
Refractory period estimates

Figure 10 a-h shows the average T pulse effectiveness as a function of C-T interval for sites tested in individual animals. Figure 11 illustrates the average refractory period curve for all the CPu and MPFC placements. The overall shape of the curves (see Figure 3 in the Introduction) resembles that reported at other self-stimulation sites (Bielajew et al., 1987a, 1987b; Bielajew and Shizgal, 1982, 1986; Fouriezos et al., 1987; Rompré and Miliaressis, 1980, 1985; Schenk and Shizgal, 1982; Silva et al., 1982; Szabo et al., 1974; Yeomans, 1975, 1979).

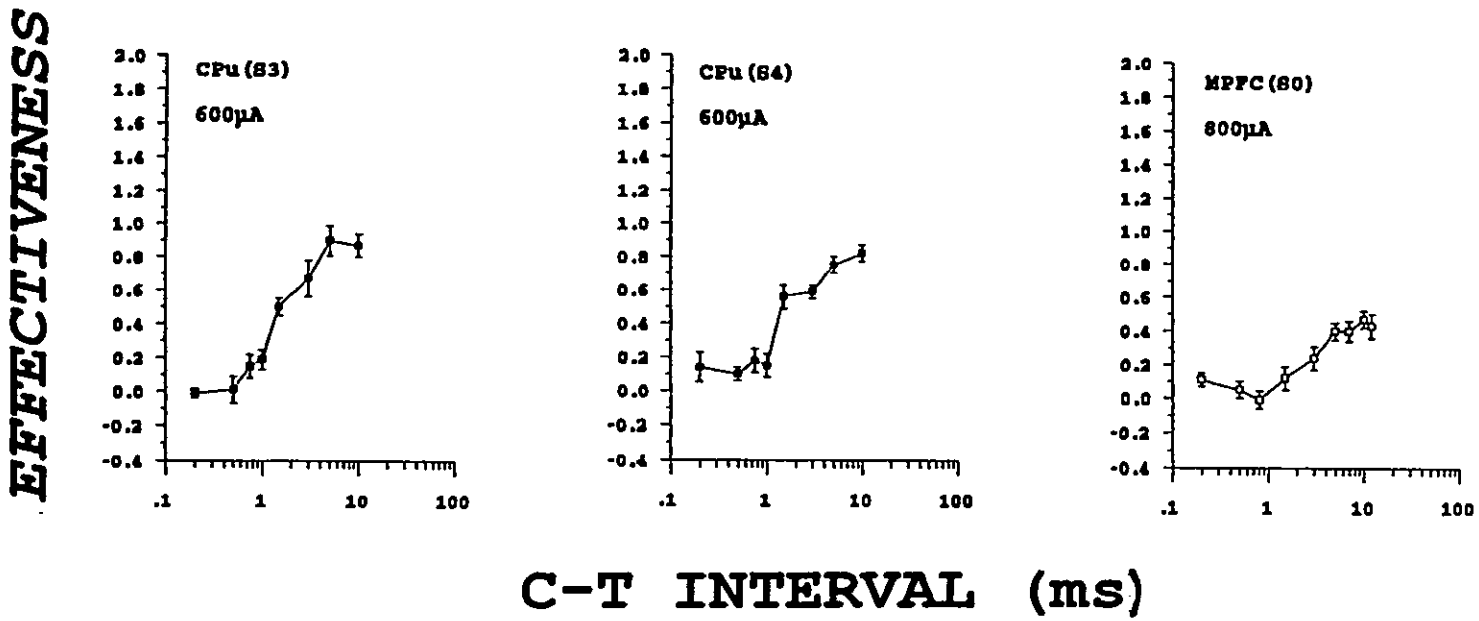
The range of refractoriness was estimated by evaluating the C-T interval at which the curves began to rise (beginning of recovery) and the C-T interval at which the curves approached an asymptote (end of recovery). Since it is difficult to remove the effects of local potential summation which obscure the beginning of recovery, (please refer to pages 26 to 28), a curve fitting method was used to estimate the onset of recovery from refractoriness. It was calculated by performing a weighted regression analysis on all pairs of effectiveness and associated C-T intervals below and including the C-T interval at which the end of recovery was estimated; the latter was evaluated using the asymptote test, described below. Initially, the two longest C-T intervals were subjected to a weighted linear regression. An effectiveness value was predicted for the next (third) C-T

Figure 10 a-h. The graphs show the average effectiveness of T-pulse stimulation ± S.E.M. as a function of C-T interval for all the CPu (filled squares) and MPFC (open squares) sites evaluated in each subject. The alphanumeric identifying the animal is located in the top left corner of each figure. The specific site from which the curve was collected, as well as the current value at which each function was obtained are identified at the top of each individual graph.

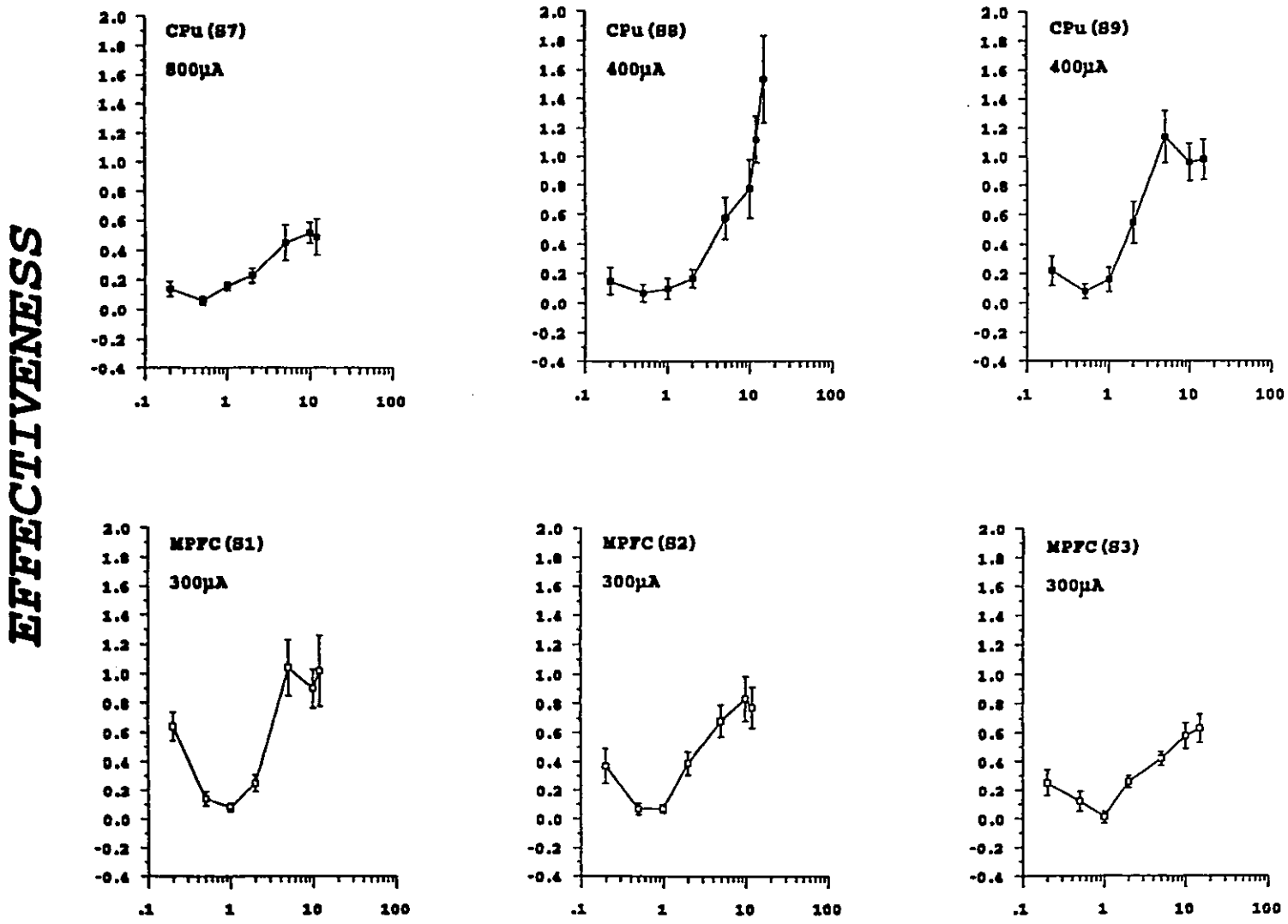
a) F2


CPu (S4)
600µA
CPu (S5)
600µA
CPu (S6)
600µA
CPu (S7)
600µA
CPu (S8)
800µA
EFFECTIVENESS
C-T INTERVAL (ms)

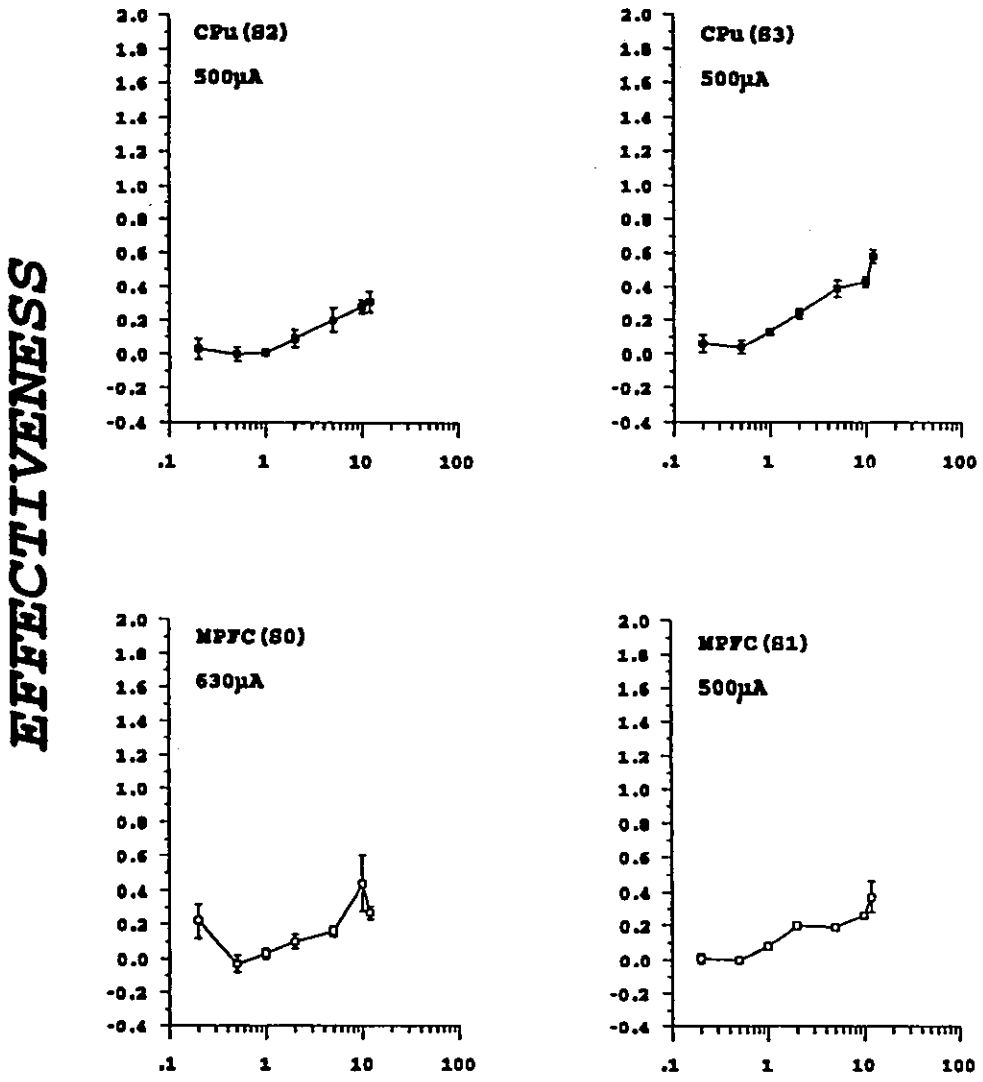
b) CP18


EFFECTIVENESS
CPu (S3)
600µA
CPu (S4)
600µA
MPFC (S0)
800µA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

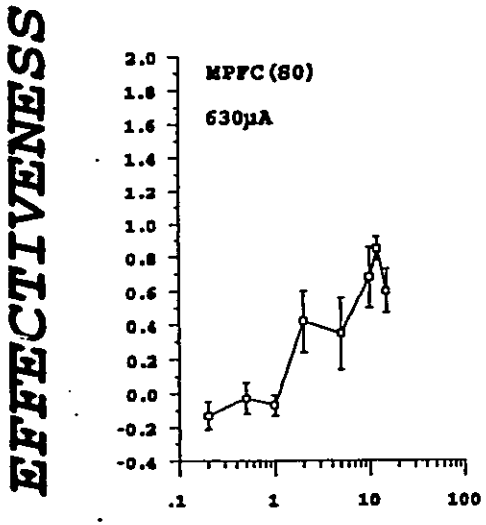
c) CP22


CPu (S7)
800µA
CPu (S8)
400µA
CPu (S9)
400µA
MPFC (S1)
300µA
MPFC (S2)
300µA
MPFC (S3)
300µA
EFFECTIVENESS
C-T INTERVAL (ms)

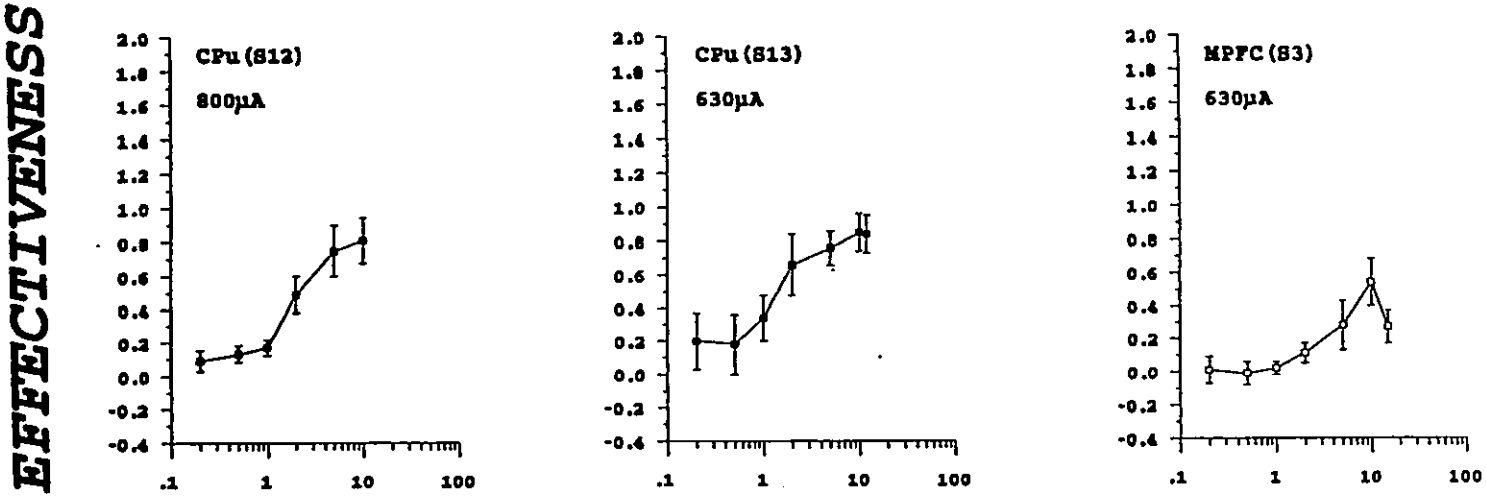
d) CP46


CPu (S2)
500µA
CPu (S3)
500µA
MPFC (S0)
630µA
MPFC (S1)
500µA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
EFFECTIVENESS
C-T INTERVAL (ms)

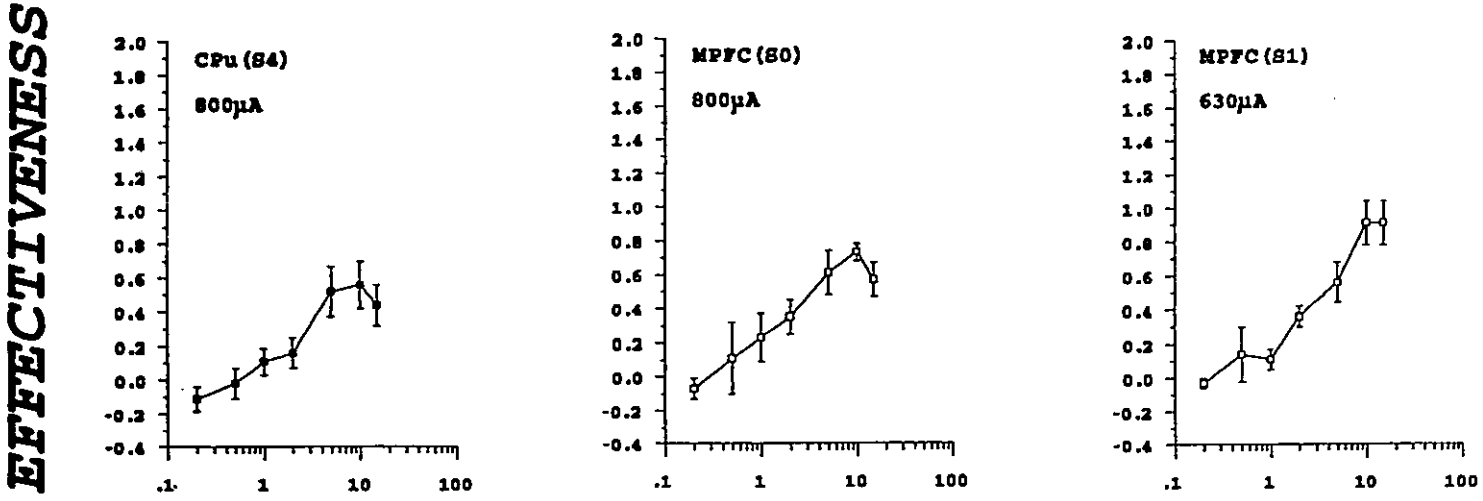
e) CH58


EFFECTIVENESS
MPFC(80)
630μA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

f) CP62


EFFECTIVENESS
CPu (S12)
800µA
CPu (S13)
630µA
MPFC (S3)
630µA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

g) CP64


EFFECTIVENESS
CPu (S4)
800µA
MPFC (S0)
800µA
MPFC (S1)
630µA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

h) CP65

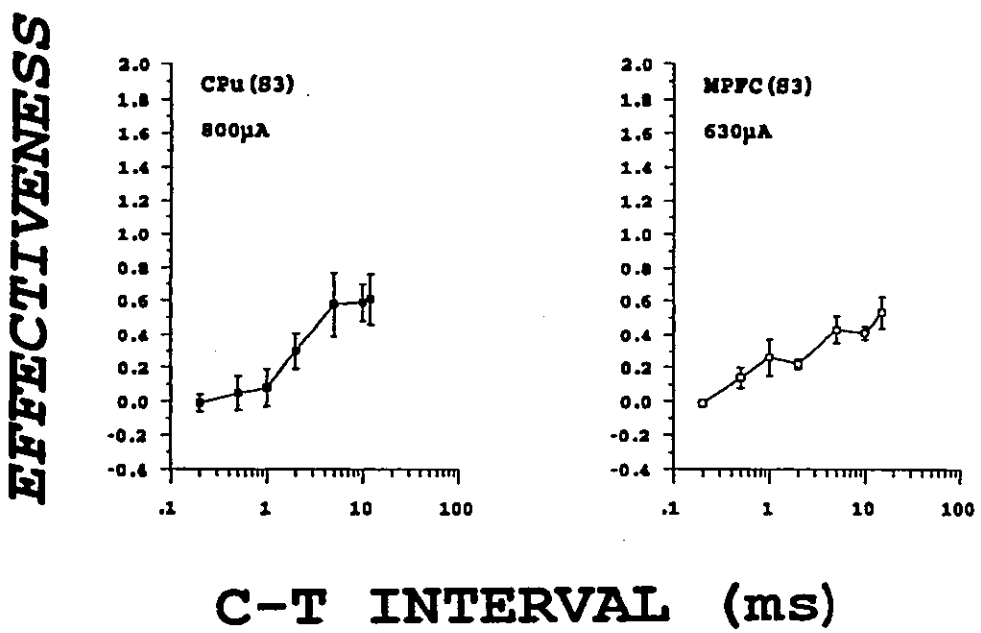
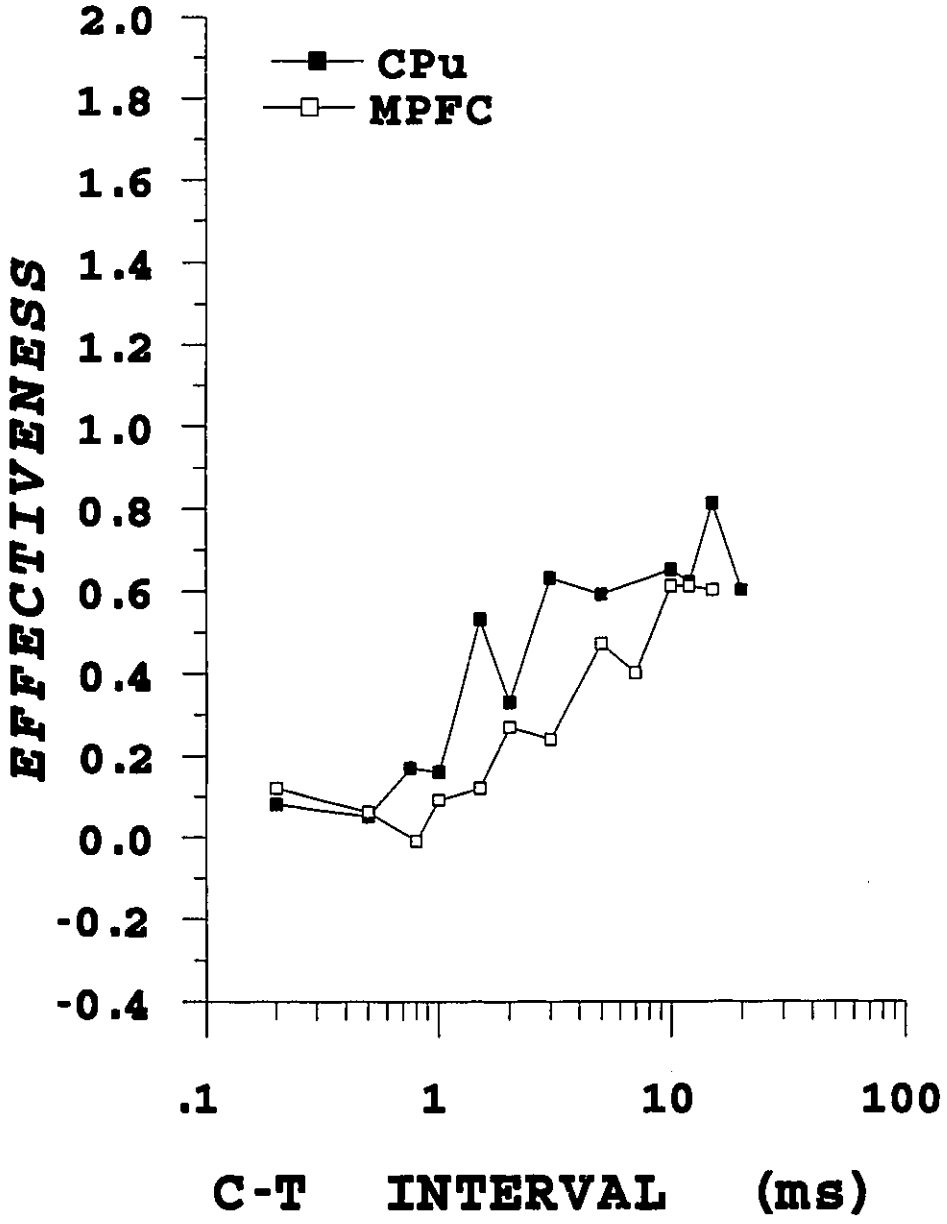

EFFECTIVENESS
CPu (S3)
800µA
MPFC (S3)
630µA
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

Figure 11. An illustration of the average CPu and MPFC functions relating the effectiveness of T-pulse stimulation to C-T interval for all the placements shown in Figure 10 a-h.


2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
CPu
MPFC
EFFECTIVENESS
.1
1
10
100
C-T INTERVAL (ms)

interval. If the actual effectiveness value associated with the third C-T interval was not significantly different from the predicted effectiveness value, then the procedure was repeated for the next (fourth) C-T interval. The analysis continued until the actual effectiveness value associated with a C-T interval was significantly different from the predicted value. At that instant it was assumed that the divergence of the effectiveness value from the regression line was due to local potential summation. The intercept, or the point at which the regression line met the abscissa (C-T interval) was defined as the beginning of recovery. This value was obtained from the regression equation fit to all the pairs of effectiveness and associated C-T intervals above and including the C-T interval marking the beginning of recovery.

The end of recovery was assessed by the asymptote test (Bielajew et al., 1981). The method consisted of comparing the S.E.M. of the effectiveness value about the longest C-T interval to the S.E.M. about the effectiveness value associated with the next shorter C-T interval. If the two S.E.M. overlapped, the values contributing to these two means were pooled and a new effectiveness value and S.E.M. calculated, and then compared to the next shorter C-T interval. The whole procedure was repeated until no overlap in the S.E.M. was found for the pooled and adjacent effectiveness values. The end of recovery was defined as the shortest C-T interval corresponding to the pooled effectiveness value.

On average, the beginning of recovery was observed at 1.4 ms for the MPFC and 0.8 ms for the CPu; the end of recovery was determined to be 7.9 ms for the MPFC and 5.4 ms for the CPu. In addition, local potential summation effects were observed at the shortest C-T intervals at both sites, but were more prominent in the MPFC.

Two different patterns seemed to emerge. Most curves rose gradually and approached a plateau at average effectiveness values of 81% for the CPu and 61% for the MPFC. In some cases, however, the effectiveness values rose gradually without showing a distinct plateau [F2/CPu(S5) and CP46/CPu(S3)] or continued to rise abruptly even at the longest C-T intervals tested [CP22/CPu(S8)]. As a rule, if the curve showed no clear plateau, then the S.E.M. about the effectiveness values associated with the longest C-T did not overlap the adjacent effectiveness value and S.E.M., so that the end of recovery from refractoriness could not be estimated.

Table 1 lists the C-T intervals at which the beginning and end of recovery were determined. The slope values obtained from each regression analysis of the rising portion of the refractory period curve for each subject are also included.

All effectiveness values that fell within the C-T intervals marking the beginning and end of recovery were subjected to an analysis of variance approach to regression (Neter, Wasserman, and Kutner, 1990). This method determines whether the CPu and MPFC refractory period data are best represented by a single

Table 1

Summary of results from Experiment 2

SUBJECT	SITE	CPu			MPFC		
		$C\text{-}T_s$	$C\text{-}T_L$	slope	$C\text{-}T_s$	$C\text{-}T_L$	slope
F2	S4	0.3	2.0	0.21			
F2	S5	1.0	*	0.03			
F2	S6	0.09	5.0	0.11			
F2	S7	1.6	10.0	0.03			
F2	S8	0.6	5.0	0.12			
CP18	S3	0.06	5.0	0.14			
CP18	S4	0.1	5.0	0.11			
CP18	S0				3.9	5.0	0.09
CP22	S7	0.3	5.0	0.09			
CP22	S8	1.1	12.0	0.10			
CP22	S9	0.4	5.0	0.20			
CP22	S1				0.8	5.0	0.15
CP22	S2				0.7	5.0	0.16
CP22	S3				0.2	10.0	0.06
CP46	S2	1.6	5.0	0.05			
CP46	S3	1.6	*	0.03			
CP46	S0				1.7	10.0	0.03
CP46	S1				1.0	10.0	0.02
CH58	S0				2.4	12.0	0.08
CP62	S12	0.3	5.0	0.15			
CP62	S13	1.0	2.0	0.37			
CP62	S3				1.2	5.0	0.08
CP64	S4	1.3	5.0	0.12			
CP64	S0				2.8	10.0	0.06
CP64	S1				0.3	10.0	0.08
CP65	S3	0.7	5.0	0.13			
CP65	S3				0.5	5.0	0.10
average		0.8	5.4	0.12	1.4	7.9	0.08
S.E.M		0.1	0.7	0.02	0.4	0.9	0.01

$C\text{-}T_s$, shortest C-T interval before the rise, estimated by a weighted regression analysis;
$C\text{-}T_L$, longest C-T interval estimated by the asymptote test;
slope refers to the slope of the regression line fitted to the rising portion of the refractory period curve;

*, missing values in cases where no plateau was evident.

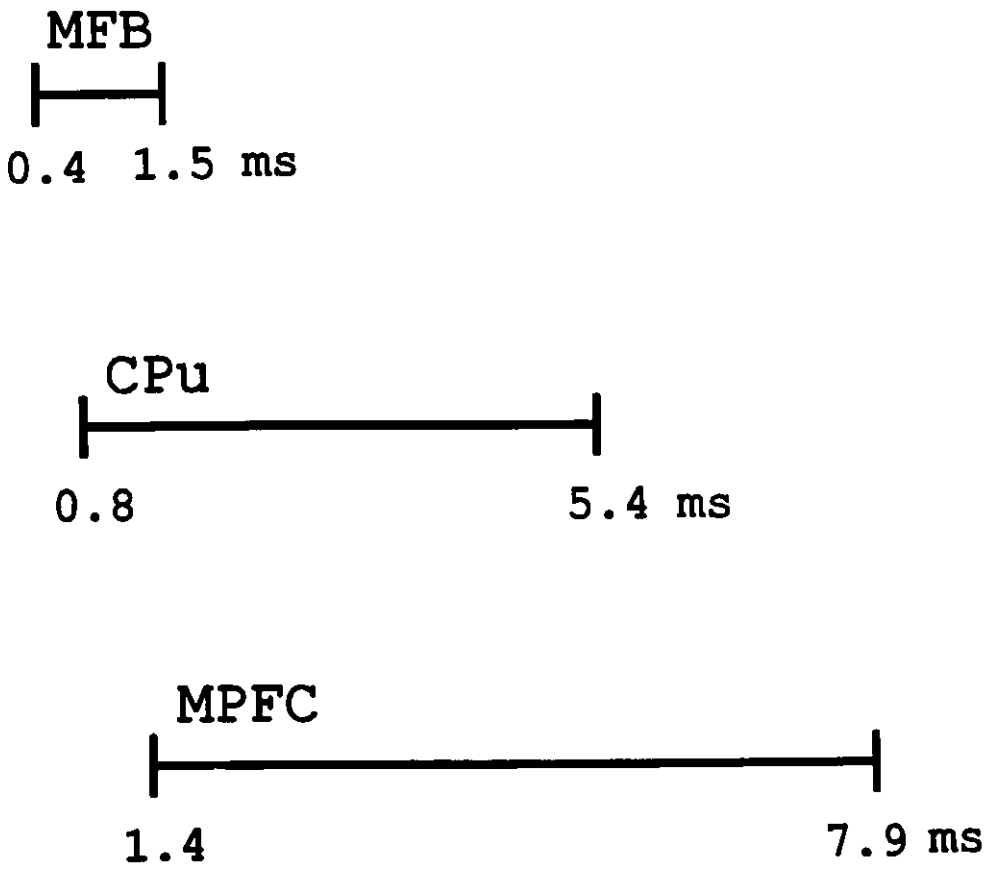
regression line or by two separate lines. A significant difference was found between the two regression lines fit to the CPu and MPFC data [$F(3, 150) = 62.9$, $p < 0.01$], thus indicating that the rising portion of the functions relating effectiveness to C-T intervals for each site are best represented by two separate lines. The subsequent post-hoc analysis determined which regression parameters contributed to this difference. Although no significant difference was found between the intercepts of the two lines, which defined the beginning of recovery, the t-tests confirmed that the slopes of the two curves were significantly different [$t(150) = 9.05$, $p < 0.01$)]. The steepness of the slope is indicative of the rate of recovery from refractoriness and also the homogeneity of the stimulated axon bundle (Bielajew, 1983; Yeomans, 1979; Yeomans et al., 1985). The initial rising portion of the refractory period curve indicates the completion of the absolute refractory periods of the fastest, largest neurons, while the later portion reflects the relative refractory periods and/or the contribution of neurons with longer absolute refractory periods (Yeomans, 1979). The average slope value for the CPu was greater than that for the MPFC. In addition, the C-T intervals marking the end of recovery from refractoriness were significantly different, $t(22) = 2.24$, $p < 0.05$. Hence, it appears that the slopes which reflect the rate of recovery and the C-T intervals at which recovery is defined to be complete are the two factors which contribute to the difference in the CPu and MPFC refractory period data.

Figure 12 compares the range of recovery from refractoriness for the MFB, CPu, and MPFC reward sites, former being a compilation of the refractory period data published in other laboratories. Although there is some overlap in the values marking the beginning of recovery for all three regions, it is particularly apparent that the end of recovery becomes progressively delayed in the rostrocaudal direction, that is from the MFB towards the CPu and MPFC.

Discussion

The present results indicate that the behaviourally derived refractory periods underlying MPFC and CPu self-stimulation tend to be longer than the values typically obtained for MFB reward structures (Bielajew and Shizgal, 1982; Bielajew et al., 1982, 1987a, 1987b; Bielajew and Fouriezos, 1985; Fouriezos et al., 1987; Panagis et al., 1995b; Rompré and Miliaressis, 1980, 1985; Schenk and Shizgal, 1982; Shizgal et al., 1980; Silva et al., 1982; Szabo et al., 1974; Trzcińska and Bielajew, 1992; Yeomans, 1975, 1979). That is, they both begin and end later, ranging on average from 0.8 to 5.4 ms in the CPu and 1.4 to 7.9 ms in the MPFC. In previous studies it was reported that recovery from refractoriness for the MPFC, basal forebrain, and some ventral pallidum sites ranged from 1.0 to 5.0 ms (Fouriezos el al., 1987; Panagis et al., 1995b; Schenk and Shizgal, 1982) and ended as late as 10.0 ms in the mediodorsal thalamus (Bielajew and

Figure 12. An illustration of the range of refractoriness obtained at most MFB placements (see Yeomans, 1990a for review), and in the CPu and MPFC regions evaluated in the present study.


MFB
0.4 1.5 ms
CPu
0.8
5.4 ms
MPFC
1.4
7.9 ms

Fouriezos, 1985). Since fiber diameter and conduction velocity appear to correlate with refractory periods (Swadlow and Waxman, 1978; Yeomans, 1990), the long estimates of recovery from refractoriness in the MPFC and CPu suggest that the stimulated elements are small, slowly conducting neurons.

Interestingly, the values obtained here are consistent with the range of recovery of possible dopaminergic neurons. The range of recovery from refractoriness of these neurons estimated behaviourally by using smaller electrode tips and larger currents (Yeomans, 1989; Yeomans et al., 1988) is similar to the data obtained electrophysiologically (Deniau et al., 1980; German et al., 1980; Lindvall and Björklund, 1974; Moore and Bloom, 1978; Wang, 1981) and span between 1.2 and 5.0 ms. These results are not surprising in light of the pattern of dopaminergic innervation in forebrain regions (Berger et al., 1991; Graybiel, 1990). There is ample evidence suggesting that the distribution of the MPFC and CPu regions supporting self-stimulation overlaps with catecholaminergic innervation in these areas (Kolb, 1990; McGregor et al., 1989; Sesack and Pickel, 1992).

The statistical analyses indicate that the rate at which the neurons underlying MPFC and CPu self-stimulation recover from refractoriness, as well as the time at which recovery is complete, both contribute significantly to the difference in the CPu and MPFC estimates, but not to the beginning of recovery which seems to occur at about the same time at both sites. Thus while there is quite a lot of overlap between the range of

refractory period estimates at the two sites, recovery at the MPFC sites continues well beyond that of the CPu.

One hypothesis to account for this difference may reflect the stimulation in the MPFC of somas or initial segments, which are known to be characterized by a longer recovery from refractoriness than axonal segments. Indeed, anatomical studies support this idea in that it has been well documented that cell bodies which form the major corticostriatal projections are contained in the MPFC pyramidal cells (DeFilipe and Farinas, 1992; Feldman, 1984; Fonnum, 1984; Fonnum, Storm-Mathisen, and Divac, 1981).

Alternatively, these differences may be explained by a greater contribution of either longer relative or absolute refractory periods to the time course of recovery from refractoriness in the CPu and MPFC (Bielajew et al., 1982; Schenk, 1982; Yeomans, 1979). These differences may suggest that a heterogenous population of neurons underlies MPFC and CPu self-stimulation and that only some of the stimulated neurons share similar characteristics (Yeomans, 1990). Because equal pulse stimulation was used here, the separate contributions of the absolute and relative refractory periods cannot be discerned. In order to accomplish this task, the unequal pulse technique must be employed (Bielajew et al., 1982; Yeomans, 1979, 1990), (see page 29).

The majority of the maximum effectiveness values did not approach the expected theoretical value of 1.0 or 100%. This

phenomenon has also been observed for other sites, including the lateral hypothalamus, ventral tegmental area, and dorsal pons (Bielajew et al., 1981, 1982; Schenk and Shizgal, 1982; Skelton and Shizgal, 1980; Yeomans, 1979; Yeomans and Davis, 1975). Interestingly, it has been recently shown that in order to obtain effectiveness values of 100%, one must test C-T intervals greater than 25 ms, at least at the level of the MFB (Fournier et al., 1991). Whether such effects reflect axonal properties remains controversial. In the present study in order to prevent the C-T interval from exceeding half of the C-C interval (reciprocal of frequency), the longest C-T interval that could be tested was 15 ms. Increasing the current, which would allow one to scale longer C-C intervals, and hence evaluate longer C-T intervals was not possible because of the epileptogenic activity observed at high currents.

However, even if such longer C-T intervals could have been tested, it is doubtful that true axonal excitability properties are observed in such cases, because values longer than 10 ms are outside the known physiological range of recovery from refractoriness for most neurons (Yeomans, 1990a). In addition, when axons are stimulated repeatedly, as is the case at longer C-T intervals, the subnormal and supernormal periods become more prominent, especially in slow-conducting fibers (Yeomans, 1990b) In such cases synaptic factors can become important, because the subnormal period slows the conduction of a T-pulse, thus allowing the C-T interval to persist long enough to affect the release of

a neurotransmitter (Yeomans, 1990a).

It is possible that the high currents used in this study evoke other populations of neurons which somehow interfere with the measurement of refractory period in reward neurons. Such populations may recover at progressively longer C-T intervals (Yeomans, 1990a), and hence affect the maximum effectiveness values. Furthermore, it has been shown that when high currents are used, high-threshold axons with longer refractory periods are recruited (Brown, 1992; Yeomans et al., 1985). Additionally, inhibitory pathways might be responsible for a failure to observe increases in effectiveness values at longer C-T intervals (Porrino, Coons, and MacGregor, 1983; Yeomans, 1975, 1990a). The high currents could evoke a response in a neighbouring population of neurons that have a suppressive effect on reward fibers, a phenomenon that has been reported in lateral and medial hypothalamic placements (Porrino et al., 1983).

Finally, it is also conceivable that progressively longer C-T intervals overstimulate seizure-prone neurons that have just recovered from refractoriness. Such C-T intervals might thus evoke subseizures, and as a consequence increase the thresholds of the reward-relevant population. For example, if the single pulse threshold (FT_{SP}) is equal to 10 and the double pulse threshold (FT_{C-T}) at a C-T interval of 10.0 ms is equal to 5, then the effectiveness value at that interval will be equal to 1.0 (10/5 - 1), or 100%. However, if the double pulse threshold is increased due to subseizures and is now equal to 7, then the

resulting effectiveness value will be equal to 0.43 (10/7 - 1), or 43%, which constitutes a 57% decrease in effectiveness at a C-T interval of 10 ms.

In conclusion, one explanation for the similarity in the MPFC and CPu estimates of recovery from refractoriness is that these areas constitute two unrelated neuronal populations with coincidentally similar physiological characteristics. Another idea is that these areas constitute two locations along the same reward bundle, and it is this hypothesis that is explored in the third experiment.

EXPERIMENT 3

The MPFC and CPu exhibit comparable behavioural and electrophysiological characteristics when electrically stimulated. For example, they are similar with respect to rate of self-stimulation acquisition (Corbett et al., 1982b, 1985), level of arousal associated with the stimulation (Schenk and Shizgal, 1982; Zacharko et al., 1990), currents used to elicit self-stimulation (Schenk et al., 1985), their [^{14}C]-2-deoxyglucose autoradiography patterns (Yadin et al., 1983), and degree of epileptogenic activity (Corbett, 1990). In addition, there is some evidence that lesions of either the MPFC or CPu produce qualitatively similar deficits (Brutkowski, 1965; Kolb, 1984). Anatomical studies have shown that descending MPFC fibers cross the medial CPu and that the orbitofrontal and dorsolateral areas of the MPFC project to the corresponding regions of the CPu. The evidence is less clear concerning the ascending CPu fibers that may contribute to MPFC self-stimulation.

In the previous experiment it was found that there were also similarities in the refractory periods of the directly stimulated neurons underlying MPFC and CPu self-stimulation, ranging from 0.8 to 5.4 ms for the CPu, and from 1.4 to 7.9 ms for the MPFC. While these values do overlap the behaviourally derived refractory periods observed at most MFB sites, their range is significantly longer, in that they begin and end later. In addition, the similarity in the refractory period profiles

between the MPFC and CPu suggests that these two regions may form part of the same reward substrate. In order to investigate this possibility, the behavioural adaptation of the collision test was employed to evaluate whether a direct axonal link exists between unilateral sites that support self-stimulation in the CPu and MPFC.

Method

Subjects

Thirty of the 67 subjects were used for the collision experiment.

Behavioural tests

Once stable responding was obtained on both sites, the current on each electrode was adjusted to produce the closest match between frequency thresholds at the two sites. The reason for this criterion was to assure that the two sites contributed equally to the effects of double-pulse stimulation. Each session began and ended with a single-pulse threshold determination at each site. In the double-pulse condition, pairs of C and T pulses were delivered to the two electrodes, either with the C pulses to the anterior (MPFC) electrode, and the T pulses to the posterior (CPu) electrode (AP condition), or in the reverse order, (PA condition). In general, C-T intervals of 0.2 to 20.0 ms were evaluated across subjects. They were administered in a

descending and ascending order, and counterbalanced across the four AP and four PA replications. Once the collision test was completed for a given pair of CPu and MPFC sites, either one or both electrodes were moved 0.32 mm, and the whole procedure repeated. The effectiveness of double-pulse stimulation was assessed using the following formula (Shizgal et al., 1980; Yeomans, 1979):

$$\mathbf{E = (FTsp_L/FT_{C\text{-}T}) - 1 \;/\; (FTsp_L/FTsp_H)} \quad \text{where}$$

E = effectiveness of double-pulse stimulation at a particular C-T interval,

$\mathbf{FTsp_L}$ = average frequency threshold of single pulses, obtained from the site yielding the lower threshold value,

$\mathbf{FTsp_H}$ = average frequency threshold of single pulses, obtained from the site yielding the higher threshold value,

$\mathbf{FT_{C\text{-}T}}$ = frequency threshold of double pulses at a given C-T interval.

The term $FTsp_L/FTsp_H$ in the denominator is a correction factor to account for the difference in the single-pulse threshold values obtained from the two placements. An E of 0 indicates that double-pulse stimulation is no more effective than trains of single pulses, so that the frequency threshold of double pulses (C and T pulses) is identical to the frequency threshold of single pulses (T pulses absent). An E of 1 indicates that

double-pulse stimulation is just as effective as trains of single pulses; thus, the frequency threshold of double-pulses is equal to half of the frequency threshold of single pulses.

Results

Histology

Figure 13 a-g illustrates the CPu and MPFC placements evaluated for axonal connectivity using the behavioural adaptation of the collision test. The same figure (right column) also features the average effectiveness values obtained in the AP and PA conditions associated with the pair of sites drawn in the left column. In total, there were nine pairs of ipsilateral and two pairs of contralateral sites tested in this study.

In most subjects, the striatal electrode tips were located in the anterior medial CPu, and in one case, subject CP62, the anterior ventrolateral CPu. The MPFC placements were found in the anterior medial MPFC, specifically in the Cg3 area, and in the Fr1/Fr2 region for subject CP46.

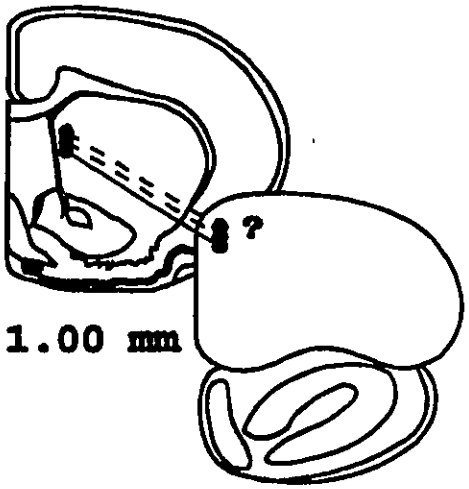
Collision test

Following the visual inspection of the data all functions were subjected to a repeated measures ANOVA analysis. Collision was inferred from statistical results that showed a significant main effect of C-T interval and no main effect of order and no interaction. Recall that the pattern constituting the signature

Figure 13 a-g. Neurographics plates that best correspond to the location of the eleven pairs of CPu (background plate) and MPFC (overlapping plate) electrode tips, which are indicated by filled circles. The consecutive sites are 0.32 mm apart. The question mark for subject CP22 reflects the uncertainty as to the exact location of that MPFC electrode tip. Panels a-e show the five ipsilateral CPu and MPFC placements and panels f and g, the two contralateral ones. Those pairs of sites which were determined to give rise to a collision-like effect are linked by solid lines and the remaining pairs in which collision-like effects were not observed, by broken lines. The anteroposterior coordinates based on the Paxinos and Watson (1986) atlas, express the distance of the plate (mm) from bregma; these are listed at the bottom of each section. The alphanumeric positioned in the upper left corner of the page identifies each subject. The effectiveness values for the collapsed AP and PA conditions are plotted as a function of C-T interval and are located on the right hand side of each illustration. Because there was a significant interaction between the AP and PA conditions in subject CP46, the two curves that represent those conditions are shown separately. The specific depth number of each pair of sites (recall that 0 refers to initial implantation site, 1 to 0.32 mm lower, etc) and the currents employed are listed at the top of each effectiveness function.

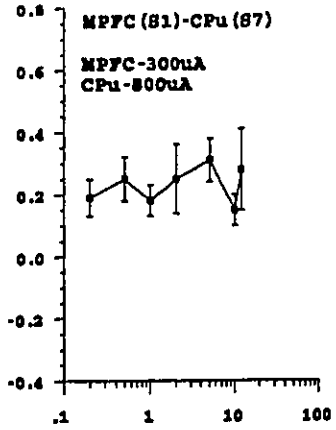
a)

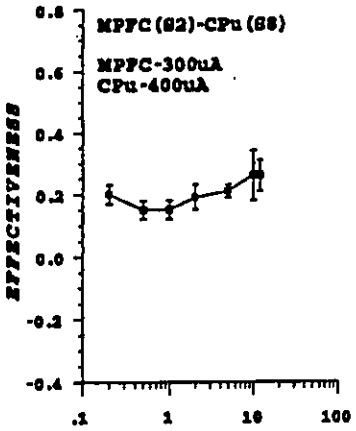
CP22

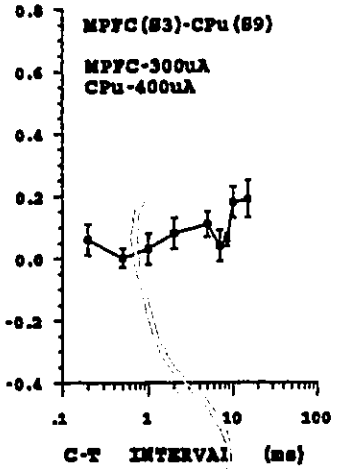


1.00 mm

5.20 mm

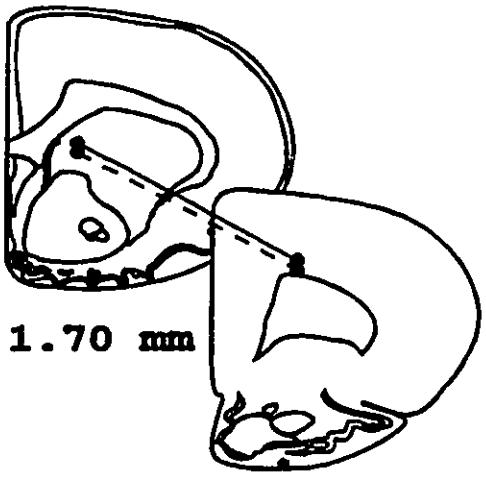






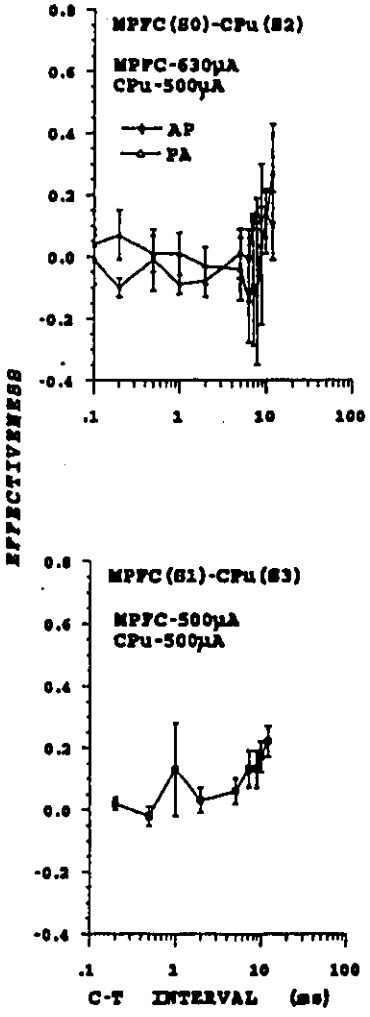
b)

CP46



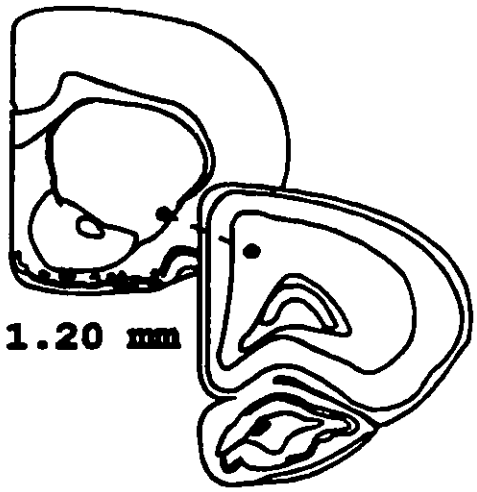
1.70 mm

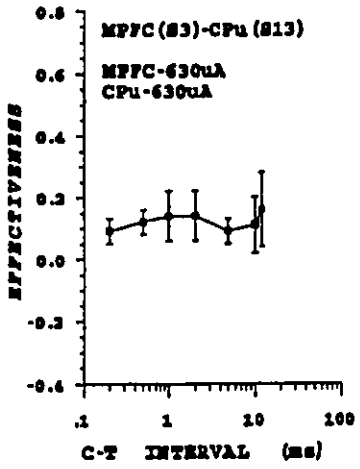
2.70 mm



c)

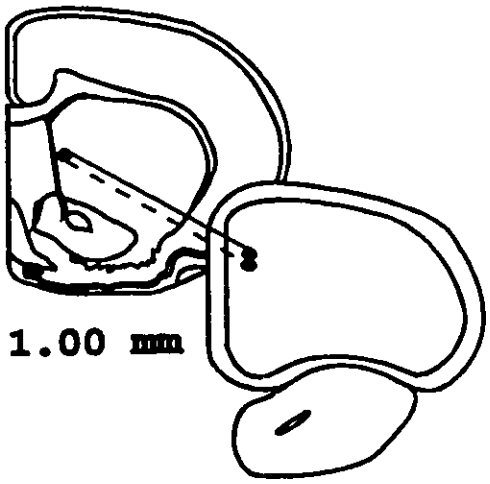
CP62


1.20 mm
3.20 mm


MPFC(S3)-CPu(S13)
MPFC-630uA
CPu-630uA
EFFECTIVENESS
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

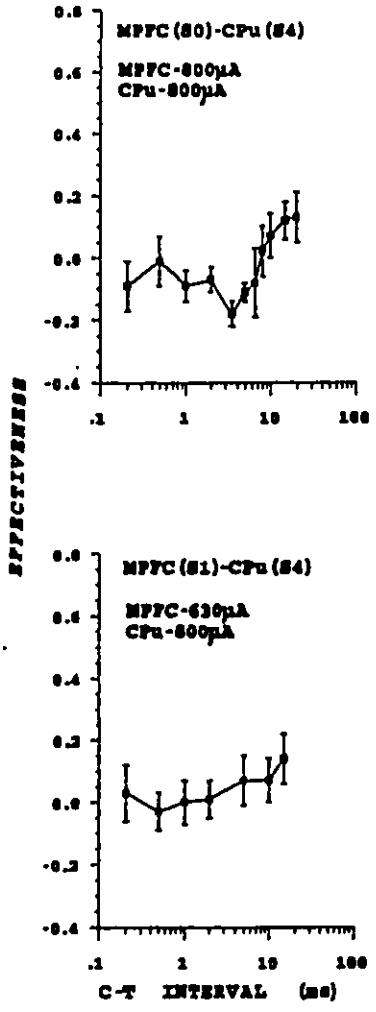
d)

CP64



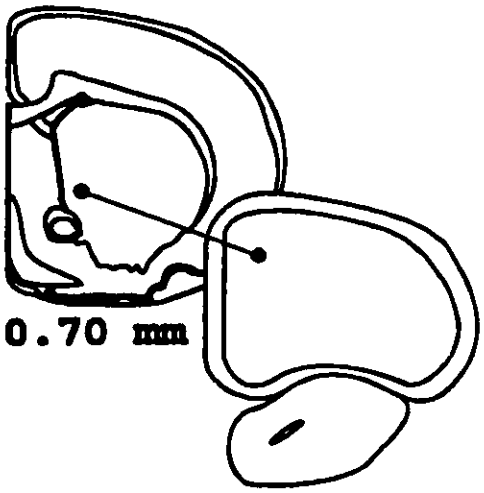
1.00 mm

4.70 mm

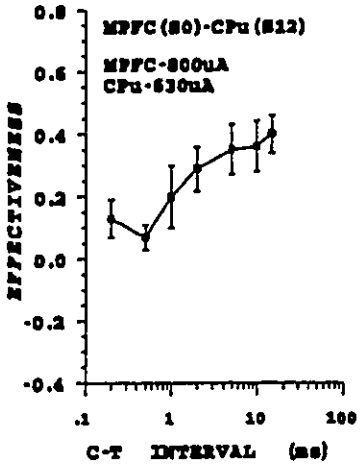


e)

CP66

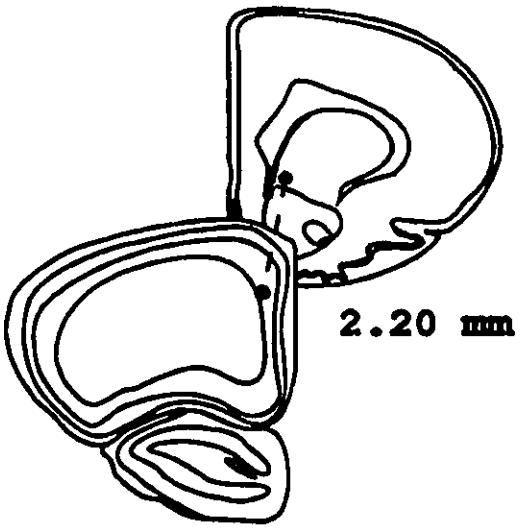

0.70 mm

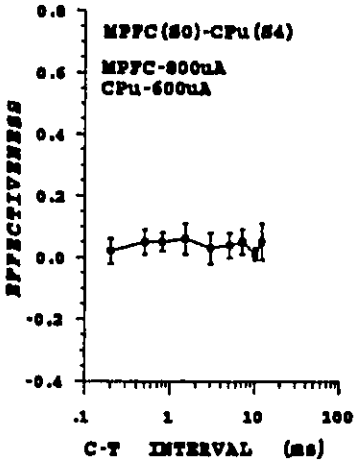
4.70 mm


MPFC(80)-CPu(812)
MPFC-800uA
CPu-630uA
EFFECTIVENESS
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

f)

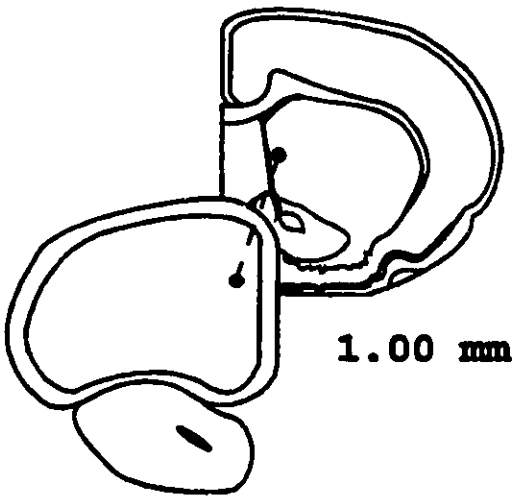
CP18


2.20 mm
4.20 mm

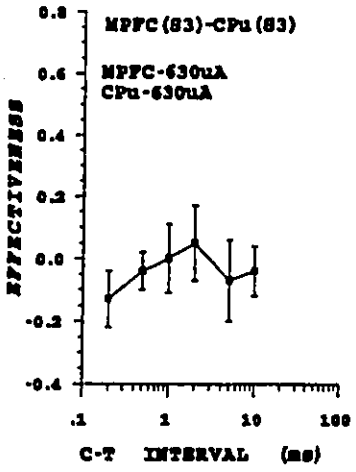

MPFC(S0)-CPu(S4)
MPFC-600uA
CPu-600uA
EFFECTIVENESS
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

g)

CP65


1.00 mm

4.70 mm


MPFC(83)-CPu(83)
MPFC-630uA
CPu-630uA
EFFECTIVENESS
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
.1
1
10
100
C-T INTERVAL (ms)

of collision has a step-like function characterized by low effectiveness values at short C-T intervals, followed by a sharp rise and a plateau at longer C-T intervals. Table 2 lists the results of that analysis; the C-T interval factor was significant and the order of presentation or condition was not in four of the nine ipsilateral pairs of sites tested, with one exception (subject CP46). Despite a significant interaction between the two conditions in this subject it was decided to include the data in the calculations of conduction velocity, because both cases and both the AP and PA curves showed a similar step-like profile and great overlap in their collision intervals. Although repeated measures tests should correct for violations in sphericity (Barcikowski and Robey, 1984), using this approach here resulted in relevant significant effects in only two of the four pairs of sites. Since visual inspection of curves suggested a step-like function in all cases, it was decided to base the interpretations of this experiment on a less conservative analysis.

The significant main effect of C-T interval was due to a step-like increase in effectiveness values between short and long C-T intervals, which is interpreted as reflecting some degree of functional connectivity between these sites (Bielajew, 1983; Bielajew and Shizgal, 1980, 1982; Boucher-Thrasher, 1986; Bushnik-Harris, 1993; Durivage and Miliaressis, 1987; Gratton and Wise, 1988b; Shizgal et al., 1980; Shizgal, 1989). The size of the collision effect, estimated from the difference between the

Table 2

Results of repeated measures ANOVA

Source	df	MS	F	p
CP22/MPFC(S3)-CPu(S9)				
C-T Interval (C-T)	8	.033	2.436	.0438*
Order of presentation of pulses (AP/PA)	1	.073	7.317	.0735
Interaction (C-T X AP/PA)	8	.009	1.140	.3734
CP46/MPFC(S0)-CPu(S2)				
C-T Interval (C-T)	11	.068	3.808	.0004**
Order of presentation of pulses (AP/PA)	1	.011	.1400	.7240
Interaction (C-T X AP/PA)	11	.056	3.712	.0006*
CP64/MPFC(S0)-CPu(S4)				
C-T Interval (C-T)	10	.075	2.484	.0264*
Order of presentation of pulses (AP/PA)	1	.279	2.701	.1988
Interaction (C-T X AP/PA)	10	.013	.8070	.6239
CP66/MPFC(S0)-CPu(S12)				
C-T Interval (C-T)	6	.126	6.472	.0009**
Order of presentation pulses (AP/PA)	1	.003	.0300	.8734
Interaction (C-T X AP/PA)	6	.015	.4240	.8534

* $p < .05$, ** $p < .01$

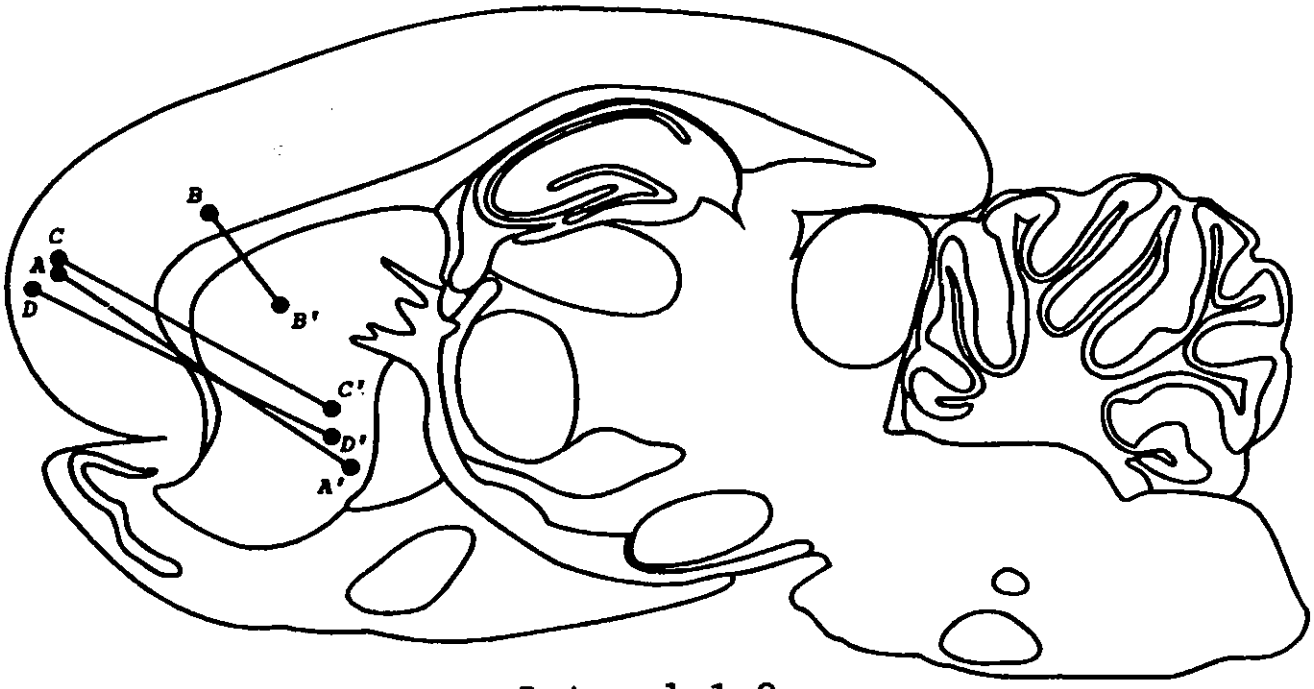
peak and minimum effectiveness values (Yeomans, 1990a), ranged from 15 to 33% across the four animals.

Figure 14 illustrates the anatomical distribution of the four CPu and MPFC pairs that were found to give rise to collision-like effects, drawn in the sagittal plane. Although the lateral coordinate was different for each electrode placement, a representative section of 1.2 mm lateral to the midsagittal suture was chosen for illustrative purposes.

Statistical analysis on the remaining five ispilateral and two contralateral curves showed no main effect of C-T interval. In these cases, concurrent delivery of double pulses resulted in a 14% summation for the ipsilateral pairs (4 to 23% range), and a 4% summation for the bilateral ones.

Table 3 lists the average effectiveness values collapsed across C-T intervals for the five ipsilateral and two contralateral conditions which did not show the characteristic step-like collision profile, and the collision intervals, conduction times, inter-electrode distances, and conduction velocities for the functions consistent with the collision model. The collision interval refers to the longest C-T interval before the rise in effectiveness. In one case, (subject CP66) the collision interval was estimated by the asymptote test, described earlier (see page 106), because it appeared to be shorter than the shortest estimate of the beginning of recovery from refractoriness. Conduction time was calculated by subtracting the refractory period estimate of the site, associated with the

Figure 14. A Neurographics sagittal section at 1.2mm showing the locations of the CPu and MPFC electrode pairs in the four cases that were identified to be associated with collision-like effects. The letters without the prime symbol refer to the MPFC and those with the symbol, to the CPu region.



LEGEND:

A-A' = CP22
B-B' = CP46
C-C' = CP64
D-D' = CP66

Lateral 1.2 mm

Table 3

Summary of collision test results

Rat	Sites	average E ± S.E.M	collision interval (ms)	conduction time (ms)	inter-electrode distance (mm)	conduction velocity (m/s)
Ipsilateral pairs						
CP22	S1-S7	0.23±0.03				
CP22	S2-S8	0.20±0.02				
CP22	S3-S9	collision	7.0	6.0	1.8	0.3
CP46	S0-S2	collision	7.1	6.6	1.4	0.2
CP46	S1-S3	0.10±0.02				
CP62	S3-S13	0.12±0.03				
CP64	S0-S4	collision	3.5	2.5	1.4	0.6
CP64	S1-S4	0.04±0.03				
CP66	S0-S12	collision	2.0	1.5	2.7	1.8
Contralateral pairs						
CP18	S0-S4	0.04±0.01				
CP65	S3-S3	0.04±0.04				

N.B. Subject CP66 lost his electrode assembly before the completion of the refractory period test. The conduction time was thus calculated on the basis of the average refractory period obtained from collapsing one MPFC and two CPu sessions.

longest value from the collision interval. The inter-electrode distance of individual pairs of MPFC and CPu sites was estimated by geometrical interpolation, from the histologically verified lateral and ventral coordinates. The conduction velocities were obtained by dividing the inter-electrode distance by the conduction time.

Across animals, the collision intervals ranged between 2.0 and 7.1 ms, conduction time from 1.7 to 7.1 ms, and the inter-electrode distance between 1.4 and 2.7 mm. Given the inter-electrode distance and the conduction time, conduction velocity for the MPFC and CPu reward fibers was estimated to range between 0.2 and 1.8 m/s, with a 0.7 m/s average. This represents the slowest estimate since it is assumed that the reward fibers coursing between any two self-stimulation sites are linked by a straight line.

Discussion

Collision-like profiles

In order for the function relating effectiveness of double pulse stimulation as a consequence of C-T interval to be consistent with the interpretation of collision, it must have specific characteristics. Traditionally, all curves have been compared to those found for paired-pulse stimulation applied to the ventral tegmental and lateral hypothalamic areas, because these were the first sites used to evaluate the collision method

using a behavioural paradigm (Shizgal et al., 1980). Collision is inferred when double-pulse effectiveness is lower at short C-T intervals than at longer ones. That portion of the function where effectiveness values are lower is termed the collision interval and is believed to reflect the sum of the inter-electrode conduction time and the refractory period. Once this sum is exceeded, collision no longer occurs and effectiveness values suddenly rise and remain elevated for the remaining C-T intervals. In addition, regardless of the order of presentation of the pairs of pulses, that is, the AP or PA condition, the curve should be symmetrical due to the bidirectional conduction property of axons (see Figure 4 in the Introduction).

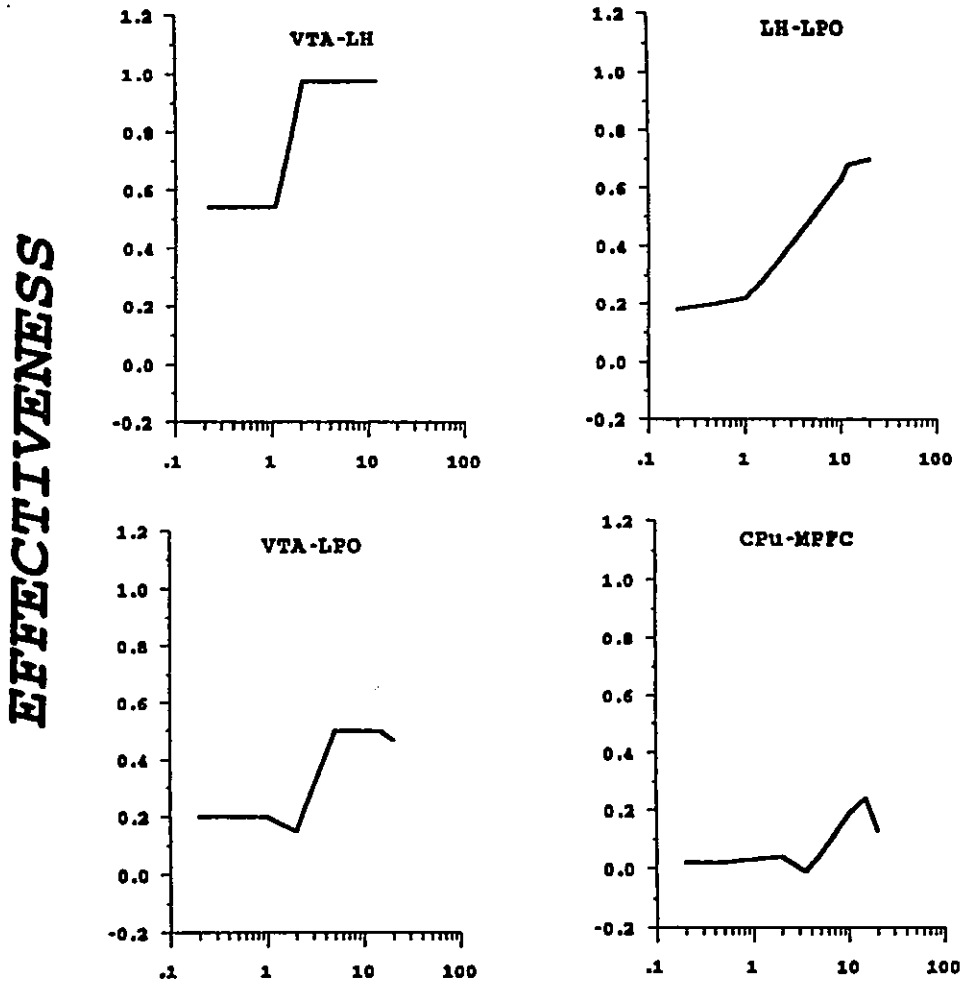
The shapes of the CPu and MPFC curves are generally consistent with such effects, and overall resemble that reported for other pairs of sites (see Figure 15) for which axonal connectivity has been observed (Bielajew, 1983; Bielajew and Shizgal, 1980, 1982; Boucher-Thrasher, 1986; Bushnik-Harris, 1993; Durivage and Miliaressis, 1987; Gratton and Wise, 1988b; Malette and Miliaressis, 1995; Murray and Shizgal., 1994; Shizgal et al., 1980; Shizgal, 1989). The step-like functions constituting the trademark of collision in the present study were based on statistical findings of significant effects of C-T interval and no significant difference between the AP and PA conditions or interaction between the two factors.

Despite these general findings there are several characteristics which distinguish the data reported here from

Figure 15. Representative effectiveness functions obtained from different intra-hemispheric pairs of self-stimulation sites at which collision-like effects have been reported. The VTA-LH pair is based on data from Durivage and Miliaressis (1987), Gratton and Wise (1988b), Malette and Miliaressis (1995); Murray and Shizgal (1994), and Shizgal et al., (1980); the LH-LPO summarizes Boucher-Thrasher (1986) and Bielajew et al.'s (1987b) collision results; the VTA-LPO panel comes from Bushnik-Harris (1993), and the CPu-MPFC pair is a composite of the data described here.

CPu: caudate-putamen
LH: lateral hypothalamus
LPO: lateral preoptic area
MPFC: medial prefrontal cortex
VTA: ventral tegmental area

INTRA-HEMISPHERIC COLLISION



that in the literature. They include the length of collision interval, gradual rise in effectiveness, the small size of the collision effect, roughly 20%, the lack of a clearly-defined plateau at the longer C-T intervals, and the low summation values (the latter are discussed at the end of the "Non-collision profiles" section on page 148).

Collision interval. The collision interval ranged from 2.0 to 7.1 ms, which distinguishes the collision-like profiles obtained in this study from earlier work. Previous reports indicate that the reward neurons along the MFB rise in effectiveness within 1.0 to 3.0 ms (Bielajew and Shizgal, 1982; Shizgal et al., 1980). However, the length of this interval is dependent on the distance between the two stimulating electrodes, as well as the conduction time of the inter-electrode segment. Thus, it is not surprising that the range of intervals at which collision occurs seems to increase in the posteroanterior direction (see Figure 15, top left to bottom right panels VTA-LH, LH-LPO, VTA-LPO and CPu-MPFC).

The longer collision intervals suggest that the reward substrate is composed of neurons with heterogenous excitability estimates and/or a wide range of conduction velocities (Bielajew and Shizgal, 1982; Durivage and Miliaressis, 1987; Murray and Shizgal, 1994; Shizgal et al., 1980). This observation is reflected in the CPu and MPFC refractory period estimates, which show recovery to be delayed until 5.4 and 7.9 ms respectively.

The recovery from refractoriness at the CPu placements was faster than at the MPFC. Thus, if the refractory period contributes to the collision interval, it does so more at the MPFC than CPu sites. A longer refractory period might result in a delayed rise in effectiveness at short C-T intervals based on an anatomical arrangement consisting of corticostriatal fibers that synapse on striatal neurons which would influence the PA more than the AP function. Further, it is also possible that the axonal bundle coursing between the MPFC and CPu thins out, which would affect the range of recovery from refractoriness and consequently the length of the collision interval, resulting in different AP and PA profiles. However, in this study no difference in the shape of the collision curves, regardless of the order of presentation of pulses, was observed, suggesting that the recovery of different populations of fibers, which are similarly distributed at both sites, rather than recovery from a unique population of fibers contributes to the shape of the collision profiles.

Conduction velocity. Conduction velocity is a cable characteristic that depends on fiber diameter and degree of myelination (Swadlow and Waxman, 1978). Larger myelinated fibers conduct faster than smaller unmyelinated fibers (Hursch, 1939; Szabo et al., 1974). The estimates of conduction velocity obtained in this study range from 0.2 to 1.8 m/s, with an average of 0.7 m/s, and are slower than the ones obtained for MFB reward fibers, which span values between 1.0 and 8.3 m/s (Bielajew,

1983; Bielajew and Shizgal, 1986; Shizgal et al., 1980). The former estimates are similar to those obtained for the unmyelinated dopaminergic fibers, which range from 0.3 to 1.5 m/s, with a 0.5 m/s average (German et al., 1980; Guyenet and Aghajanian, 1978; Yim and Mogenson, 1980). The estimates of conduction velocity, however, should be interpreted with caution as they are based on estimates of refractory period, inter-electrode distance and collision interval, each of which are subject to measurement error (Yeomans, 1990a).

The single-electrode curves reflecting the recovery from refractoriness are often not identical, as is the case here. These differences can occur when one electrode stimulates main fibers and the other one initial segments, collaterals, or axon terminals (Yeomans, 1990a). Interestingly, the MPFC sends direct and collateral projections to the CPu, which suggests that such effects might be seen here. Further, refractory period estimates of different populations of neurons often include local potential summation and supernormal period contributions, effects which may obscure the initial rise in effectiveness and the overall rate of recovery. The contribution of the subnormal period becomes more important in slow-conducting fibers and is believed to reflect synaptic activity especially at longer C-T intervals (Yeomans, 1990b). Recall that in order to calculate conduction time, the beginning of recovery from refractoriness is subtracted from the collision interval. Lastly, the estimates of inter-electrode distance are derived by geometrical interpolations from

histological verification of the electrode tips. The latter can be altered by distortions resulting from freezing and thawing of the tissue, and the alignement of the electrodes within the axonal bundle (Yeomans, 1990a). In addition, conduction velocity can vary along the length of an axon and hence further alter the estimates (Yeomans, 1995). Thus, the conduction velocity values estimated behaviourally might not accurately reflect the speed of conduction of the reward fibers.

The observation that the estimates of conduction velocity obtained in this study and those values usually found for dopaminergic neurons are similar warrants further elaboration. The consensus has long been that the catecholaminergic neurons constitute a later stage in the reward circuity (Shizgal et al., 1980; Wise, 1978, 1980), and instead of being directly activated in self-stimulation studies modulate the transmission of rewarding information (Gallistel, 1986; Gallistel and Freyd, 1987; Miliaressis et al., 1986, 1991). Moreover, some of the first stage neurons appear to carry the reward-relevant signal in a rostrocaudal direction, opposite to the direction of conduction of dopaminergic fibers (Bielajew, 1983; Bielajew and Shizgal, 1986). While this may characterize MFB reward neurons, it may not describe the properties of the forebrain reward substrate. It has been determined that self-stimulation obtained from MPFC is accompanied by an enhanced release of dopamine and its metabolites (Mora and Myers, 1977). Further, the CPu and ventromedial cortex of the rat receive dopaminergic afferents from the

ventral tegmental area (Beckstead, Domesick, and Nauta, 1979). In addition, the CPu accepts dopaminergic innervation from the substantia nigra pars compacta (Beckstead et al., 1979). Both of these dopaminergic signals travel in the caudorostral direction, but that does not suggest that the reward fibers are the same as these ascending dopaminergic fibers. On the basis of the data presented here, the direction of conduction in the reward-relevant bundle between the MPFC and CPu cannot be estimated. In order to accomplish that the technique of the hyperpolarization block would have to be employed (Bielajew and Shizgal, 1986).

Although it has been argued that pulse durations, longer than the ones employed in this experiment, are needed to stimulate dopaminergic cells, more recent reports (Bielajew, 1991; Gratton et al., 1988; Miliaressis et al., 1991) suggest that the region in which dopaminergic neurons can be excited around the electrode tip decreases as the pulse duration increases, at least at the level of the MFB. Some argue that dopaminergic cells can be fired transynaptically by the typical self-stimulation parameters (Gratton et al., 1988; Yeomans, et al., 1988). In fact, dopamine can be released by such parameters and the size of this effect depends on current and frequency (Nicolaysen, Ikeda, Justice, and Neill, 1988). It is thus possible that at least some of the fibers contributing to the estimates of conduction velocity could be dopaminergic.

Another neurotransmitter which could also influence the estimates of conduction velocity is glutamate. From anatomical

studies it has been determined that pyramidal cells forming corticostriatal projections seem to be glutamatergic and aspartergic in nature (Conti, Rustioni, Petrusz, and Towle, 1987; Dinopoulus, Dori, Davies, and Parnavelas, 1989). Interestingly, when the corticostriatal projection area, the same region often confirmed in behavioural studies to support MPFC and CPu self-stimulation, is antidromically activated, conduction velocities ranging from 0.6 to 0.8 m/s are obtained (Ferino et al., 1987), values which are in agreement with the present behavioural estimates. From purely electrophysiological studies there is also evidence for phasic excitatory modulation of striatal dopaminergic release by corticostriatal neurons (Taber and Fibiger, 1993), suggesting an interaction between the two neurotransmitter systems.

Rising portion. The rising portion of the CPu and MPFC collision curves is gradual, resembling the refractory period curves in this respect, and contrasts with the sharp increases in effectiveness reported earlier for concurrent ventral tegmental-lateral hypothalamic area stimulation (Bielajew and Shizgal, 1982, 1986; Durivage and Miliaressis, 1987; Gratton and Wise, 1988b; Shizgal et al., 1980). Recently, Murray and Shizgal (1994) argue that the range of C-T intervals used in earlier studies had greater resolution, and thus allowed for the observation of sharper changes in effectiveness values as C-T interval was increased. In addition, they point out that the

inadequate sampling of the rate-number functions might have contributed to a steeper rise in effectiveness in the first collision studies. The rate-number curves from which collision functions are derived could have been artificially steepened by selecting pulse numbers that were greater than one-half of the interval over which the rate-number function rose.

In the original collision studies, the number of pulses in the train was decreased in 0.1 $\log_{10}$ unit steps in the single and double pulse determinations, whereas in Murray and Shizgal's (1994) and Boucher-Thrasher's (1986) studies, as well as in the present case, the number of pulses was decreased in 0.05 $\log_{10}$ unit steps. In contrast, the resolution used by Bielajew and Shizgal, 1982, 1986, Bushnik-Harris (1993), and Shizgal et al. (1980) was 0.1 $\log_{10}$ unit steps. Finally, the use of a more stringent pre-screening criterium, by selecting only those subjects that showed a sharp rise in effectiveness values, might have also explained why only selective patterns were observed in earlier studies (Bielajew and Shizgal, 1982, 1986; Bushnik-Harris, 1993; Shizgal et al., 1980). Indeed, it appears that the slopes of the functions based on 0.1 $\log_{10}$ unit steps (VTA-LH and VTA-LPO pairs) are steeper than the ones based on 0.05 $\log_{10}$ unit steps (LH-LPO and CPu-MPFC pairs).

Size of the effect. Generally, the size of the collision effect (the difference in the effectiveness values between the point at which the curve begins to rise and the value at which it

approaches an asymptote) has been reported to be roughly 45% for the lateral hypothalamic and ventral tegmental area placements (Huntrage and Milliaressis, 1987; Gratton and Wise, 1988b; Milliaressis and Malette, 1995; Murray and Shizgal, 1994; Shizgal et al., 1993), 40% for the lateral preoptic area (Boucher-Thrasher, 1986) and ventral tegmental and lateral preoptic areas (Bushnik-Harris, 1993). Theoretically, if all of the reward-relevant fibers course between the two stimulation fields, effectiveness values of 100% should be obtained (please refer to Figure 4). However, the failure to obtain this in the majority of the collision curves suggests that collision did not occur in all stimulated axons. For that to occur, it would require that the electrodes be perfectly aligned within the fiber bundle, such that all stimulated axons course through both effective stimulation fields (Bielajew and Shizgal, 1982; Yeomans, 1990a). Interestingly, the magnitude of the collision effect gradually diminishes from roughly 45% to 20% in the ventral tegmental-lateral hypothalamic, lateral hypothalamic-lateral preoptic, ventral tegmental-lateral preoptic, and dorsal pairs, suggesting that the distribution of the reward-relevant fibers undergoing collision and possibly their anatomical location in relation to the stimulating electrodes changes in the posterodorsal direction. It should be noted that increases in effectiveness might have been observed in these pairs had longer C-C intervals been tested. However, this option was not possible here, because of the range of C-C

intervals used.

Lastly, the transynaptic and mixed collision models cannot explain the present results, because these models require a specific difference in AP and PA curves, as illustrated in Figure 5. In the present experiment no significant difference was seen between the two conditions, except for subject CP46. However, in this particular case the shape of the two conditions was very similar, and the rise in effectiveness seen at longer C-T intervals occurred at the same time.

Non-collision profiles

Ipsilateral pairs

Not all functions relating the changes in effectiveness as a function of C-T interval were consistent with a collision profile in this study. In five of the nine cases in which stimulation was concurrently delivered to the two sites, the effectiveness values showed little variability across C-T interval. The overall shape resembled a flat line, similar to the one described in Figure 4. These flat, non-zero functions have been explained as representing either a misalignment of stimulation field, or activation of two fiber bundles with converging outputs (Bielajew et al., 1981; Schenk and Shizgal, 1982). In the former case, increasing the current, or moving one or both electrodes has been shown to give rise to a collision effect in some pairs of sites (Bielajew, 1983; Bielajew and Shizgal, 1982, 1986; Bushnik-Harris, 1993). In the present experiment, a move of merely 0.32

mm at one of the two sites resulted in the emergence of a collision-like function (see Figure 13 a and d).

Contralateral pairs

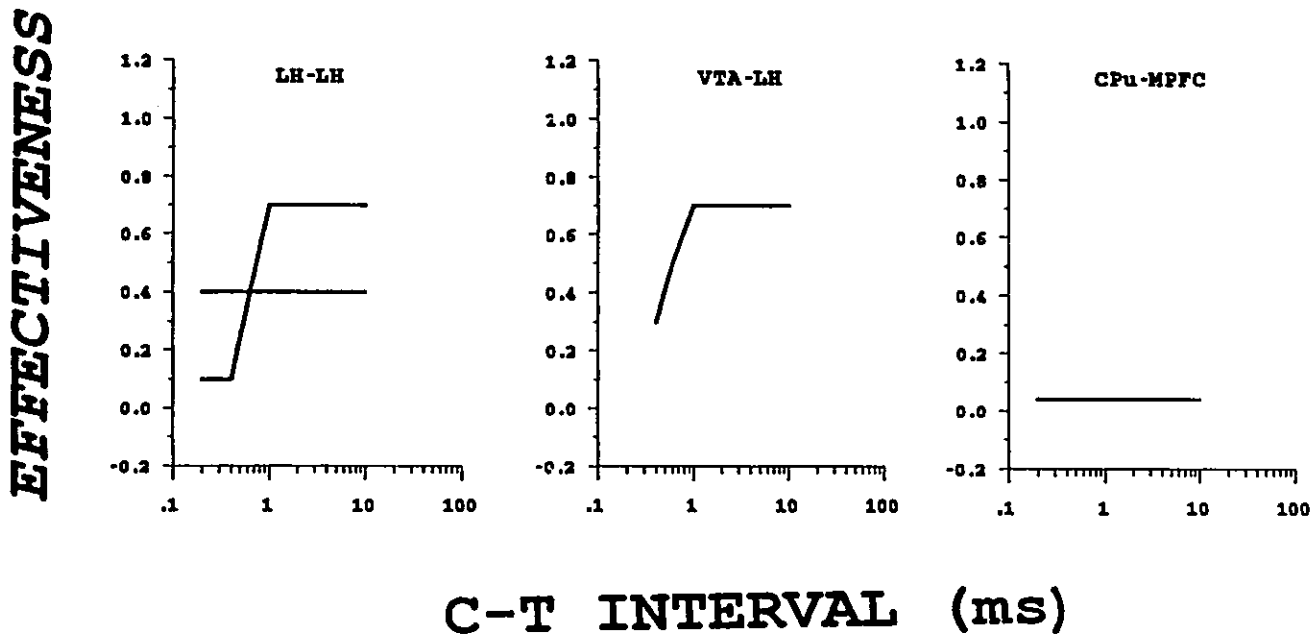
When concurrent stimulation is delivered to contralateral structures, a variety of patterns is observed, some of which resemble typical collision-like profiles (see Figure 16). For example, when bilateral lateral hypothalamic sites (panel LH-LH) were evaluated, either a pattern consistent with direct axonal connectivity was observed (Malette and Miliaressis, 1995), indicating the possible activation of collaterals, or a flat function, suggestive of some integration of the reward signals, but no direct axonal linkage (Bielajew and Shizgal, 1982, 1986; Shizgal et al., 1980). In other cases (panel VTA-LH), a curve similar to a collision-like effect was seen (rising portion and plateau), but without a clearly defined collision interval, and hence could not be interpreted as a collision effect (Malette and Miliaressis, 1995).

The pattern obtained for two CPu and MPFC pairs (panel CPu-MPFC) in this study is consistent with a flat function. Such a pattern is similar to the one typically seen for unilateral or intra-hemispheric sites at which no evidence of collision was obtained (Bielajew and Shizgal, 1982, 1986; Shizgal et al., 1980).

Interestingly, the data from Figures 13d and g are in agreement with the results of the ipsilateral and contralateral

Figure 16. Representation of the results of different collision studies where concurrent stimulation was applied to structures in different hemispheres. The straight line in the LH-LH panel was obtained from Shizgal et al. (1980) and the step-like function from Malette and Miliaressis, (1995). The VTA-LH curve is one of the representative graphs from Malette and Miliaressis (1995) and the CPu-MPFC panel represents a summary of the results obtained in this study. For explanation of the acronyms please refer to the caption of Figure 15.

INTER-HEMISPHERIC COLLISION



collision tests obtained in this study. The pairs of MPFC and CPu sites appear to be located in the same anatomical regions, except that subject CP65 was implanted with a contralateral set of electrodes and subject CP64 with ipsilateral ones. A collision effect was obtained for CP64, but not for CP65, further suggesting that contralateral fibers do not send direct axonal projections to the other hemisphere.

Transference and axonal connectivity

All of the ipsilateral MPFC-CPu pairs of sites for which significant collision effects were found were the same ones that gave rise to the transference effect. The sites (Figure 13 c and g - one ipsilateral and one contralateral pair) that did not show collision also failed to demonstrate transference of rewarding CPu stimulation to MPFC. However, these animals eventually did learn to bar press for MPFC stimulation after many training sessions on both electrodes. Although it is premature to draw any strong conclusion about a possible correlation between collision effects and transference, it might be interesting to further evaluate this relationship. If the MPFC and CPu sites are axonally linked, as suggested by these data, the experience on the CPu site would be simultaneously activating the MPFC one. If this factor facilitates acquisition, then such an arrangement would be more powerful than one in which the two sites are indirectly linked.

The problem of low summation

The summation levels obtained in this study were generally low. Traditionally, step-like collision functions reach effectiveness values of nearly 1.0 or 100% at longer C-T intervals (Bielajew, 1983; Bielajew and Shizgal, 1980, 1982; Boucher-Thrasher, 1986; Bushnik-Harris, 1993; Durivage and Miliaressis, 1987; Gratton and Wise, 1988; Shizgal et al., 1980; Shizgal, 1989). Similarly, summation levels found for the functions that are not consistent with a collision-like profile are usually higher. For example, 40 to 70% summation has been reported for concurrent stimulation applied to lateral hypothalamic and ventral tegmental self-stimulation sites (Shizgal et al., 1980) and 60% for the LH and periaqueductal gray regions (Bielajew et al., 1981), suggesting that these placements extensively integrate their reward signals. In other words, when the two electrodes activate different first stage neurons, the output of these neurons converges at some later point in the reward circuity. In contrast, the summation levels between the intra-hemispheric MPFC and CPu sites were roughly 14%, which indicates that the frontal cortex and striatal self-stimulation sites integrate poorly for some reason or relay their signals to different integrators. Similarly, the levels of summation found for inter-hemispheric pairs of CPu and MPFC sites had an average of 4%, indicating that roughly the same number of single as double pulses were necessary to maintain criterion responding. Hence, adding stimulation to the second site did not seem to

improve the rewarding effect produced by stimulation of either site alone, whether that site was located ipsilaterally or contralaterally, although ipsilateral concurrent stimulation was somewhat more effective. Since the contralateral MPFC and CPu pairs are transhemispherically and thus most likely transynaptically related, it is not surprising that stimulation at these two sites combines less perfectly than for ipsilateral MPFC and CPu pairs.

The low summation values reported here are similar to the ones found for paired pulse studies of the MPFC and cingulate cortex (Silva et al., 1982) and MPFC and lateral hypothalamic placements, which reported an average of 10% summation (Schenk and Shizgal, 1982).

Bielajew and Shizgal (1980) suggested that the low summation could be explained by either incomplete convergence of the behaviourally relevant signals, which can occur when the trajectories of some of the activated neurons diverge, or when convergence is indirect, that is, it occurs after the behaviourally relevant signals have been degraded through several synapses.

It appears that both mechanisms might explain the present results. There is ample anatomical and electrophysiological evidence that could imply the divergence of signals by evoking fine collaterals of fast corticostriatal fibers (Donoghue and Kitai, 1981; Wilson, 1986). These studies have confirmed the existence of corticofugal fibers that send ipsilateral and

contralateral collaterals to subcortical structures, such as the CPu, and also striatofugal fibers that send ipsilateral collaterals (DeFilipe and Farinas, 1992; Donoghue and Kitai, 1981; Feldman, 1984; Ferino et al., 1987; Fonnum, 1984; Fonnum et al., 1981). Recently, Malette and Miliaressis (1995) suggested that some of the collision-like profiles obtained in their study of contralateral concurrent stimulation of the lateral hypothalamus and ventral tegmental area might be explained by the activation of axon collaterals. Their functions resembled the typical collision profiles obtained for ipsilateral ventral tegmental and lateral hypothalamic areas (please see Figure 16, panels LH-LH and VTA-LH). Malette and Miliaressis (1995) suggested that each electrode stimulated a different collateral of the same neuron and that the antidromic potential elicited by each electrode failed to activate the collateral stimulated by the second electrode, and hence invoked a conduction failure at the branchpoint of the two collaterals. It is conceivable that the presence of a collateral in the effective stimulation field might cause a re-direction of the signal to two different integrators (Gallistel et al., 1981; Leon and Gallistel, 1992), and hence explain the small collision effects obtained in the present experiment. There is already some evidence that more than one integrator combines the rewarding effects of action potentials generated in different reward-relevant axons within the MFB (Leon and Gallistel, 1992), thus implicating more than one system of reinforcement in that region (Anderson and

Miliaressis, 1994). It is quite likely that similar mechanisms might exist outside the MFB. Alternatively, the integrator which "sees" the action potentials generated by the MPFC and CPu electrodes could be leaky, suggesting that the post-synaptic signal is subjected to decay before it reaches the last stage in the reward circuity (Gallistel, 1975). Once again, this decay process might result in low summation values.

In conclusion, the results of the present study suggest that at least some of the reward-relevant neurons coursing between the MPFC and CPu are axonally connected. The conduction velocity estimates of these neurons tend to be longer than the ones typically obtained for the MFB. There is a wealth of anatomical, electrophysiological and pharmacological data that suggest that the fibers stimulated are dopaminergic and glutamatergic ones. If that is the case then it is possible that the size of the collision effect may in fact be quite large, but is masked by a combination of excitatory and inhibitory effects.

GENERAL DISCUSSION

The experiments described in this thesis have shed some light on the characteristics and the organization of forebrain reward circuitry. The findings of high required currents, low success rate for self-stimulation, and uneven distribution of charge values suggests the involvement of less excitable, widely distributed and smaller, reward-relevant fibers than has been profiled for the MFB. Furthermore, the similarity in their recovery from refractoriness, and finally the results obtained from the collision experiment support the idea that a subset of MPFC and CPu reward fibers are functionally connected. In summary, the properties of the structures are suggestive of an intricate neural circuit.

A model of such a circuit is presented in Figure 17. It is composed of a main corticostriatal projection from the MPFC to the CPu, and a secondary, collateral fiber, that projects to an unidentified neuron. The latter in turn influences the original corticostriatal fiber, possibly through a positive feedback mechanism similar to long-term potentiation, and in that way completes the loop. One of the neurotransmitters which controls the activity in this circuit is glutamate. It has been shown that electrical stimulation of the MPFC results in an increase in aspartate and glutamate in the striatum and the hippocampus, and that this release appears to be related to current intensity (Godukhin, Zharikova, and Novoselov, 1980; Perschak & Cuénod, 1990). The currents employed here to evoke MPFC and CPu self-

Figure 17. This model of MPFC and CPu reward circuity is based in part on the work of Alexander and Crutcher (1990), Carlsson and Carlsson (1990), Dichiara and Morelli (1993), Dichiara, Morelli, and Consolo (1994), Graybiel (1990), and Parent and Hazrati (1995a, 1995b). The involvement of other potential neurotransmitters is purposely ignored for clarification purposes (Graybiel, 1990; Kolb and Tees, 1990). The hatched circles indicate stimulation fields; the question mark on the cell body refers to the uncertainty about identity of the specific origin of that afferent. The arrows point to the direction of propagation of reward signals.

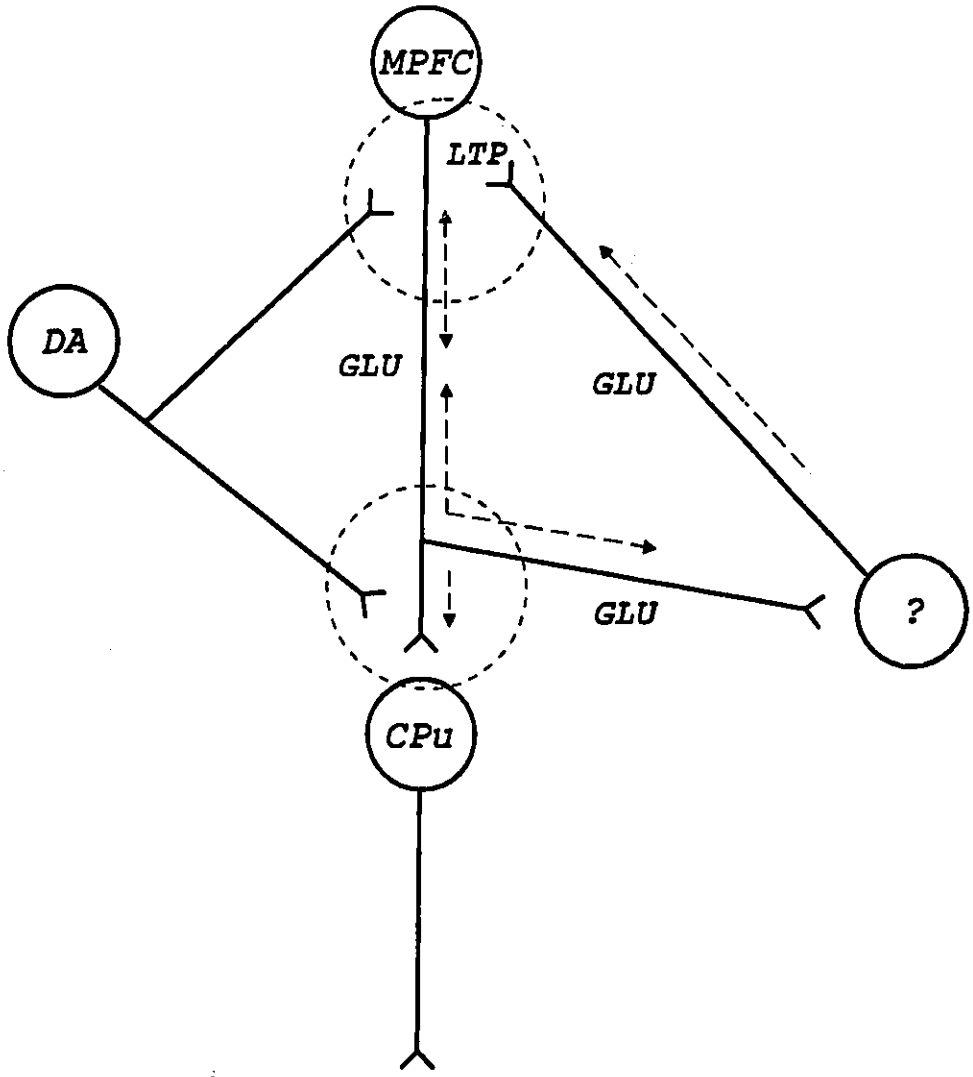
CPu: caudate-putamen

DA: dopamine

GLU: glutamate

LTP: long term potentiation

MPFC: medial prefrontal cortex


MPFC
LTP
DA
GLU
GLU
GLU
?
CPu

stimulation ranged from 200 to 1200 μA, and thus were within the margin sufficient to induce a release of glutamate.

Another neurotransmitter which makes direct contact with the initial segment and the terminal of the corticostriatal fiber is dopamine. The limbic regions of both the MPFC and CPu are thought to contain high concentrations of dopaminergic fibers (Berger et al., 1991; Lindvall and Björklund 1974; Sesack and Pickel, 1992; Ungerstedt, 1971) which respond best to frequencies below 40 Hz and currents between 50 to 100 μA, when a pulse duration of 0.5 seconds is used (Taber et al., 1993). The middle range of frequencies as well as the currents employed here were probably adequate to activate such neurons.

According to the model, if dopaminergic terminals are contained within the effective stimulation field, then they could directly contribute to the overall estimates of recovery from refractoriness. The distribution of the estimates of excitability cycle can be modified by stimulating competing or inhibitory axons with different refractory periods (Yeomans, 1990a). The estimates of refractoriness obtained from stimulation of the MPFC showed a greater overall range than that observed at the CPu site. This observation fits with the idea that the former is located near somas or initial segments. It is well known that refractory periods of the other neural elements tend to be longer than that of the axon.

The model also proposes a significant interaction between the ascending dopaminergic and descending glutamatergic fibers.

proposed that dopamine blocks, or modulates this corticostriatal excitatory input by acting on NMDA and AMPA glutamate receptors (Cheramy, Romo, Godeheu, Baruch, and Glowinski, 1986; Decse, Godeheu, Galli, Artaud, Cheramy, and Glowinski, 1992; Kalivas, Duffy, and Barrow, 1989; Krebs, Trovero, Desban, Gauchy, Glowinski, and Kemel, 1991; Levi and Raiteri, 1993; Moghaddam, Gruen, Roth, Bunney, and Adams, 1990). The ventral tegmental area constitutes the most significant dopaminergic projection to the MPFC and CPu. Its interactive effects with glutamatergic fibers might explain why transference was not observed from this region to the MPFC. If glutamate release is somewhat hindered, then facilitation of MPFC self-stimulation acquisition may be consequently inhibited (Garcia-Munoz et al., 1991a; Garcia-Munoz, Young, and Groves, 1991b). This potential interaction between dopamine and glutamate may explain why 6-OHDA lesions do not cause long-lasting changes in MPFC and CPu self-stimulation (Corbett et al., 1982; Ferrer et al., 1981; Gerfen and Clavier, 1979; Phillips and Fibiger, 1979; Phillips et al., 1976; Simon et al., 1979; Tiwari et al., 1996). The glutamatergic fibers, if indeed involved in reward, could overcompensate for the loss of dopaminergic innervation, and even potentially become overactive without the dopaminergic influence (Garcia-Munoz et al., 1991b).

The estimates of excitability cycles obtained in this study are consistent with the anatomical assignments proposed here in that the refractory period values estimated for the CPu and MPFC match those obtained electrophysiologically for dopaminergic and

glutamatergic fibers (Feger, Robeldo, and Renwart, 1991; Smith, Franke, Rosenheimer, Zufali, and Hatt, 1991; Wilson, 1986). The latency of the corticostriatal excitatory post-synaptic potentials measured intracellularly, ranges from 3.0 to 10.0 ms (Wilson, 1986). Interestingly, the thalamo-subthalamic projection in rats, which constitutes part of the basal ganglia circuity and uses glutamate as its neurotransmitter, appears to have a refractory period in the 5 to 7 ms range (Feger et al., 1991).

Likewise, the conduction velocity estimates reported here for the two regions are in the range expected for glutamatergic (Ferino et al., 1987) and dopaminergic fibers (German et al., 1980; Guyenet and Aghajanian, 1978; Yim and Mogenson, 1980), and span from 0.6 to 0.8 m/s and 0.3 to 1.5 m/s respectively.

Despite the anatomical and electrophysiological evidence implicating dopamine in forebrain reward circuitry, it has been proposed that this neurotransmitter may not be responsible for the reward signal generated by the neurons directly underneath the electrode tip, but may rather serve to modulate that signal at a later stage in the reward circuitry (Gallistel, 1986; Miliaressis et al., 1986; Wise and Rompré, 1989). This argument developed from the finding that at least at the level of the MFB, the frequency thresholds, refractory period estimates, and conduction velocities were incompatible with those which characterize the directly stimulated reward fibers (Gallistel et al., 1981; Wise, 1978, 1980). The psychophysical estimates

obtained in this thesis support the hypothesis that dopamine may indeed be directly involved in MPFC and CPu self-stimulation. Recent studies are also with agreement with this notion in that at least some of the dopaminergic neurons are thought be activated transynaptically by the parameters typically employed in brain stimulation reward studies (Gratton et al., 1988; Yeomans, et al., 1988), at least at the level of the MFB.

Although not much research has focused on the possible influences of glutamate on the brain's reward substrates, the results of the transference experiment hint at such an involvement. Self-stimulation evoked from both the MPFC and CPu is almost in all cases accompanied by seizures, and stimulating the MPFC site alone for many consecutive sessions tends to gradually increase the rate of self-stimulation. These observations are consistent with previous studies interested in the acquisition of MPFC self-stimulation (Corbett and Stellar, 1983; Corbett et al., 1985; Schenk, 1982). This gradual acquisition was proposed to result from the MPFC electrode strengthening or modifying neural connections between the MPFC site and some other reward structure (Corbett et al., 1982b), such as the CPu, in a manner similar to long-term potentiation. Long-term potentiation is defined as a stable and long-lasting increase in the magnitude of a postsynaptic response that can be produced as a result of a behavioral learning experience. Although this phenomenon has been mostly studied in the hippocampus, it has also been observed in the neocortex (Teyler,

1989; Wilson and Racine, 1983). This idea is strengthened by the fact that the facilitatory effects of non-contingent stimulation on the acquisition of MPFC self-stimulation can be blocked by ketamine, an antagonist of the glutamatergic NMDA receptor (Cobo et al., 1989; Corbett, 1990; Corbett et al., 1982, 1985; Robertson et al., 1982a, 1986), and by diazepam, an anticonvulsant (Robertson et al., 1982b). Thus, it is possible that the stimulation becomes more rewarding with repetition via the process of synaptic enhancement through a positive feedback mechanism, such as the one involved in long-term potentiation and kindling (Cepeda, Peacock, Levine, and Buchwald, 1991; Goddard, McIntyre, and Leech, 1969; Schwartz, Foster, French, Whetsell, and Kohler, 1984).

The presence of the corticostriatal collateral in the model may be responsible for the low summation values obtained in the collision tests. It is possible that the "split signals", one propagating orthodromically to its terminal in the CPu, and the other to an unknown neuron (see Figure 17), sum in separate integrators. This type of anatomical arrangement could also explain the shape of the collision profiles. Yeomans and Buckenham (1992) conducted a study aimed at evaluating the direct axonal connectivity between two sites that supported turning. The slow symmetric collision effects observed in their study, characterized by long collision intervals and an overall small collision effect, are similar to the ones seen here and might result from activation of the fine collaterals of corticostriatal

fibers.

In conclusion, this set of experiments comprises the first detailed study of this nature of the relationship between two forebrain reward regions. Not only was it demonstrated that the areas underlying reward form a close relationship, but additionally that their characteristics differ from those that have been documented for the MFB. Just as a map of a city is needed before one can comfortably navigate and fathom the neighbourhoods, so it is necessary to chart and describe the function of different brain areas before one can fully appreciate their complex interconnections.

REFERENCES

Abercrombie, E. D., Keefe, K. A., DiFrischia, D. S., and Zigmond, M. J. (1989). Differential effect of stress on in vivo dopamine release in striatum, nucleus accumbens, and medial frontal cortex. ***Journal of Neurochemistry, 52,*** 1655-1658.

Alexander, G. E., and Crutcher, M. D. (1990). Functional architecture of basal ganglia circuits: neural substrates pf parallel processing. ***Trends in Neurosciences, 13,*** 266-271.

Alexander, G. E., DeLong, M. R., and Strick, P. L. (1986). Parallel organization of functionally segregated circuits linking basal ganglia and cortex. ***Annual Review in Neuroscience, 9,*** 357-381.

Anderson R., and Miliaressis, E. (1994). Does the MFB convey functionally different reward signals? ***Behavioural Brain Research, 60,*** 55-61.

Arbuthnott, G. W., Crow, T. J., Fuxe, K., Olson, L., and Ungerstedt, U. (1970). Depletion of catecholamines in vivo induced by electrical stimulation of central monoamine pathways. ***Brain Research, 24,*** 471-483.

Bakshi, V. P., and Kelley, A. E. (1993). Striatal regulation of morphine-induced hyperphagia: an antomical mapping study. ***Psychopharmacology., 111,*** 207-214.

Balaban, P. M., and Chase, R. (1989). Self-stimulation in snails. ***Neuroscience Research Communications, 4,*** 139-146.

Balleine, B. W. (1991). The acquisition of self-stimulation of the medial prefrontal cortex following exposure to escapable and inescapable footshock. ***Behavioural Brain Research, 43,*** 167-174.

Balleine, B. W., McGregor, I. S., and Atrens, D. A. (1989). Controlability of prestimulation of the medial prefrontal cortex determines the facilitation of self-stimulation and kindled seizures. ***Physiology and Behaviour, 46,*** 239-245.

Barcikowski, R. S., and Robey, R. R. (1984). Decisions in single group repeated measures analysis: Statistical tests and three computer packages. ***The American Statistician, 38,*** 148-150.

Beckstead, R. M. (1979). An autoradiographic examination of corticocortical and subthalamic projections of the mediodorsal projection (prefrontal) cortex in the rat. ***Comparative Neurology, 184,*** 46-62.

Beckstead, R. M., Domesick, V. B., and Nauta, W. J. H. (1979). Efferent connections of the substantia nigra and ventral tegmental area in the rat. ***Brain Research, 175,*** 191-217.

Beninger, R. J., D'Amico, C. M., and Ranaldi, R. (1993). Microinjections of flupenthixol into the caudate putamen of rats produce intrasession declines in food-rewarded operant responding. ***Phramacology Biochemistry and Behavour, 45,*** 343-350.

Beninger, R. J., Laferrière, A., and Milner, P. M. (1978). An investigation of responding on schedules of electrical brain stimulation reinforcement. ***Canadian Journal of Psychology, 32,*** 106-115.

Berger, B., Gaspar, P., and Verney, C. (1991). Dopaminergic innervation of the cerebral cortex: unexpected differences between rodents and primates. ***Trends in Neurosciences, 14,*** 21-27.

Bielajew, C. H. (1983). ***Psychophysical inference of the direction of normal conduction in the substrates for medial forebrain bundle self-stimulation.*** Unpublished doctoral dissertation, Concordia University, Montreal.

Bielajew, C. H. (1991). Distribution of cytochrome oxidase in response to rewarding brain stimulation: effects of different pulse durations. ***Brain Research Bulletin, 26,*** 379-384.

Bielajew, C., and Fouriezos, G., (1985). Post-stimulation excitability of mediodorsal thalamic self-stimulation. ***Behavioural Brain Research, 17,*** 97-101.

Bielajew, C., Jordan., C., Ferme-Enright, J., and Shizgal, P., (1981). Refractory periods and anatomical linkage of the substrates for lateral hypothalamic and periaqueductal gray self-stimulation. ***Physiology and Behaviour, 27,*** 95-104.

Bielajew, C., Jurgens, S., and Fouriezos, G., (1987a). The effects of pulse duration on refractory periods of neurons mediating brain-stimulation reward. ***Behavioural Brain Research, 24,*** 233-241.

Bielajew, C., Lapointe, M,. Kiss, I., and Shizgal, P., (1982). Absolute and relative refractory periods of the substrates for lateral and ventral midbrain self-stimulation. ***Physiology and Behaviour, 28,*** 125-132.

Bielajew, C., and Shizgal, P. (1980). Dissociation of the substrates for medial forebrain bundle self-stimulation and stimulation-escape using a two-electrode stimulation technique. ***Physiology and Behaviour, 25,*** 707-711.

Bielajew, C., and Shizgal, P., (1982). Behaviorally derived measures of conduction velocity in the substrates for rewarding medial forebrain bundle stimulation. ***Brain Research, 237,*** 107-119.

Bielajew, C., and Shizgal. (1986). Evidence implicating descending fibers in self-stimulation of the medial forebrain bundle. ***Journal of Neuroscience, 6,*** 919-929.

Bielajew, C., Thrasher, A., and Fouriezos, G. (1987b). Self-stimulation sites in the lateral hypothalamic and lateral preoptic areas are functionally connected. ***Canadian of Psychology Abstracts, 28,*** 36.

Bielajew, C., and Trzcińska, M (1994). Characteristics of stimulation-induced feeding sites in the sulcal prefrontal cortex. ***Behavioural Brain Research, 61,*** 29-35.

Bishop, M. P., Elder, S. T., and Heath, R. G. (1963). Intracranial self-stimulation in man. ***Science, 140,*** 394-396.

Boucher-Thrasher, A. (1986). ***Evidence for anatomical connectivity between lateral hypothalamic and lateral preoptic area brain stimulation sites.*** Unpublished master's thesis, University of Ottawa, Ottawa.

Bower, G. H., and Miller, N. E. (1958). Rewarding and punishing effects from stimulating the same place in the rat's brain. ***Journal of Comparative and Physiological Psychology, 51,*** 669-674.

Boyd, E. S., and Gardner, L. C. (1962). Positive and negative reinforcement from intracranial stimulation of a teleost. ***Science, 136,*** 648-649.

Brodie, D. A., Moreno, O. M., Malis, J. L., and Boren, J. J. (1960). Rewarding properties of intracranial stimulation. ***Science, 131,*** 929-930.

Brown, C. M. (1992). ***Parametric analysis of post-stimulation neuronal excitability in the lateral hypothalamus.*** Unpublished honours thesis, University of Ottawa, Ottawa.

Bruner, A. (1966). Facilitation of classical conditioning in rabbits by reinforcing brain stimulation. ***Psychonomic Sciences, 6,*** 211-212.

Brutkowski, S. (1965). Functions of prefrontal cortex in animals. ***Physiological Review, 45,*** 721-746.

Burchfiel, J. L., and Applegate, C. D. (1989). Stepwise progression of kinding: Perspectives from the kindling antagonism model. ***Neuroscience and Biobehavioural Reviews, 13,*** 289-299.

Burchfiel, J. L., Serpa, K. A., and Duffy, F. H. (1982). Further studies of antagonism of seizure development between concurrently developing kindled limbic foci in the rat. ***Experimental Neurology, 75,*** 476-489.

Burnham, W. (1975). Primary and "transfer" seizure development in the kindled rat. ***Canadian Journal of Neurological Sciences, 2,*** 417-428.

Bushnik-Harris, T. L. (1993). ***Brain stimulation reward in the lateral preoptic area: An examination of its substrate and functional connectivity to the ventral tegmental area.*** Unpublished doctoral dissertation, University of Ottawa, Ottawa.

Bushnik-Harris, T., and Bielajew, C. (1993). An evaluation of the functional connectivity between the lateral preoptic and ventral tegmental area reward sites. ***Society for Neuroscience Abstracts, 19,*** 809.

Cagguila, A. R., and Hoebel, B. G. (1966). "Copulation-reward site" in the posterior hypothalamus. ***Science, 153,*** 1284-1285.

Carlsson, M., and Carlsson, A. (1990). Interactions between glutamatergic and monoaminergic systems within the basal ganglia - implications for schizophrenia and Parkinson's disease. ***Trends in Neurosciences, 13,*** 272-276.

Carr, G. D., and White, N. M. (1983). Conditioned place preference from intra-accumbens but not intracaudate amphetamine injections. ***Life Sciences, 33,*** 2551-2557.

Carr, G. D., and White, N. M. (1986). Anatomical dissociation of amphetamine's rewarding and aversive effects: an intracranial microinjection study. ***Psychopharmacology (Berlin), 89,*** 340-346.

Carter, C. J. (1982). Topographical distribution of possible glutamatergic pathways from the frontal cortex to the striatum and substantia nigra in rats. ***Neuropharmacology, 21,*** 379-383.

Cavalheiro, E. A., and Turski, L. (1986). Intrastriatal N-methyl-D-aspartate prevents amygdala kindled seizures in rats. ***Brain Research, 377,*** 173-176.

Cepeda, C., Peacock, W., Levine, M. S., and Buchwald, N. A. (1991). Iontophoretic application of NMDA produces different types of excitatory responses in developing human cortical and caudate neurons. ***Neuroscience Letters, 126,*** 167-171.

Cheramy, A., Romo, R., Godeheu, G., Baruch, P. and Glowinski, J. (1986). In vivo presynaptic control of dopamine release in the cat caudate-nucleus - II. Facilitatory or inhibitory influence of L-glutamate. ***Neuroscience, 19,*** 1081-1090.

Christopher, S. M., and Butter, C. M. (1968). Consumatory behaviors and locomotor exploration evoked from self-stimulation sites in rats. ***Journal of Comparative and Physiological Psychology, 66,*** 335-339.

Clavier, R. M., and Gerfen, C. R. (1979). Self-stimulation of the sulcal prefrontal cortex in the rat: direct evidence for ascending dopaminergic mediation. ***Neurosciences Letters, 12,*** 183-187.

Clavier, R. M., and Gerfen, C. R. (1982). Intracranial self-stimulation in the thalamus of the rat. ***Brain Research Bulletin, 8,*** 353-358.

Cobo, M., Ferrer, J. M. R., and Mora, F. (1989). The role of the lateral cortico-cortical prefrontal pathway in self-stimulation of the medial prefrontal cortex in the rat. ***Behavioural Brain Research, 31,*** 257-265.

Conover, K. L., and Shizgal, P. (1994). Competition and summation between rewarding effects of sucrose and lateral hypothalamic stimulation in the rat. ***Behavioural Neuroscience, 108,*** 537-548.

Conover, K. L., Woodside, B., and Shizgal, P. (1994). Effects of sodium depletion on competition and summation between rewarding effects of salt and lateral hypothalamic stimulation in the rat. ***Behavioural Neuroscience, 108,*** 549-558.

Conti, F., Rustioni, A., Petrusz, P., and Towle, A. C. (1987). Glutamate-positive neurons in the somatic sensory cortex of rats and monkeys. ***Journal of Neuroscience, 7,*** 1887-1901.

Cooper, R., and Taylor, L. (1967). Thalamic reticular system and central gray: self-stimulation. ***Science, 156,*** 102-103.

Corbett, D. (1990). Ketamine blocks the plasticity associated with prefrontal cortex self-stimulation. ***Pharmacology Biochemistry and Behaviour, 37,*** 685-688.

Corbett, D. (1991). Cocaine enhances the reward value of medial prefrontal cortex self-stimulation. ***Neuroreport, 2,*** 805-808.

Corbett, D. (1992). Chronic morphine fails to enhance the reward value of prefrontal cortex self-stimulation. ***Pharmacology Biochemistry and Behaviour, 42,*** 451-455.

Corbett, D., Fox, E., and Milner, P. M (1982a). Fiber pathways associated with cerebellar self-stimulation in the rat: a retrograde and anterograde tracing study. ***Behavioural Brain Research, 6,*** 167-184.

Corbett, D., Laferrière, A., and Milner, P. M. (1982b). Plasticity of the medial prefrontal cortex facilitated acquisition of intracranial self-stimulation by pretraining stimulation. ***Physiology and Behaviour, 28,*** 531-534.

Corbett, D., Laferrière, A., and Milner, P. M. (1982c). Elimination of medial prefrontal cortex self-stimulation following transection of efferents to the sulcal cortex in the rat. ***Physiology and Behaviour, 29,*** 425-431.

Corbett, D., Silva, L. R., and Stellar, J. R. (1985). An investigation of the factors affecting development of frontal cortex self-stimulation. ***Physiology and Behaviour, 34,*** 89-95.

Corbett, D., and Stellar, J. R. (1983). Neurological reactivity during medial prefrontal cortex stimulation: effects of self-stimulation experience. ***Physiology and Behaviour, 31,*** 771-776.

Davies, G. D. (1958). Caudate lesions and spontaneous locomotion in the monkey. ***Neurology, 8,*** 135-139.

Decse, J. M., Godeheu, G., Galli, T., Artaud, F., Cheramy, A., and Glowinski, J. (1992). L-Glutamate-evoked release of dopamine from synaptosomes of the rat striatum: involvement of AMPA and N-methyl-D-aspartate receptors. ***Neuroscience, 47,*** 333-339.

DeFelipe, J., and Farinas, I. (1992). The pyramidal neuron of the cerebral cortex: morphological and chemical characteristics of the synaptic inputs. ***Progress in Neurobiology, 39,*** 563-607.

Delgado, J. (1973). ***Physical control of the mind,*** New York: Harper & Row Publishers.

Deniau, J. M., Thierry, A. M., and Feger, J., (1980). Electrohpysiological identification of mesencephalic ventromedial tegmental (VMT) neurons projecting to the frontal cortex, septum, and nucleus accumbens. ***Brain Research, 189,*** 315-326.

Desban, M., Kemel, M. L., Glowinski, J., and Gauchy, C. (1993). Spatial organization of patch and matrix compartments in the rat striatum. ***Neuroscience, 57,*** 661-671.

Deutch, A. Y., Lee, M. C., and Iadarola, M. J. (1992). Regionally specific effects of atypical antipsychotic drugs on striatal fos expression: the nucleus accumbens shell as a locus of antipsychotic action. ***Mollecular and Cellular Neuroscience, 3,*** 332-341.

Deutsch, J. A. (1964). Behavioral measurement of the neural refractory period and its application to intracranial self-stimulation. ***Journal of Comparative and Physiological Psychology, 58,*** 1-9.

DiChiara, G., and Morelli, M. (1993). Dopamine-acetylcholine-glutamate interactions in the striatum. ***Advanced Neurology, 60,*** 102-106.

Dichiara, G., Morelli, M., and Consolo, S. (1994). Modulatory functions of neurotransmitters in the striatum: Ach/dopamine/NMDA interactions. ***Trends in Neurosciences, 17,*** 228-233.

Dinopoulus, A., Dori, I., Davies, S. W., and Parnavelas, J. G. (1989). Neurochemical heterogenity among corticofugal and callosal projections. ***Experimental Neurology, 105,*** 36-44.

Divac, I., Fonnum, F., and Storm-Mathisen, J. (1977). High affinity uptake of glutamate in terminals of corticostriatal axons. ***Nature (London), 266,*** 377-378.

Divac, I., Kosmal, A., Björklund, A., and Lindvall, O. (1978). Subcortical projections to the prefrontal cortex in the rat as revealed by the horseradish peroxidase technique. ***Neuroscience, 3,*** 785-796.

Divac, I., Mogensen, J., Marinkovic, S., and Martensson, R. (1987). On the projections from the neostriatum to the cerebral cortex: The "displaced" neurons. ***Neuroscience, 21,*** 197-205.

Donoghue, J. P., and Kitai, S. T. (1981). A collateral pathway to the neostriatum from corticofugal neurons of the rat sensory-motor cortex: An intracellular HRP study. ***Journal of Comparative Neurology, 201,*** 1-13.

Douglin, A., and Glassman, R. B. (1979). Gradual increase in self-stimulation response rates: effect of electrode loci. ***Physiological Psychology, 7,*** 135-138.

Druhan, J. P., Levy, M., and Shizgal, P. (1993). Effects of varying reinforcement schedule, reward current, and pretrial priming stimulation on discrete-trial performance for brain stimulation reward. ***Psychobiology, 21,*** 37-42.

Duchowny, M. S., and Burchfiel, J. L. (1981). Facilitation and antagonism of kindled seizure development in the limbic system of the rat. ***Electroencephalography and Clinical Neurophysiology, 51,*** 403-416.

Dunn, A. J. (1988). Stress-related activation of cerebral dopaminergic systems. ***Annals of the New York Academy of Sciences, 537,*** 188-205.

Dunn, A. J., and File, S. E. (1983). Cold restraint alters dopamine metabolism in frontal cortex, nucleus accumbens and neostriatum. ***Physiology and Behaviour, 31,*** 511-513.

Durivage, A., and Miliaressis, E., (1987). Anatomical dissociation of the substrates of medial forebrain self-stimulation and exploration. ***Behavioural Neuroscience, 101,*** 57-61.

Edmonds, D. E., and Gallistel, C. R. (1974). Parametric analysis of brain stimulation reward in the rat: III. Effects of performance variables on the reward summation function. ***Journal of Comparative Physiological Psychology, 87,*** 876-883.

Edmonds, D. E., Stellar, J. R., and Gallistel, C. R. (1974). Parametric analysis of brain stimulation reward in the rat: II. Temporal summation in the reward system. ***Journal of Comparative and Physiological Psychology, 87,*** 860-869.

Ettenberg, A., Laferrière, A., Milner, P. M., and White, N. (1981). Response involvement in brain-stimulation reward. ***Physiology and Behaviour, 27,*** 641-647.

Evans, K. R., and Eikelboom, R. (1987). Feeding induced by ventricular bromocriptine and amphetamine: a possible excitatory role for dopamine in eating behavior. ***Behavioural Neuroscience, 101,*** 591-593.

Everitt, B. J. (1990). Sexual motivation: A neural and behavioural analysis of the mechanisms underlying appetitive and copulatory respones of male rats. ***Neuroscience and Biobehavioural Reviews, 14,*** 217-232.

Feger, J., Robeldo, P., and Renwart, N. (1991). The subthalamic nucleus: new data, new questions. In G. Bernardi, M. B. Carpenter, G. D. Chiara., M. Morelli, and P. Stanzione (Eds.), ***The Basal Ganglia III,*** New York: Plenum Press.

Feldman, M. L. (1984). Morphology of the neocortical pyramidal neurons. In A. Peters and E. G. Jones (Eds.), ***Cerebral cortex, vol.1, Cellular components of the cerebral cortex,*** New York: Plenum Press.

Ferino, F., Thierry, A. M., Saffroy, M., and Glowinski, J (1987). Interhemispheric and subcortical collaterals of the medial prefrontal corrtical neurons in the rat. ***Brain Research, 417,*** 257-266.

Ferrer, J. M. R., Cobo, M., and Mora, F. (1987). The basolateral limbic circuit and self-stimulation of the medial prefrontal cortex in the rat. ***Physiology and Behaviour, 40,*** 291-295.

Ferrer, J. M. R., Cobo, M., and Mora, F. (1988). Peptides and self-stimulation of the medial prefrontral cortex in the rat: Effects of intracerebral microinjections of substance P and cholecystokinin. ***Peptides, 9,*** 1-7.

Ferrer, J. M. R., Myers, R. D., and Mora, F. (1985). Suppression of self-stimulation of the medial prefrontal cortex after local micro-injection of kainic acid in the rat. ***Brain Research Bulletin, 15,*** 225-228.

Ferrer, J. M. R., Sanguinetti, A. M., Vives, F., and Mora, F. (1983). Effects of agonists and antagonists of D_1 and D_2 dopamine receptors on self-stimulation of the medial prefrontal cortex in the rat. ***Pharmacology Biochemistry and. Behaviour., 19,*** 211-217.

Ferron, A., Thierry, A. M., Le Douarin, C., and Glowinski, J. (1984). Inhibitory influences of the mesocortical dopaminergic system on the spontaneous activity or excitatory response induced from the thalamic mediodorsal nucleus in the rat medial prefrontal cortex. ***Brain Research, 302,*** 257-265.

Fonnum, F. (1984). Glutamate: a neurotransmitter in mammalian brain. ***Journal of Neurochemistry, 42,*** 1-11.

Fonnum, F., Storm-Mathisen, J., and Divac, I. (1981). Biochemical evidence for glutamate as neurotransmitter in the corticostriatal and corticothalamic fibers in rat brain. ***Neuroscience, 6,*** 863-873.

Fouriezos, G., Bielajew, C., and Pagotto, W. (1990). Task difficulty increases thresholds of rewarding brain stimulation. ***Behavioural Brain Research, 37,*** 1-7.

Fouriezos, G., Walker, S., Rick, J., and Bielajew, C. (1987). Refractoriness of neurons mediating intracranial self-stimulation in the anterior basal forebrain. ***Behavioural Brain Research, 24,*** 73-80.

Fouriezos, G., and Wise, R. A. (1984). Current-distance relation for rewarding brain stimulation. ***Behavioural Brain Research, 14,*** 85-89.

Fournier, T., Bazana, G., Brown, C., and Fouriezos, G. (1991). Behavioural estimates of post-stimulation excitability of reward neurons at very long inter-pulse intervals. ***Poster presented at the first Annual Meeting of the Canadian Society for Brain, Behaviour, and Cognitive Sciences,*** Calgary, June, 1991.

Frank, R., and Williams, H. (1985). Both response effort and current intensity affect self-stimulation train duration thresholds. ***Physiology Biochemistry and Behaviour, 22,*** 527-530.

Frankland, P. W., Scott, B. W., and Yeomans, J. S. (1995). Axons and synapses mediating electrically evoked startle: collision tests and latency analysis. ***Brain Research, 670,*** 97-111.

Fuster, J. M. (1980). ***The prefrontal cortex: Anatomy, physiology, and neuropsychology of the frontal lobe.*** New York: Raven Press.

Gallistel, C. R. (1974). Note on temporal summation in the reward system. ***Journal of Comparative Physiological Psychology, 87,*** 870-875.

Gallistel, C. R. (1975). Motivation as central organizing process: The psychophysical approach to its functional and neurophysiological analysis. In J. K. Cole and T. B. Sonderegger (Eds.), ***Nebraska Symposium on Motivation,*** Lincoln: University of Nebraska Press.

Gallistel, C. R. (1978). Self-stimulation in the rat: Quantitative characteristics of the reward pathway. ***Journal of Comparative Physiological Psychology, 92,*** 977-998.

Gallistel, C. R. (1986). The role of the dopaminergic projections in MFB self-stimulation. ***Behavioural Brain Research, 20,*** 313-321.

Gallistel, C. R., Boytim, M., Gomita, Y., and Klebanoff, L. (1982). Does pimozide block the reinforcing effect of brain stimulation? ***Pharmacology Biochemistry and Behaviour, 17,*** 769-781.

Gallistel, C. R., and Freyd, G. (1987). Quantitative determination of the effects of catecholaminergic agonists and anatoganists on the rewarding efficacy of brain stimulation. ***Pharmacology Biochemistry and Behaviour, 26,*** 731-741.

Gallistel, C. R., Gomita, Y., Yadin, E., and Campbell, K. A. (1985). Forebrain origins and terminations of the medial forebrain bundle metabolically activated by rewarding stimulation or by reward-blocking doses of pimozide. ***Journal of Neuroscience, 5,*** 1246-1261.

Gallistel, C. R., and Leon, M. (1991). Measuring the subjective magnitude of brain stimulation reward by titration with rate of reward. ***Behavioural Neuroscience, 105,*** 913-925.

Gallistel, C. R., Shizgal, P., and Yeomans, J. S (1981). A portrait of the substrate for self-stimulation. ***Psychological Review, 88,*** 228-273.

Garcia-Munoz, M., Young, S. J., and Groves, P. M. (1991a). Terminal excitability of the corticostriatal pathway. I. Regulation by dopamine receptor stimulation. ***Brain Research, 551,*** 195-206.

Garcia-Munoz, M., Young, S. J., and Groves, P. M. (1991b). Terminal excitability of the corticostriatal pathway. II. Regulation by glutamate receptor stimulation. ***Brain Research, 551,*** 207-215.

Gerfen, C. R. (1984). The neostriatal mosaic: Compartmentalization of corticostriatal input and striatonigral output system. ***Nature, 311,*** 461-464.

Gerfen, C. R. (1989). The neostriatal mosaic: Striatal patch-matrix organization is related to cortical lamination. ***Science, 246,*** 385-388.

Gerfen, C. R. (1992). The neostriatal mosaic: Multiple levels of compartmental organization. ***Trends in Neurosciences, 15,*** 133-139.

Gerfen, C. R., and Clavier, R. M. (1981). Intracranial self-stimulation from the sulcal prefrontal cortex in the rat: The effects of 6-hydroxydopamine or kaininc acid lesions at the site of stimulation. ***Brain Research, 224,*** 291-304.

German, D., Dalsass, M., and Kiser, R. (1980). Electrophysiological examination of the ventral tegmental (A10) area in the rat. ***Brain Research, 181,*** 191-197.

Goddard, G. V., McIntyre, D. C., and Leech, C. K. (1969). A permanent change in brain function as resulting from daily electrical stimulation. ***Experimental Neurology, 25,*** 295-330.

Godukhin, O. V., Zharikova, A. D., and Novoselov, V. I. (1980). The release of labelled L-glutamic acid from rat neostriatum in vivo following stimulation of frontal cortex. ***Neuroscience, 5,*** 2151-2154.

Goeders, N. E., Dworkin, S.I., and Smith, J.E (1986). Neuropharmacological assessment of cocaine self-administration into the medial prefrontal cortex. ***Pharmacology Biochemistry and Behaviour, 24,*** 1429-1440.

Goeders, N. E., and Smith, J. E. (1983). Reinforcing properties of cocaine in the medial prefrontal cortex: Primary action on presynaptic dopaminergic terminals. ***Pharmacology Biochemistry and Behaviour, 25,*** 191-199.

Goodman, I. J., and Brown, J. L. (1966). Stimulation of positively and negatively reinforcing sites in the avian brain. ***Life Sciences, 5,*** 693-704.

Gratton, A., Hoffer, B. J., and Gerhardt, G. A. (1988). Effects of electrical stimulation of brain reward sites on release of dopamine in rat: an in vivo electrochemical study. ***Brain Research Bulletin, 21,*** 319-324.

Gratton, A., and Wise, R. A. (1985). Hypothalamic reward mechanism: two first-stage fiber populations with a cholinergic component. ***Science, 277,*** 545-548.

Gratton, A., and Wise, R. A. (1988a). Comparisons of refractory periods for medial forebrain bundle fibers subserving stimulation-induced feeding and brain stimulation reward: a psychophysiological study. ***Brain Research, 438,*** 256-263.

Gratton, A., and Wise, R. A. (1988b). Comparisons of connectivity and conduction velocities for medial forebrain bundle fibers subserving stimulation-induced feeding and brain stimulation reward. ***Brain Research, 438,*** 264-270.

Graybiel, A. M. (1984). Neurochemically specified subsystems in the basal ganglia. Functions of the basal ganglia. ***CIBA Foundation symposium,*** London: Pitman.

Graybiel, A. M. (1990). Neurotransmitters and neuromodulators in the basal ganglia. ***Trends in Neurosciences, 13,*** 244-254.

Guyenet, P. G., and Aghajanian, G. K. (1978). Antidromic identification of dopaminergic and other output neurons in the rat substantia nibra. ***Brain Research, 150,*** 69-84.

Hamilton, A., Stellar, J., and Hart, E. (1985). Reward, performance, and the response strength method in self-stimulating rats: validation and neuroleptics. ***Physiology and Behaviour, 35,*** 897-904.

Handford, A., and Ackermann, R. F. (1993). Electrogenic status epilepticus induced from numerous limbic sites. ***Epilepsy Research, 15,*** 21-26.

Harris, T., and Bielajew, C. (1988). An examination of current/frequency trade-offs for rewarding brain stimulation along the medial forebrain bundle. ***Milner Symposium on Neural Basis of Reward and Reinforcement.*** L'Esterel.

Harris, T., and Bielajew, (1990). Suppression of seizures without altering excitability properties of reward-related neurons by the novel benzodiazepine - brotizolam. ***Society for Neuroscience Abstracts, 16,*** 593.

Harris, T., and Bielajew, C. (1991). Diazepam alters brain-stimulation reward thresholds in seizure-prone sites. ***Behavioural Brain Research, 46,*** 167-173.

Harris, T., Bielajew, C., and Merali, Z. (1991). The effect of peripherally administered bombesin on LH stimulation-induced feeding and self-stimulation. ***Society for Neuroscience Abstracts, 17,*** 1237.

Hawkins, R. D., Puerto, A., Roll, P. L., and Yeomans, J. S. (1983). Refractory periods of neurons mediating stimulation-elicited eating and brain stimulation reward: Interval scale measurement and tests of a model of neural integration. ***Behavioural Neuroscience, 97,*** 416-432.

Heath, R. (1972). Pleasure and brain activity in man. ***Journal of Nervous and Mental Disease, 154,*** 3-18.

Heimer, L. (1978). The olfactory cortex and the ventral striatum. In K. E. Livingston & O. Hornykiewicz (Eds.), ***Limbic mechanisms,*** New York: Plenum Press.

Heimer, L., Alheid, G. F., and Zaborszky, L. (1985). Basal ganglia. In G. Paxinos (Eds.), ***The rat nervous system. vol 1. Forebrain and midbrain.*** New York,: Academic Press.

Herberg, L. J., and Blundell, J. E. (1969). Epileptiform seizures and the rats choice of stimulus duration in electrical self-stimulation. ***Journal of Comparative and Physiological Psychology, 67,*** 442-447.

Herberg, L. J., and Rose, I. C. (1994). Kindled epileptic seizures, postictal refractoriness, status epilepticus, and electrical self-stimulation. ***Neuroscience and Biobehavioural Reviews, 18,*** 411-420.

Herkenham, M. (1987). Mismatches between neurotransmitter and receptor localizations in brain: observations and implications. ***Neuroscience, 23,*** 1-38.

Herkenham, M. and Pert, C. B. (1981). Mosaic distribution of opiate receptors, parafascicular projections and acetylcholinesterase in rat striatum. ***Nature, 291,*** 415-418.

Herman, J. F., Guillonneau, D., Dantzer, R., Scatton, B., Semerdjian-Rouquier, L., and LeMoal, M. (1982). Differential effects of inescapable footshocks on dopamine turnover in cortical and limbic areas of the rat. ***Life Sciences, 30,*** 2207-2214.

Hernandez, L., and Hoebel, B. G. (1990). Feeding can enhance dopamine turnover in the prefrontal cortex. ***Brain Research Bulletin, 25,*** 975-979.

Heyman, G. M., Kinzie, D. L., and Seiden, L. S. (1986). Chlorpromazine and pimozide alter reinforcing efficacy and motor performance. ***Psychopharmacology, 88,*** 346-353.

Hoebel, B. G., and Teitelbaum, P. (1962). Hypothalamic control of feeding and self-stimulation. ***Science, 135,*** 375-377.

Howarth, C. I., and Deutsch, J. A. (1962). Drive decay: The cause of fast "extinction" of habits learned for brain stimulation. ***Science, 137,*** 35-36.

Hursch, J. B. (1939). Conduction velocity and diameter of nerve fibers. ***American Journal of Physiology, 127,*** 131-139.

Huston, J. P., and Borbely, A. A. (1973). Operant conditioning in forebrain ablated rats by use of rewarding hypothalamic stimulation. ***Brain Research, 50,*** 467-472.

Huston, J. P., and Borbely, A. A. (1974). The thalamic rat: General behavior operant learning with rewarding hypothalamic stimulation and effects of amphetamine. ***Physiology and Behaviour, 12,*** 433-448.

Iversen, S. D., and Dunnett, S. B. (1990). Functional organization of striatum as studied with neural grafts. ***Neuropsychologia, 28,*** 601-626.

Jayaraman, A. (1980). Anatomical evidence for cortical projections from the striatum in the cat. ***Brain Research, 195,*** 29-36.

Joseph, M. H., and Hodges, H. (1990). Lever pressing for food reward and changes in dopamine turnover and uric acid in rat caudate and nucleus accumbens studies chronicaly by in vivo voltammetry. ***Journal of Neuroscience Methods, 34,*** 143-149.

Justesen, D. R., Sharp, J. C., and Porter, R. B. (1963). Self-stimulation of the caudate nucleus by instrumentally naive cats. ***Journal of Comparative and Physiological Psychology, 56,*** 371-374.

Kalivas, P. W., Duffy, P., and Barrow, J. (1989). Regulation of the mesocortical dopamine system by glutamic acid receptor subtypes. ***Journal of Pharmacological and Experimental Therapeutics, 251,*** 378-387.

Kandel, E.R., and Schwartz, J. H. (1985). ***Principles of neural science,*** New York: Elsevier.

Kane, F., Coulombe, D., and Miliaressis, E. (1991). Amygdaloid self-stimulation. A moveable electrode mapping study. ***Behavioural Neuroscience, 105,*** 926-932.

Kitai, S. T., Kocsis, J. D., Preston, R. J., and Sugimori, M. (1976). Monosynaptic inputs to caudate neurons identified by intracellular injection of horseradish peroxidase. ***Brain Research, 109,*** 601-606.

Kocsis, J. D., Sugimori, M., and Kiatai, S. T. (1977). Convergence of excitatory synaptic inputs to caudate spiny neurons. ***Brain Research, 124,*** 403-413.

Kolb, B. (1984). Functions of the frontal cortex of the rat: A comparative review. ***Brain Research Review, 8,*** 65-98.

Kolb, B. (1990). Prefrontal cortex. In B. Kolb & C. Tees (Eds.), ***The cerebral cortex of the rat,*** London: The MIT Press.

Kolb, B., and Tees, (1990). ***Prefrontal cortex,*** London: MIT Press.

Krebs, M. O., Trovero, F., Desban, M., Gauchy, C., Glowinski, J., and Kemel, M. L. (1991). Distinct presynaptic regulation of dopamine release through NMDA receptors in striosome- and matrix-enriched areas of the rat striatum. ***Journal of Neuroscience, 11,*** 1256-1262.

Krettek, J. E., and Price, J. L (1977). The cortical projections of the mediodorsal nucleus and adjacent thalamic nuclei in the rat. ***Journal of Comparative Neurology, 171,*** 157-192.

Lenzer, I. I. (1971). Differences between behavior reinforced by electrical stimulation of the brain and conventionally reinforced behavior: An associative analysis. ***Psychological Bulletin, 78,*** 103-118.

Leon, M., and Gallistel, C. R. (1992). The function relating the subjective magnitude of brain stimulation reward to stimulation strength varies with site of stimulation. ***Behavioural Brain Research, 52,*** 183-193.

Leonard, C. M. (1969). The prefrontal cortex of the rat. I. Cortical projection of the mediodorsal nucleus. II. Efferent connections. ***Brain Research, 12,*** 321-343.

Levi, G., and Raiteri, M. (1993). Carrier-mediated release of neurotransmitters. ***Trends in Neurosciences, 16,*** 415-419.

Lindvall, O., and Björklund, A., (1974). The organization of ascending catecholamine neuron systems in the rat brain as revealed by the glyoxylic acid fluorescence technique. ***Acta Physiologica Scandinavica, 412,*** 1-48.

Lindvall, O., and Björklund, A., (1983). Dopamine- and norepinephrine-containing neuron systems: Their anatomy in the rat brain. In P. C. Emson (Eds.), ***Chemical Neuroanatomy,*** New York: Raven Press.

Lindvall, O., Björklund, A., and Divac, I. (1978). Organization of catecholamine neurons projecting to the frontal cortex in the rat. ***Brain Research, 142,*** 1-24.

Lineberry, C. G., and Vierck, C. J. (1975). Attenuation of pain reactivity by caudate nucleus stimulation in monkeys. ***Brain Research, 98,*** 110-134.

Lucas, K. (1913). The effect of alcohol on the excitation, conduction, and recovery process in nerve. ***Journal of Physiology, 46,*** 470-505.

Lucas, K. (1917). ***The conduction of the nervous impulse.*** Aberdeen: University Press.

MacMillan, C. J., Simantirakis, P., and Shizgal, P. (1985). Self-stimulation of the lateral hypothalamus and ventrolateral tegmentum: Excitability characteristics of the directly stimulated substrates. ***Physiology and Behaviour, 35,*** 711-723.

Malette, J., and Miliaressis, E. (1990). The notion of response invariance in trade-off studies of self-stimulation. ***Behavioural Brain Research, 40,*** 37-44.

Malette, J., and Miliaressis, E. (1995). Interhemispheric links in brain stimulation reward. Behav. ***Behavioural Brain Research*** (in press).

Matthews, G. (1978). Strength-duration properties of single units driven by electrical stimulation of the lateral hypothalamus in rats. ***Brain Research Bulletin, 3,*** 171-174.

McGeorge, A. J., and Faull, R. L. M (1987). The organization and collateralization of corticostriate neurons in the motor and sensory cortex of the rat brain. ***Brain Research, 423,*** 318-324.

McGeorge, A. J., and Faull, R. L. M (1989). The organization of the projection from the cerebral cortex to the striatum in the rat. ***Neuroscience, 29,*** 503-507.

McGreer, P. L., McGreer, E. G., Scherer, U., and Singh, K. (1977). A glutamatergic corticostriatal path? ***Brain Research, 128,*** 369-373.

McGregor, I. S. (1992). Determinants of slow acquisition of medial and sulcal prefrontal cortex self-stimulation: An individual differences approach. ***Physiology and Behaviour, 51,*** 1219-1225.

McGregor, J. S., and Atrens, D. M. (1991). Prefrontal cortex self-stimulation and energy balance. ***Neuroscience, 105,*** 870-883.

McGregor, I. S., Balleine, B. W., and Atrens, D. M (1989). Footshock stress facilitates self-stimulation of the medial prefrontal cortex but not the lateral hypothalamus in the rat. ***Brain Research, 490,*** 48-58.

Miliaressis, E. (1981a). A miniature, moveable electrode for brain stimulation in small animals. ***Brain Research Bulletin, 7,*** 715-718.

Miliaressis, E. (1981b). Refractoriness of neurons subserving circling following stimulation of the median raphe region in the rat. ***Physiology and Behaviour, 26,*** 709-713.

Miliaressis, E., Bouchard, A., and Jacobowitz, D. M. (1975). Strong positive reward in median raphe: Specific inhibition by para-chlorophenylalanine. ***Brain Research, 98,*** 194-201.

Miliaressis, E., Emond, C., and Merali, Z. (1991). Re-evaluation of the role of dopamine in intracranial self-stimulation using in vivo microdialysis. ***Behavioural Brain Research, 46,*** 43-48.

Miliaressis, E., and Malette, J. (1987). Summation and saturation properties in the rewarding effect of brain stimulation. ***Physiology and Behaviour, 41,*** 595-604.

Miliaressis, E., Malette, J., and Coulombe, D. (1986a). The effects of pimozide on the reinforcing efficacy of central grey stimulation in the rat. ***Behavioural Brain Research, 21,*** 95-100.

Miliaressis, E., and Philippe, L. (1983). A dual moveable electrode and its application to the behavioral version of the collision test. ***Brain Research Bulletin, 10,*** 573-577.

Miliaressis, E., and Philippe, L. (1984). The pontine substrate of circling behavior. ***Brain Research, 293,*** 143-152.

Miliaressis, E., and Rompré, P-P. (1980). Self-stimulation and circling: Differentiation of the neural substrata by behavioral measurement with the use of a double pulse technique. ***Physiology and Behaviour, 25,*** 939-943.

Miliaressis, E., and Rompré, P-P. (1987). Effects of concominant motor reactions on the measurement of rewarding efficacy of brain stimulation. ***Behavioural Neuroscience, 101,*** 827-831.

Miliaressis, E., Rompré, P-P., and Durivage, A., (1982). Psychophysical method for mapping behavioral substrates using a moveable electrode. ***Brain Research Bulletin, 8,*** 693-701.

Miliaressis, E., Rompré, P-P., Laviolette, P., Phillippe, L., and Coulombe, D. (1986b). The curve-shift paradigm in self-stimulation. ***Physiology and Behaviour, 37,*** 85-91.

Milner, P. (1989). The discovery of self-stimulation and other stories. ***Neuroscience and Biobehavioural Review, 13,*** 61-67.

Milner, P. (1991). Brain-stimulation reward: A review. ***Canadian Journal of Psychology, 45,*** 1-36.

Milner, P., and Laferrière, A. (1982). Strength-duration characteristics of lateral hypothalamic and periaqueductal gray reward-path neurons. ***Physiology and Behaviour, 29,*** 857-864.

Miserendino, M. J. D., and Coons, E. E. (1989). Electrical stimulation of the medial prefrontal cortex supports both "pure reward" and "reward-escape" behavior in rats. ***Brain Research, 483,*** 226-232.

Mitchell, J. B., and Gratton, A. (1992). Partial dopamine depletion of the prefrontal cortex leads to enhanced mesolimbic dopamine release elicited by repeated exposure to naturally reinforcing stimuli. ***Journal of Neuroscience, 12,*** 3609-3618.

Mogenson, G. J., and Cioe, J. (1977). Central reinforcement - a bridge between brain function and behavior. In W. K. Honig and Staddon, J. E. R (Eds.), ***Handbook of operant behavior.*** New Jersey: Prentice-Hall.

Mogenson, G. J., Jones, D., and Yim, C. Y. (1980). From motivation to action: Functional interface between the limbic system and the motor system. ***Progress in Neurobiology, 14,*** 69-97.

Mogenson, G. J., and Stevenson, J. A. F. (1966). Drinking and self-stimulation with electrical stimulation of lateral hypothalamus. ***Physiology and Behaviour, 1,*** 251-254.

Moghaddam, B., Gruen, R. J., Roth, R. H., Bunney, B. S., and Adams, R. N. (1990). Effect of L-glutamate on the release of striatal dopamine: in vivo dialysis and electrochemical studies. ***Brain Research, 518,*** 55-60.

Moore, R. Y., and Bloom, E. E., (1978). Central catecholamine neuron system: Anatomy and physiology of the dopamine system. ***Annual Review of Neuroscience, 1,*** 129-169.

Mora, F. (1978). The neurochemical substrates of prefrontal cortex self-stimulation: A review and an interpretation of some recent data. ***Life Sciences, 22,*** 919-930.

Mora, F., and Ferrer, J. M. R. (1986). Neurotransmitters, pathways and circuits as the neural susbtrates of self-stimulation of the prefrontal cortex: facts and speculations. ***Behavioural Brain Research, 22,*** 127-140.

Mora, F., and Myers, R. D. (1977). Brain self-stimulation: direct evidence for the involvement of dopamine in the prefrontal cortex. ***Science, 197,*** 1387-1389.

Mundl, W. (1980). A constant-current stimulator. ***Physiology and Behaviour, 24,*** 991-993.

Murray, B., and Shizgal, P. (1994). Evidence implicating both slow- and fast-conducting fibers in the rewarding effect of medial forebrain bundle stimulation. ***Behavioural Brain Research, 63,*** 47-60.

Nassif, S., Cardo, B., Libersat, F., and Velley, L. (1985). Comparison of deficits in electrical self-stimulation after ibotenic acid lesion of the lateral hypothalamus and the medial prefrontal cortex. ***Brain Research, 332,*** 247-257.

Nauta, W. J. H., and Domesick, V. B (1984). Afferent and efferent relationships of the basal ganglia. Functions of the basal ganglia, ***CIBA Foundation Symposium,*** London: Pitman.

Neter, J., Wasserman, W., and Kutner, M. (1990). ***Applied linear statistical models,*** Homewood, IL: Irwin

Newman, R. and Winans, S. S. (1980). An experimental study of the ventral striatum of the golden hamster. II. Neuronal connections of the olfactory tubercle. ***Journal of Comparative Neurology, 191,*** 193-212.

Nicolaysen, L. C., Ikeda, M., Justice, Jr., J. B., and Neill, D. B. (1988). Dopamine release at behaviorally relevant parameters of nigrostriatal stimulation: effects of current and frequency. ***Brain Research, 460,*** 50-59.

Nieuwenhuys, R., Geeraedts, L. M. G., and Veening, J. G. (1982). The nedial forebrain bundle of the rat. I. General introduction. ***Journal of Comparative Neurology, 206,*** 49-81.

Olds, J. (1958). Self-stimulation of the brain: Its use to study local effects of hunger, sex, and drugs. ***Science, 127,*** 315-324.

Olds, J., and Milner, P. (1954). Positive reinforcement produced by electrical stimulation of septal area and other regions of rat brain. ***Journal of Comparative Physiological Psychology, 47,*** 419-427.

Olds, M. E., and Olds, J. (1963). Approach-avoidance analysis of rat diencephalon. ***Journal of Comparative Neurology, 120,*** 256-295.

Ott, T., Destrade, C., and Ruthrich, H. (1980). Introduction of self-stimulation derived from a brain region lacking in dopaminergic innervation. ***Behavioural Neural Biology, 28,*** 512-516.

Panagis, G., Miliaressis, E., Anagnostakis, Y., and Spyraki, C. (1995a). Ventral pallidum self-stimulation: A moveable electrode mapping study. ***Behavioural Brain Research*** (in press).

Panagis, G., Spyraki, C., and Miliaressis, E. (1995b). Post-stimulation excitability of ventral pallidum self-stimulation neurons. ***Behavioural Neuroscience*** (in press).

Papp, M., and Bal, A. (1987). Separation of the motivational and motor consequences of 6-hydroxydopamine lesions of the mesolimbic and nigrostriatal systems in rats. ***Behavioural Brain Research, 23,*** 221-229.

Parent, A. (1990). Extrinsic connections of the basal ganglia. ***Trends in Neurosciences, 13,*** 254-258.

Parent, A., and Hazrati, L-N. (1995a). Functional anatomy of the basal ganglia. I. The cortico-basal ganglia-thalamo-cortical loop. ***Brain Research Reviews, 20,*** 91-127.

Parent, A., and Hazrati, L-N. (1995b). Functional anatomy of the basal ganglia. II. The place of subthalamic nucleus and external pallidum in basal ganglia circuity. ***Brain Research Reviews, 20,*** 128-154.

Paxinos, G., and Watson, C (1986). ***The rat brain in stereotaxic coordinates.*** Australia: Academic Press.

Perschak, H., and Cuénod, M. (1990). *In vivo* release of endogenous glutamate and aspartate in the rat striatum during stimulation of the cortex. ***Neuroscience, 35,*** 283-287.

Pfaus, J. G., Kleopoulos, S. P., Mobbs, C. V., Gibbs, R. B., and Pfaff, D. W. (1993). Sexual stimulation activates c-fos within estrogen-concentrating regions of the female rat forebrain. ***Brain Research, 624,*** 253-267.

Phillips, A. G. (1970). Enhancement and inhibition of olfactory bulb self-stimulation by odours. ***Physiology and Behaviour, 5,*** 1127-1131.

Phillips, A. G. (1984). Brain reward circuity: A case for separate systems. ***Brain Research Bulletin, 12,*** 195-201.

Phillips, A. G., Carter, D. A., and Fibiger, H. C., (1976). Dopaminergic substrates of intracranial self-stimulation in the caudate nucleus. ***Brain Research, 104,*** 221-232.

Phillips, A. G., and Fibiger, H. C. (1978). The role of dopamine in maintaining intracranial self-stimulation in the ventral tegmentum, nucleus accumbens, and medial prefrontal cortex. ***Canadian Journal of Psychology, 32,*** 58-66.

Phillips, A. G., and Fibiger, H. C. (1989). Neuroanatomical bases of intracranial self-stimulation: untangling the Gordian knot. In J. M. Liebman and Cooper, S. J. (Eds.), ***The neuropharmacological basis of reward.*** Oxford: Clarendon Press.

Porrino, L. J., Coons, E. E., and MacGregor, B. (1983). Two types of medial hypothalamic inhibition of lateral hypothalamic reward. ***Brain Research, 277,*** 269-282.

Prado-Alcala, R. A., Kent, E. W., and Reid. L. D., (1975). Intracranial self-stimulation effects along the route of nigrostriatal bundle. ***Brain Research, 84,*** 531-540.

Prado-Alcala, R. A., Streather, A., and Wise, R. A (1984). Brain stimulation reward and dopamine terminal fields. II. Septal and cortical projections. ***Brain Research, 301,*** 209-219.

Prado-Alcala, R. A., and Wise, R. A. (1984). Brain stimulation reward and dopamine terminal fields. I. Caudate-putamen, nucleus accumbens and amygdala. ***Brain Research, 297,*** 265-273.

Purpura, D. P., Housepian, E. M., and Grundfest, H. (1958). Analysis of caudate-cortical connections in neuroaxially intact and telencephale isole cats. ***Archives of Italian Biology, 96,*** 145-167.

Ranck, J. B. Jr., (1975). Which elements are excited in electrical stimulation of mammalian central nervous system: A review. ***Brain Research, 98,*** 417-440.

Reid. L. D., Gibson, W. E., Gledhill, S. M., and Porter, P. (1964). Anticonvulsant drugs and self-stimulating behaviour. ***Journal of Comparative Physiological Psychology, 57,*** 353-356.

Reymann, K., Mulcko, S., Ott, T., and Matthies, H. (1986). Opioid-receptor blockage reduces nose-poke self-stimulation derived from medial entorhinal cortex. ***Pharmacology Biochemistry and Behaviour, 24,*** 439-443.

Rick, J. Th., and Fouriezos, G. (1992). Pulse jitter retards kindling without affecting brain-stimulation reward. ***Brain***

Robbins, T. W., Cador, M., Taylor, J. R., and Everitt, B. J. (1989). Limbic-striatal interactions in reward-related processes. ***Neuroscience and Biobehavioural Reviews, 13,*** 155-162.

Robertson, A. (1989). Multiple reward systems and the prefrontal cortex. ***Neuroscience and Biobehavioural Reviews, 13,*** 163-170.

Robertson, A., Laferrière, A., and Franklin, K. B. J. (1981). Amphetamine and increases in current intensity modulate reward in the hypothalamus and substantia nigra but not in the prefrontal cortex. ***Physiology and Behaviour, 28,*** 869-872.

Robertson, A., Laferrière, A., and Milner, P. M., (1982a). Development of brain stimulation reward in the medial prefrontal cortex: Facilitation by prior electrical stimulation of the sulcal prefrontal cortex. ***Physiology and Behaviour, 28,*** 869-872.

Robertson, A., Laferrière, A., and Milner, P. M (1982b). Treatment with anticonvulsant drugs retards the development of brain-stimulation reward in the prefrontal cortex. ***Physiology and Behaviour, 29,*** 275-280.

Robertson, A., Laferrière, A., and Milner, P. M. (1986). The role of cortico-cortical projections in self-stimulation of the prelimbic and sulcal prefrontal cortex in rats. ***Behavioural Brain Research., 21,*** 129-142.

Robertson, A., and Mogenson, G. J. (1978). Evidence for a role of dopamine in self-stimulation of the nucleus accumbens of the

Rolls, E. T. (1974). The neural basis of brain-stimulation reward. ***Progress in Neurobiology, 3,*** 71-160.

Rolls, E. T., and Kelly, P. H. (1972). Neural basis of stimulus-bound locomotor activity in the rat. ***Journal of Comparative and Physiological Psychology, 81,*** 173-182.

Rompré, P-P., and Boye, S. (1989). Localization of reward-relevant neurons in the pontine tegmentum: a moveable electrode mapping study. ***Brain Research, 496,*** 295-302.

Rompré, P-P., and Miliaressis, E., (1980). A comparison of the excitability cycles of the hypothalamic fibers involved in self-stimulation and exploration. ***Physiology and Behaviour, 24,*** 995-998.

Rompré, P-P., and Miliaressis, E., (1985). Pontine and mesencephalic substrates of self-stimulation. ***Brain Research, 359,*** 246-259.

Rompré, P-P., and Shizgal, P. (1986). Electrophysiological characteristics of neurons in forebrain regions implicated in self-stimulation of the medial forebrain bundle in the rat. ***Brain Research, 364,*** 338-349.

Rose, M. (1974). Pain-reducing properties of rewarding brain stimulation in the rat. ***Journal of Comparative and Physiological Psychology, 87,*** 607-617.

Rossi III, J., and Panksepp, J. (1992). Analysis of the relationship between self-stimulation sniffing and brain-stimulation sniffing. ***Physiology and Behaviour, 51,*** 805-813.

Routtenberg, A. (1971). Forebrain pathways of reward in Rattus Norvegicus. ***Journal of Comparative and Physiological Psychology, 75,*** 269-276.

Routtenberg, A. (1976). ***Self-stimulation pathways: origins and terminations - a three stage technique.*** Amsterdam: Elsevier.

Routtenberg, A., and Lindy, J. (1965). Effects of the availability of rewarding septal and hypothalamic stimulation on bar pressing for food under conditions of deprivation. ***Journal of Comparative and Physiological Psychology, 60,*** 158-161.

Routtenberg, A., and Sloan, M., (1972). Self-stimulation of the frontal cortex of Rattus Norvegicus. ***Brain Biology, 7,*** 567-572.

Sax, L., and Gallistel, C. R. (1991). Characteristics of spatiotemporal integration in the priming and rewarding effects of medial forebrain bundle stimulation. ***Behavioural Neuroscience, 105,*** 884-900.

Schenk, S. (1982). ***The substrate for prefrontal cortical self-stimulation: A psychophysical investigation.*** Unpublished doctoral dissertation, Concordia University, Montreal.

Schenk, S., Prince, C., and Shizgal, P., (1985). Spatio-temporal integration in the substrates for self-stimulation of the prefrontal cortex. ***Physiology and Behaviour, 35,*** 303-306.

Schenk, S., and Shizgal, P., (1982). The substrates for lateral hypothalamic and medial prefrontal cortex self-stimulation have different refractory period and show poor spatial summation. ***Physiology and Behaviour, 28,*** 133-138.

Schenk, S., and Snow, S. (1994). Sensitization of cocaine's motor activating properties produced by electrical kindling of the medial prefrontal cortex but not of the hippocampus. ***Brain Research, 659,*** 17-22.

Schultz, W., Apicella, P., Scarnati, E., and Ljungberg, T. (1992). Neuronal activity in monkey ventral striatum related to the expectation of reward. ***Journal of Neuroscience, 12,*** 4595-4610.

Schwartz, R., Foster, A. C., French, E. D., Whetsell, W. O., and Kohler, C., (1984). Excitotoxic models for neurodegenerative disorders. ***Life Sciences, 35,*** 19-32.

Sesack, S. R. and Bunney., B. S. (1989). Pharmacological characterization of the receptor mediating electrophysiological responses to dopamine in the rat medial prefrontal cortex: a microiontophoretic study. ***Journal of Pharmacological and Experimental Therapeutics, 248,*** 1323-1333.

Sesack, S. R., Deutch, A. Y., Roth, R. H., and Bunney, B. S. (1989). Topographical organization of the efferent projections of the medial prefrontal cortex in the rat: An anterograde tract-tracing study with Phaseolus vulgaris leucoagglutinin. ***Journal of Comparative Neurology, 290,*** 213-242.

Sesack, S. R., and Pickel, V. M. (1992). Prefrontal cortical efferents in the rat synapse on unlabeled neuronal targets of catecholamine terminals in the nucleus accumbens septi and on dopamine neurons in the ventral tegmental area. ***Journal of Comparative Neurology, 320,*** 145-160.

Shizgal, P. (1989). Toward a cellular analysis of intracranial self-stimulation: Contributions of collision studies. ***Neuroscience and Biobehavioural Reviews, 13,*** 81-90.

Shizgal, P., Bielajew, C., Corbett, D., Skelton, R., and Yeomans, J., (1980). Behavioral methods for inferring anatomical linkage between rewarding brain stimulation sites. ***Journal of Comparative and Physiological Psychology, 94,*** 227-237.

Shizgal, P., and Murray, B. (1989). Neuronal basis of intracranial self-stimulation. In J. M. Liebman and S. J. Cooper (Eds.), ***The neuropharmacological basis of reward,*** Oxford: Clarendon Press.

Shizgal, P., Schindler, D., and Rompré, P-P. (1989). Forebrain neurons driven by rewarding stimulation of the medial forebrain bundle in the rat: comparison of psychophysical and electrophysiological estimates of refractory periods. ***Brain Research, 499,*** 234-248.

Silva, L. R., Vogel, J. A., and Corbett, D. (1982). Frontal cortex self-stimulation: evidence for independent substrates within areas 32 and 24. ***Society for Neuroscience Abstracts, 8,*** 625.

Simon, H., Stinus, L., Tassin, J. P., Lavielle, S., Blanc, G., Thierry, A., Glowinsky, J., and Le Moal, M. (1979). Is the dopaminergic mesocortical system necessary for intracranial self-stimulation? ***Behavioural and Neural Biology, 27,*** 125-145.

Skelton, R. W., and Shizgal, P. (1980). Parametric analysis of ON- and OFF- responding for hypothalamic stimulation. ***Physiology and Behaviour, 25,*** 699-706.

Smith, D. O., Franke, C., Rosenheimer, J. L., Zufali, F., and Hatt, H. (1991). Desensitization and resensitization rates of glutamate-activated channels may regulate motoneuron excitability. ***Journal of Neurophysiology, 66,*** 1166-1175.

Spence, S. J., Silverman, J. A., and Corbett, D., (1985). Cortical and ventral tegmental systems exert opposing influences on self-stimulation from the prefrontal cortex. ***Behavioural Brain Research, 17,*** 117-124.

Spiller, A.E., and Racine, R. J. (1994). Transfer kindling between sites in the entorhinal cortex-perforant path-dentate gyrus system. ***Brain Research, 635,*** 130-138.

Staines, W. A., Nagy, J. I., Vincent, S. R., and Fibiger, H. C. (1980). Neurotransmitters contained in the efferents of the striatum. ***Brain Research, 194,*** 391-402.

Stamm, J. S. (1964). Retardation and facilitation in learning by stimulation of frontal cortex in monkey. In J. M. Warren & K. Akert (Eds.), ***The frontal granular cortex and behaviour,*** New York: McGraw-Hill.

Stark, P., and Boyd, E. S. (1961). Electrical self-stimulation by dogs through chronically implanted electrodes in the hypothalamus. ***Federal Proceedings, 20,*** 328.

Steiner, S. S., Beer, B., and Shaffer, M. M. (1969). Escape from self-produced rates of brain stimulation. ***Science, 163,*** 90-91.

Stellar, J. R., Brooks, F., and Mills, L. (1979). Approach and withdrawal analysis of the effects of hypothalamic stimulation and lesions in rats. ***Journal of Comparative and Physiological Psychology, 93,*** 446-466.

Stellar, J. R., and Corbett, D. (1989). Regional neuroleptic microinjections indicate a role for nucleus accumbens in lateral hypothalamic self-stimulation reward. ***Brain Research, 477,*** 126-143.

Stellar, J. R., and Stellar, E (1985). ***The neurobiology of motivation and reward.*** New York: Springer-Verlag.

Sutherland, R. J., and Nakajima, S. (1981). Self-stimulation of the habenular complex in the rat. ***Journal of Comparative and Physiological Psychology, 95,*** 781-791.

Swadlow, H. A., and Waxman, S. E. (1978). ***Activity-dependent variations in the conduction properties of central axons.*** New York:: Raven Press.

Szabo, I., Lenard, L., and Kosaras, B., (1974). Drive decay theory of self-stimulation: refractory periods and axon diameters in hypothalamic reward loci. ***Physiology and Behaviour, 12,*** 329-343.

Szabo, I., and Milner, P. (1973). Intracranial self-stimulation in the rat: Cable properties of the depolarized membrane. ***Acta Physiologica Academiae Scientarium Hungaricae, 43,*** 65-70.

Taber, M., and Fibiger, H. C. (1993). Electrical stimulation of the medial prefrontal cortex increases dopamine release in the striatum. ***Neuropsychopharmacology, 9,*** 271-275.

Tehovnik, E. J., and Yeomans, J. S. (1986). Two converging brainstem pathways mediating circling behavior. ***Brain Research, 385,*** 329-342.

Tehovnik, E. J., and Yeomans, J. S. (1988). Contraversive circling elicited from the internal capsule and substantia nigra: evidence for a continuous axon bundle mediating circling. ***Brain Res., 441,*** 269-280.

Teyler, T. J. (1989). Comparative aspects of hippocampal and neocortical long-term potentiation. ***Journal of Neuroscie Methods, 28,*** 101-108.

Trzcińska, M., and Bielajew, C. (1992). Behaviourally derived estimates of excitability in striatal and medial prefrontal cortical self-stimulation sites. ***Behavioural Brain Research, 48,*** 1-8.

Tsang, W-K., and Stutz, R. M. (1984). Subject control as a determinant of the reinforcing properties of intracranial stimulation. ***Physiology and Behaviour, 32,*** 795-802.

Ungerstedt, U. (1971). Stereotaxic mapping of monoamine pathways in the rat brain. ***Acta Physiologica Scandinavice, 82 (Suppl.367),*** 1-48.

Ursin, R., Ursin, H., and Olds, J. (1966). Self-stimulation of hippocampus in rats. ***Journal of Comparative and Physiological Psychology, 61,*** 353-359.

Vachon, M. P. and Miliaressis, E., (1992). Dorsal diencephalic self-stimulation: a moveable electrode mapping study. ***Behavioural Neuroscience, 106,*** 981-991.

Vachon, M. P. and Miliaressis, E., (1994). Post-stimulation excitability of diencephalic self-stimulation neurons. ***Behavioural Brain Research, 60,*** 177-182.

Valenstein, E. S. (1964). Problems of measurement and interpretation with reinforcing brain stimulation. ***Psychological Review, 71,*** 415-436.

Valenstein, E. S. (1973). ***Brain control: A critical examination of brain stimulation and psychosurgery,*** New York: J. Wiley & Sons.

Valenstein, E. S., Cox, V. G., and Kakolewski, J. W. (1968). Modification of motivated behavior elicited by electrical stimulation of the hypothalamus. ***Science, 159,*** 1119-1121.

Veening, J. G., Cornelissen, F. M., and Lieven, P. A. J. M. (1980). The topical organization of the afferents to the caudatoputamen of the rat. A Horseradish peroxidase study. ***Neuroscience, 5,*** 1253-1268.

Vives, F., Gayoso, M. J., Osorio, C., and Mora, F. (1983). Afferent pathways to points of self-stimulation in the medial prefrontal cortex of the rat, as revealed by the horseradish peroxidase technique. ***Behavioural Brain Research, 8,*** 23-32.

Vives, F., and Mora, F. (1986). Effects of agonists and antagonists of cholinergic receptors on self-stimulation of the medial pre-frontal cortex in the rat. ***General Pharmacology, 17,*** 63-67.

Vives, F., Morales, A. B., and Mora, F. (1986). Lesions of connections of the medial prefrontal cortex in rats: differential effects of self-stimulation and spontaneous motor activity. ***Physiology and Behaviour, 36,*** 47-52.

Voorn, P., Jorritsma-Byham, B., Van Dijk, C., and Buijs, R. M. (1986). The dopaminergic innervation of the ventral striatum in the rat: A light- and electron-microscopical study with antibodies against dopamine. ***Journal of Comparative Neurology, 251,*** 84-99.

Wang, R. Y. (1981). Dopaminergic neurons in the rat ventral tegmental area. I. Identification and characterization. ***Brain Research Reviews, 3,*** 123-140.

Wang, T., and French, E. D. (1993). L-Glutamate excitation of A10 dopamine neurons is preferentially mediated by activation of NMDA receptors: extra- and intracellular electrophysiological studies in brain slices. ***Brain Research, 627,*** 299-306.

Waraczynski, M. and. Kaplan, J. M. (1990). Frequency-response characteristics provide a functional separation between stimulation-bound feeding and self-stimulation. ***Physiology and Behaviour, 47,*** 843-851.

Waxman, S. G., and Bennett, M. V. L., (1972). Relative conduction velocities of small myelinated fibers in the central nervous system. ***Nature, 238,*** 217-219.

Weinreich, D., and Clark, L. D. (1970). Anticonvulsant drugs and self-stimulation rates in rats. ***Archives of International Pharmacodynamics, 185,*** 269-273.

White, N. M. (1989). A functional hypothesis concerning the striatal matrix and patches: mediation of S-R memory and reward. ***Life Sciences, 45,*** 1943-1957.

Wilcott, R. C. (1979). Cortical stimulation and suppression of activity in the rat. ***Physiology and Behaviour, 23,*** 1041-1048.

Wilcott, R. C., Sabol, B. A., and Yurcheshen, R. P., (1976). Frontal cortex and response suppression in the rat. ***Brain Behaviour and Evolution, 13,*** 116-124.

Wilson, C. J. (1986). Postsynaptic potentials evoked in spiny neostriatal projection neurons by stimulation of ipsilateral and contralateral neocortex. ***Brain Research, 367,*** 201-213.

Wilson, C. J. (1987). Morphology and synaptic connections of crossed corticostriatal neurons in the rat. ***Journal of Comparative Neurology, 263,*** 567-580.

Wilson, D. A. and Racine, R. (1983). The postnatal development of post-activation potentiation in the rat neocortex. ***Developmental Brain Research, 7,*** 271-276.

Wise, R. A. (1978). Catecholamine theories of reward: A critical review. ***Brain Research, 152,*** 215-247.

Wise, R. A. (1980). Action of drugs of abuse on brain reward systems. ***Pharmacology Biochemistry and Behaviour, 13 (suppl. 1),*** 213-223.

Wise, R. A., and Hoffman, D. C. (1992). Localization of drug reward mechanisms by intracranial injections. ***Synapse, 10,*** 247-263.

Wise, R. A., and Rompré, P-P. (1989). Brain dopamine and reward. ***Annual Review of Psychology, 40,*** 191-225.

Yadin, E., Guarini, V., and Gallistel, C. R. (1983). Unilaterally activated systems in rats self-stimulating at sites in the medial forebrain bundle, medial prefrontal cortex, or locus coeruleus. ***Brain Research, 266,*** 39-50.

Yeomans, J. S. (1975). Quantitative measurement of neuronal post-stimulation excitability with behavioral methods. ***Physiology and Behaviour, 15,*** 593-602.

Yeomans, J. S. (1979). The absolute refractory periods of self-stimulation neurons. ***Physiology and Behaviour, 22,*** 911-919.

Yeomans, J. S. (1988). Mechanisms of brain-stimulation reward. ***Progress in Psychobiology and Physiological Psychology, 13,*** 227-266.

Yeomans, J. S. (1989). Two substrates for medial forebrain bundle self-stimulation: Myelinated axons and dopamine axons. ***Neuroscience and Biobehavioural Reviews, 13,*** 91-98.

Yeomans, J. S. (1990a). ***Principles of brain stimulation.*** New York: Oxford University Press.

Yeomans, J. S. (1990b). Temporal summation decay in hypothalamic self-stimulation: threshold changes at long intrapair intervals due to axonal subnormal periods. ***Behavioural Neuroscience, 104,*** 991-999.

Yeomans, J. S. (1995). Electrically evoked behvaiours: axons and synapses mapped with collision tests. ***Behavioural Brain Research, 67,*** 121-132.

Yeomans, J. S. and Buckenham, K. (1992). Electrically evoked turning: asymmetric and symmetric collision between anteriomedial cortex and striatum. ***Brain Research, 510,*** 279-292.

Yeomans, J. S., and Cochrane, K. A. (1993). Collision-like interactions between acoustic and electrical signals that produce startle reflexes in reticular formation sites. ***Brain Research, 617,*** 320-328.

Yeomans, J. S., and Davis, J. K. (1975). Behavioral measurement of post-stimulation excitability of neurons mediating self-stimulation by varying the voltage of paired pulses. ***Behavioural Biology, 15,*** 435-447.

Yeomans, J. S., Hempel, C. M. E., and Chapman, C. A. (1993a). Axons and synapses mediating startle-like responses evoked by electrical stimulation of the reticular formation in rats: symmetric and assymetric collision effects. ***Brain Research, 617,*** 309-319.

Yeomans, J. S., Maidment, N. T., and Bunney, B. S. (1988). Excitability properties of medial forebrain bundle axons of A9 and A10 dopamine cells. ***Brain Research, 450,*** 86-93.

Yeomans, J. S., Mathur, A., and Tampakeras, M. (1993b). Rewarding brain stimulation: Role of tegmental cholinergic neurons that activate dopamine neurons. ***Behavioural Neuroscience, 107,*** 1077-1087.

Yeomans, J. S., Matthews, G. G., Hawkins, R. D., Bellman, K., and Doppelt, H. (1979). Characteristics of self-stimulation neurons by their local potential summation properties. ***Physiology and Behaviour, 22,*** 921-929.

Yeomans, J. S., Mercouris, N., and Ellard, C., (1985). Behaviorally measured refractory periods are lengthened by reducing tip exposure or raising current. ***Behavioural Neuroscience, 99,*** 913-928.

Yeomans, J. S., Pearce, R., Wen, D., and Hawkins, R. D. (1984). Mapping midbrain sites for circling using current-frequency trade-off data. ***Physiology and Behaviour, 32,*** 287-294.

Yeomans, J. S., and Tehovnik, E. J. (1988). Turning responses evoked by stimulation of visuomotor pathways. ***Brain Research, 472,*** 235-259.

Yim, C., and Mogenson, G. (1980). Electrophysiological studies of neurons in the ventral tegmental area of Tsai. ***Brain Research, 181,*** 301-313.

Zacharko, R. M., Bowers, W. J., Kokkinidis, L., and Anisman, H. (1983). Region-specific reductions of intracranial self-stimulation after uncontrollable stress: Possible effects on reward processes. ***Behavioural Brain Research, 9,*** 129-141.

Zacharko, R. M., Kasian, M., Irwin, J., Zalcman, S., Lalonde, G., MacNeil, G., and Anisman, H. (1990). Behavioural characterization of intracranial self-stimulation from mesolimbic, mesocortical, nigrostriatal, hypothalamic and extra-hypothalamic sites in the non-inbred CD-1 mouse strain. ***Behavioural Brain Research, 36,*** 251-281.

Zeng, D., and Stuesse, S. L. (1991). Morphological heterogenity within the cingulate cortex in rat: a horseradish peroxidase transport study. ***Brain Research, 565,*** 290-300.

Zilles, K., and Wree, A (1985). Cortex: Areal and laminar structure. In G. Paxinos (Eds.), ***The rat nervous system. vol.1. Forebrain and midbrain,*** New York: Academic Press.