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REVIEW

REWARD CONTINGENT POSITIVE VARIATION (RCPV) AND POST-REINFORCEMENT EEG SYNCHRONIZATION (PRS) IN THE CAT: PHYSIOLOGICAL ASPECTS, THE EFFECT OF MORPHINE AND LSD-25, AND A NEW INTERPRETATION OF CHOLINERGIC MECHANISMS

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A. PHYSIOLOGIC ASPECTS OF REWARD-INDUCED PHENOMENA

(1) *The role of gustatory input*

In cats trained to press a lever for a small amount of milk reward, the electrocorticogram (ECoG) over the primary and secondary visual projections, including part of the suprasylvian association cortex, shows a dramatic shift from desynchronized to highly synchronized waves of 6–9 Hz during consumption of the reward (Clemente *et al.*, 1964; Buchwald *et al.*, 1964; Roth *et al.*, 1967; Marczynski *et al.*, 1968). This phenomenon, known as postreinforcement synchronization (PRS), was shown to depend on the palatability and appropriateness of the gustatory reward (Sterman & Wyrwicka, 1967). This is demonstrated by comparing one reward with another offered alternatively. For instance, as illustrated in Fig. 1, when water is presented at the onset of an experimental session to a food and water deprived cat, fully developed PRS is observed, and subsequent introduction of milk either does not change or slightly augments the PRS. However, if water is reintroduced later, the PRS is totally suppressed, although the lever pressing and consummatory behaviour show no change (Marczynski *et al.*, 1968, 1971a). The PRS is associated with an epicortical steady potential shift termed the reward contingent positive variation (RCPV) which usually outlasts the PRS by 2–3 sec (Rick & Marczynski, in preparation). The maximum amplitude of the RCPV is recorded over the primary and secondary visual projections with reference to subjacent white matter, bone, or remote cortical areas (Marczynski *et al.*, 1971a). In infants during bottle feeding or other pleasurable emotional stimuli, 5.5–6.0 Hz rhythmic ECoG waves are observed which have been termed hedonic hypersynchrony (Maulsby, 1971) and may be analogous to PRS since, as in the cat, they are restricted to the occipital region.

(2) *Reward-induced inhibition of the ascending reticular activating system (ARAS)*

It can be argued along several lines of evidence that the PRS–RCPV phenomenon results from a very transient but powerful suppression of the ARAS. First of all, the patterns and topographic distribution of the PRS and RCPV are indistinguishable from ECoG synchronization observed in a satiated drowsy animal (Roth *et al.*, 1967; Marczynski *et al.*, 1971a). Secondly, flash-driven ECoG activity at the frequency of PRS oscillations in a relaxed animal elicits a steady potential shift with amplitude and topographic distribution indistinguishable from the RCPV, and if the flash trains are repeated several times at random intervals, they produce slow wave sleep in an almost stimulus-bound manner (Marczynski & Sherry, 1972). Thirdly, studies of patterns of cortical and subcortical evoked potentials to auditory (Marczynski *et al.*, 1968), somatosensory (Marczynski & Hackett, 1969) and visual stimuli (Hackett & Marczynski, 1971) clearly show that during the 3–5 sec lasting bursts of PRS the processing of input, as judged on the basis of the shape of average evoked potentials, is similar to or sometimes indistinguishable from that characteristic of slow wave sleep or sleep onset. Also, transmission in the lateral geniculate nucleus and at the cortical level is strongly suppressed during slow bursts of PRS to a level seen during slow wave sleep (Hackett & Marczynski, 1971).

(3) *The role of earning the reward*

Although phenonena very similar to PRS–RCPV are observed during *ad libitum* consummatory responses (e.g. when the cat is lapping milk from a bowl), during petting, and particularly when the animal is grooming in the genital region (Fig. 1, bottom), these ECoG patterns are best developed and are most consistent during operantly conditioned behaviour. In

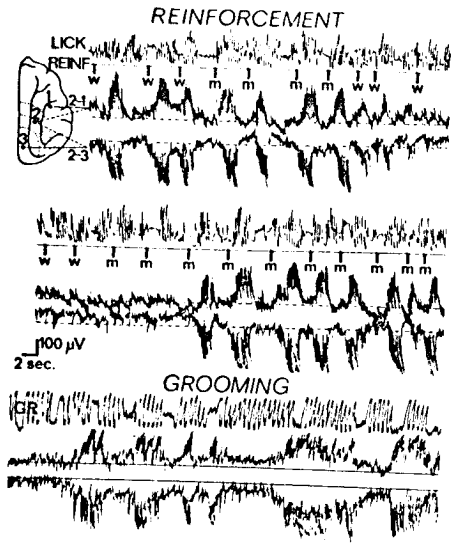

Fig. 1. *Top and middle* (a continuous record): The effect of reduction in quality of reward on the occurrence of PRS–RCPV responses. The d.c. recording from the anterior ectosylvian (1) and posterior marginal (3) gyri with reference to the medial suprasylvian gyrus (2). Substitution of milk (m) for water (w) rewards (approx 2 min after the deprived animal was allowed to press a lever) did not produce any significant change in the PRS–RCPV patterns. However, reintroduction of water abolished the PRS–RCPVs, although the rate of performance and consummatory responses were not changed. Subsequent reintroduction of milk reward restored the ECoG responses. Dotted lines of the diagram of the right hemisphere show the approximate distribution of isopotential lines in about 50 μV intervals during RCPV whose maximum potential of 180–250 μV occurs over the primary visual cortex (3). In this electrode arrangement the mirror reversal patterns of PRS–RCPV is explained by a decreasing potential gradient of RCPV along the '3–2–1' line.

Bottom: Steady potential shifts and bursts of PRS-like activity in the same subject after satiation during grooming. Note the similarity between the ECoG patterns during operant behavior and those during grooming. Note also a development of a relatively large (300–400 μV) epidurally recorded potential gradient between the electrode (3) and (1). These potentials as well as similar ones during sleep onset (not shown) do not depend on photic input. Abbreviations: LICK-, and GR-potentials caused by licking and grooming respectively. (From Marczynski *et al.*, 1971a).

most animals the PRS diminishes to the point of sporadic occurrence or complete disappearance once the lever is removed from the test chamber and the appropriate amounts of milk reward are presented freely by the experimenter at intervals corresponding to the rate of the animals lever presses. Thus, it appears that PRS is reduced in a non-operant paradigm as if the lever pressing and the effort involved in earning the reward adds to its value (Poshel & Ho, 1972). This anthropomorphic explanation may have a strong basis in known electrophysiologic mechanisms. First, there is substantial evidence that thalamocortical synchronization of the alpha type is based on recurrent hyperpolarizing inhibition of a large population of neurons as shown diagramatically in Fig. 2. This results in a tonic inhibition of a large number of relay neurons while, at the same time, other neurons show rhythmic rebound discharges which, via axon collaterals and inhibitory interneurons, recruit more neurons into the phasing process (Andersen & Andersson, 1968). This mechanism cannot function without a minimum synaptic "pressure" applied to the thalamic relay nuclei by sensory afferents as shown by our studies of the role of photic input in PRS. Secondly, the phasing of neuronal activity by hyperpolarizing inhibition, and resulting rebound rhythmic discharges that are essential for the emergence of PRS as well as the human alpha activity, can only occur during periods of slackening or active suppression of the tonus of the ARAS and its desynchronizing influences at the thalamo-cortical level. These influences dominate during arousal and are most likely mediated by the inhibitory catecholaminergic projections to the inhibitory interneurons (cf. Krnjevic, 1974), although the final proof for such an innervation is not yet at hand. The fact is that the desynchronizing ARAS influences exert a powerful control over the amplitude of the IPSPs in the thalamus (Purpura *et al.*, 1966) and cortex by modulating the firing rate of the inhibitory interneurons (cf. Steriade & Deschenes, 1973) and those located in the nuc. reticularis thalami (cf. Waszak, 1974) whose hyperpolarizing action is most likely GABA-mediated (cf. Curtis & Johnston, 1974).

Since transmission of sensory input in thalamic nuclei is correlated with the tonus of the ARAS which exerts a powerful facilitatory influence (most likely via a cholinergic mechanism as discussed later), a strong and tonic depression of the ARAS would result in reduction of synaptic "pressure" on the recurrent inhibitory phasing circuitry, leading to irregular slow wave patterns characteristic of sleep. In this condition no regular waxing and waning trains of alpha waves or PRS can be generated. On the other hand, a strong tonus of the ARAS (e.g. during an orienting reaction) will block the PRS and alpha-type synchronization by directly interfering with the function of the recurrent inhibitory phasing mechanisms. Hence, it can be concluded that a moderate tonus of the ARAS, or rather its fluctuation within moderate limits, is theoretically the most favorable condition for the emergence of PRS and other alpha-like activity. These conditions, including the proper dynamic balance between facilitation of sensory input and postreinforcement depression of the ARAS, are fulfilled during operant behavior.

(4) *Diffuse photic input and gustatory-induced ECoG phenomena*

The PRS–RCPV occurs only when the animal is fully habituated to the environment; any novel stimulus or change in the immediate surrounding abolishes

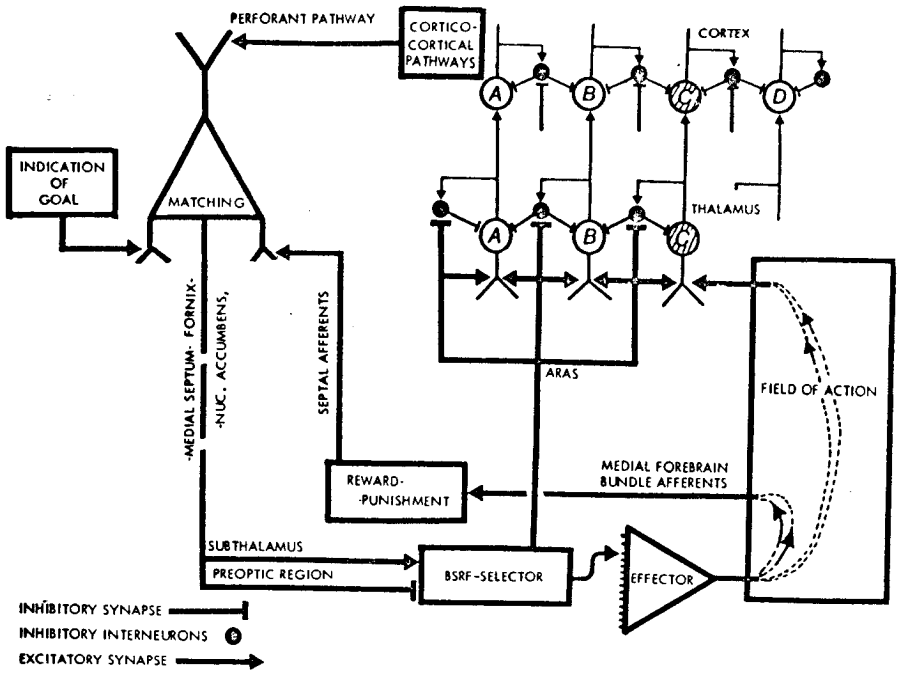

Fig. 2. Diagrammatic representation of the information flow most likely responsible for the emergence of the PRS–RCPV responses. For further explanations, see the text.

the PRS (Clemente *et al.*, 1964; Marczynski *et al.*, 1968). Suppression of PRS after turning off the light in an experimental chamber was ascribed to the orienting response and resulting activation of the ARAS (Buchwald *et al.*, 1964). However, later studies in which the cats have been trained to perform in the dark for several weeks to eliminate the "novelty" effect of the darkness showed that presence of light is necessary for the occurrence of PRS–RCPV. Moreover, it became clear that unpatterned light input and not the perception of the reward is essential, since PRS–RCPV in cats trained in the dark and wearing translucent "milky" contact lenses also depends on the presence of light (Marczynski *et al.*, 1971b). Mild electric stimuli applied to the optic nerve or the lateral geniculate nucleus during consummatory responses in the dark restore the PRS–RCPV to such an extent that the patterns and topographic distributions are indistinguishable from the spontaneous phenomenon. The same electrical stimuli are, however, ineffective during other behavioral states such as relaxed wakefulness or nonrewarded lever pressing performance (Rick & Marczynski, in preparation). Finally, in order to eliminate the orienting reaction to darkness and, at the same time, exclude the possibility of a negatively reinforcing quality of darkness, cats were trained for 2–3 months in an experimental paradigm in which "light-off" and/or "tone on" heralded the availability of milk reward. Animals learned to press the lever in the dark in response to these positively reinforced cues. Despite the long training, light was still necessary for the emergence of PRS–RCPV which, in most instances, was triggered in a stimulus-bound manner only if the light was turned on during the consummatory response and not during other phases of behavior (Rick & Marczynski, in preparation). All these observations taken together support the notion that a specific gustatory input plus a minimum synaptic drive provided by an unpatterned input in the visual pathways is necessary for the emergence of the PRS–RCPV phenomenon. This fact supports the general cybernetic law that self-organizing systems feed not only on well-structured input, i.e. neg-entropy but also on raw "noisy" energy (von Foerster, 1960; Nicolis *et al.*, 1975). The pharmacological aspects of the PRS–RCPV, particularly the effects of LSD, and of anticholinergic and cholinomimetic drugs discussed below, lend further support to this hypothesis.

(5) *Information flow and reward-induced thalamo-cortical synchronization*

Taking into account most observations so far accumulated on the electrophysiologic characteristics of the PRS–RCPV phenomenon, and the established functional anatomic relationships between the brainstem reticular formation, thalamus, neocortex and the limbic system (cf. Smythies & Adey, 1970), one can postulate the general flow of information, as depicted in a very diagrammatic form in Fig. 2, which also incorporates some of the cybernetic concepts of brain organization (MacKay, 1965). First of all, a general

indication of Goal must be internally generated in substrates responsible for the maintenance of the "milieu interior" or propagation of the species. The hypothalamic nuclei responsible for the regulation of hunger, thirst and sex, as well as the reward-punishment systems, are likely to be the main source of this activity which projects onto various parts of the limbic system, represented in Fig. 2 as a single pyramidal cell of the hippocampus (Matching). In higher mammals, and particularly in primates and man, the frontal association cortex plays an increasingly important role in the modulation of these drive-induced synaptic pressures on the limbic system. The hippocampal pyramidal cell was selected because its input–output relationships are very well documented anatomically and electrophysiologically. The limbic descending pathways can exert strong facilitatory or inhibitory influences on the brainstem reticular formation (BSRF), which fulfills two principal functions. First, it selects and determines the general mode of behavior and, in conjunction with the non-specific thalamic nuclei (not shown in Fig. 2), may determine more refined behavioral patterns by influencing the Effector which represents the motor repertoir of the body. The work of the Scheibels disclosing the profuse axonal ramifications, both ascending and descending, originating from a single BSRF neuron which may simultaneously affect hundreds of neurons in the thalamus, brainstem and cortex, clearly indicates that BSRF neurons play a role in selecting behavioral modes. The second major role is the modulation of thalamic relay nuclei, where it exerts a strong facilitatory influence on synaptic transmission, most likely through its cholinergic component, and, at the same time, regulates the function of the inhibitory interneurons through a different neurohormonal mechanism (see below).

During performance in the Field of Action, a wide range of sensory input is received which is processed in two distinct channels: one is the "visceral" or "interoceptive", which runs through the medial forebrain bundle, and is "evaluated" in the reward–punishment systems. Subsequently, the transforms of this input are projected to the limbic system (Matching), as shown in Fig. 2, to the basal dendrites of the hippocampal pyramidal cell. The second channel of sensory stimuli, experienced during a successful or unsuccessful interaction with the environment, projects to the specific thalamic nuclei and the neocortex, where the precise physical and spatial characteristics of the input are analyzed. Well described cortico-cortical pathways (Van Hoesen *et al.*, 1972) from primary receiving cortices to association areas, the temporal lobe and the entorhinal cortex project the transforms of polymodality interaction into the limbic system (Matching), onto the distal parts of the apical dendrites of the hippocampal cell. Hence, the latter simultaneously receives information from the "visceral" channel and the cortical pathways. By matching the two channels of information, complex processes of categorizing and assignment of values to the abstracted physical properties of the environmental stimuli take place, particularly in terms of reward–punishment criteria. The fulfillment of the general goal, or failure of fulfillment, can thus be identified with transforms of sensory input from the environment. If a particular mode of behavior is successful, it is reinforced and repeated many times until perfected, as in the case of lever pressing behavior in the cat. With the development of the animal's habituation to the environment and fixation of the refined behavior leading to reward, the limbic system appears to exert during consumption of reward a strong inhibitory influence on the BSRF-Selector and the ARAS. Hence, a sudden shift in the tonus of the ARAS occurs, from the desynchronizing influences on the thalamo-cortical system, associated with increased transmission of sensory input, to a sudden removal by the limbic system of the ARAS desynchronizing influences, thus allowing the recurrent inhibitory circuits to function, and PRS at the cortical and thalamic levels to emerge. The generation of PRS is possible because of the rapid recruitment of a larger population of thalamic relay neurons via inhibitory interneurons which establish contact not with one, but with many relay neurons (In Fig. 2, only two neurons are shown to be affected). Although the majority of relay neurons remain hyperpolarized and silent, many of them, after developing a 100–150 m sec IPSP which corresponds to a PRS rhythm of 6.7–10 Hz, show a rebound discharge and, in turn, recruit more neurons in the rhythmic IPSP–EPSP sequences and postinhibitory discharges (Andersen & Andersson, 1968) reflected at the cortical and the lateral geniculate level as PRS. In the visual cortex and in the thalamus the majority of neurons during PRS remain silent and hyperpolarized (Karmos & Marczynski, unpublished) thus contributing to the surface slow positive steady potential shift i.e. RCPV, illustrated in Fig. 1 top and middle. The electrogenesis of ECoG patterns during grooming (Fig. 1, bottom) is most likely similar to that of PRS-RCPV, although the "electromotive" energy for the recurrent inhibitory circuits is provided by other modalities than visual since these ECoG patterns do not depend on photic input, although their topographic distribution is indistinguishable from that of the PRS–RCPV (Marczynski *et al.*, 1971a).

As is evident from the outline of the postulated PRS mechanisms, a sufficient synaptic drive provided to the thalamic nuclei, and particulary to the lateral geniculate nucleus, is necessary for the prompt initiation of the recurrent inhibitory phasing mechanism. The appearance of PRS is dependent on the palatability and appropriateness of the gustatory reward because the goal becomes gradually refined by positive feedback from the reward-punishment system. Thus, if the expected reward is replaced by another less palatable, the sensory transform does not "match" the more specific indication of Goal, and no

PRS–RCPV is generated, as illustrated in Fig. 1, top and middle.

B. PHARMACOLOGICAL ASPECTS OF PRS AND RCPV

(1) *LSD-25 (Marczynski, 1972)*

In cats trained to perform in the dark and not displaying PRS–RCPV, LSD-25 administered in doses of 15–30 μg/kg i.m. restores these responses which normally are seen only in the presence of light. Thus, it appears that LSD through its action substitutes for the unpatterned light input. The effectiveness of LSD can be compared to that of electrical stimulation of the optic nerve or the lateral geniculate, and the patterns of PRS, (i.e. the amplitude and duration of the 7–9 Hz spindle-like burst of high voltage slow waves) do not significantly differ from those observed without the drug treatment in the presence of light. The action of LSD appears to be specific, since other drugs such as morphine, barbiturates in small doses, major tranquilizers like chlorpromazine, despite their ability to enhance the PRS–RCPV phenomenon in the presence of light, are not able to restore these responses in the dark (Marczynski & Hackett, in press). The mechanism of LSD action is not clear. However, a possible explanation for the ability of LSD to substitute for photic input is the observation that this hallucinogen administered i.v. to cats markedly stimulates the discharge rate of approx two-thirds of the retinal ganglion cells while at the same time inhibits the remaining one-third (Mouriz-Garcia *et al.*, 1969). This effect resembles that of photic stimulation of the retina, which is known to activate the on-center ganglion cells while inhibiting those located off-center (Barlow & Levick, 1969).

Another interesting effect of LSD is that the reward-induced patterns after a dose of 15–25 μg/kg i.m. occur not only during consumption but equally often during non-rewarded lever pressing when the animal licks the empty cup or looks at the manipulanda. This uncoupling between the ECoG and consummatory responses occurs much more often (up to 70% of the total number of PRS–RCPVs) in the presence of light. Its occurrence does not appear to be related to retinal stimulation since comparable percentages of dissociated responses are observed after morphine or barbiturates which do not restore PRS–RCPV in the absence of light. Whether this phenomenon is correlated with slowing of the PRS oscillations, as in the case of morphine, remains to be investigated. Larger doses of LSD-25 than 30 μg/kg i.m. interfere with operant performance even in well deprived animals.

(2) *Dual effect of morphine (Marczynski & Hackett, in press)*

Contrary to the general belief that cats typically respond to morphine with behavioral excitement and a manic-like syndrome, it was shown that doses of 0.1–0.4 mg/kg, i.m. cause a dramatic (up to 210% over control) enhancement of the PRS–RCPV responses quantified in units of RCPV (one unit is equal to a 100 μV positive steady potential shift lasting one second; for technique of measuring the RCPV responses, see: Marczynski, 1971a,b). Higher doses of morphine (0.7–0.9 mg/kg, i.m.) have clearly a biphasic action: the initial strong potentiation of the PRS–RCPV responses is followed within 10–20 min by their suppression and abolition, despite the fact that the animals continue lever pressing for some time. In the later phase, however, restless behavior interferes with performance. Chlorpromazine (1.0–1.5 mg/kg, i.m.) promptly restores the performance and the PRS–RCPV.

During the peak effect of the potentiating doses of morphine, the reward-related EEG phenomena also occur prior to or after the nonrewarded lever press when the "frustrated" animal licks the empty cup. This dissociation or uncoupling of the PRS–RCPV from consummatory responses may reflect the euphoric action of morphine, and reaches in some animals 85% of the total PRS–RCPV occurrences. The uncoupling phenomenon is much more conspicuous in animals whose control frequency of PRS oscillations is higher, and after morphine shows a more significant slowing. Despite the strong potentiation of the PRS–RCPV in the presence of ambient light, morphine in contrast to LSD-25, is not able to restore the reward-induced phenomena in the dark.

It can be argued that the morphine-induced potentiation of PRS–RCPV is caused by a different neurohormonal mechanism than the suppression of these responses by higher doses. The latter is most likely caused by activation of the dopaminergic system or its receptors, since: (a) central catecholamine depletors, such as reserpine or tetrabenazine, or central dopaminergic receptor blocking agents, such as haloperidol or chlorpromazine, prevent the behavioral and autonomic responses to larger doses of morphine; (b) alpha- or beta-adrenergic receptor blocking agents, such as phenoxybenzamine or propranolol, do not antagonize the morphine mania; and (c) anticholinergics (atropine), antihistaminic drugs (mepyramine), and antiserotonin agents, such as LSD-25, do not prevent morphine mania (cf. Dhasmana *et al.*, 1972). The mechanism of the dramatic enhancing effect of smaller doses of morphine on the PRS–RCPV has to be investigated. Because of the clear-cut biphasic action of morphine on the PRS–RCPV phenomenon, cats may be much more valuable as experimental animals than other species.

(3) *Cholinergic mechanisms*

(a) *Blockade by antimuscarinic drugs.* Since the time of the observation first made by Wikler in dogs (1952, cf. Longo, 1966) it has been known that atropinized animals (cats, rabbits, rats, monkeys and man) display high voltage slow wave ECoG activity regardless of whether they remain in a sleep posture with eyes closed or explore the environment. The patterns of

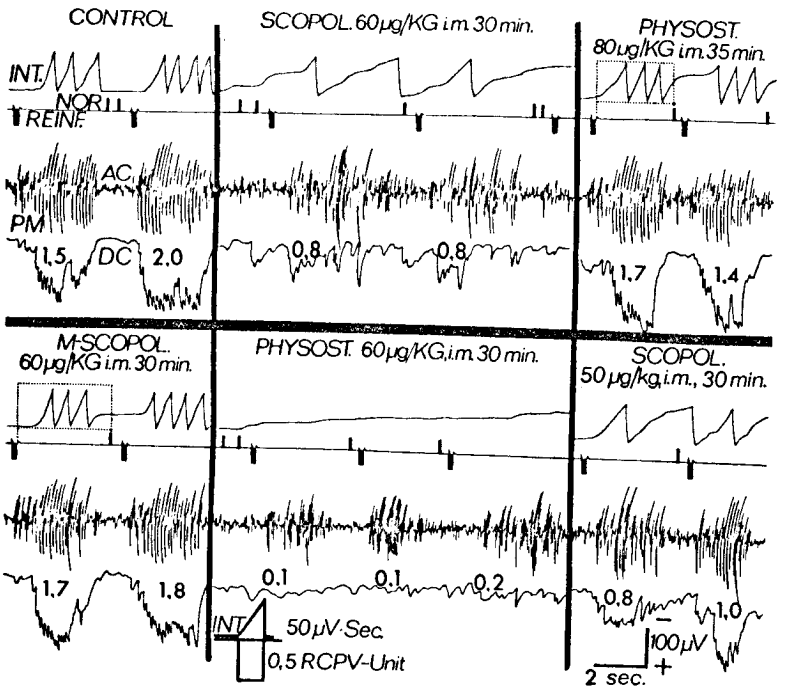

Fig. 3. *Top*: The effect of scopolamine hydrobromide and physostigmine salicylate on PRS and RCPV recorded from the primary visual cortex (posterior marginal gyrus) with respect to subjacent white matter. The d.c. record is filtered to half-amplitude response at 3 c/sec, and this channel is integrated in the first one (INT) in which two full scale upward deflections are equal to one arbitrary unit of RCPV. The deflections were summated in the time period between the rewarded bar press and the next bar press as marked by dotted lines in the upper right record and the bottom left record. Note that scopolamine suppressed the PRS–RCPV. The duration of the choppy PRS is not changed. Physostigmine promptly restored the PRS–RCPV.

Bottom: 48 hr later, in the same cat (pretreated with peripherally acting scopolamine methylbromide) physostigmine salicylate abolished the PRS–RCPV. The PRS bursts were replaced by smaller amplitude and faster frequency (16–18 c/sec) spindle bursts which were not associated with any significant steady potential shifts. Subsequent administration of scopolamine hydrobromide, in some instances, partially restored the PRS–RCPV. However, central catecholaminergic blocking agents, such as chlorpromazine, were more effective in restoring these responses (not shown). (From Marczynski, 1971).

this dissociated ECoG, although resembling slow waves characteristic of sleep, substantially differ from the latter because of the presence of high voltage spikes intermingled in a random fashion with the slow wave activity (Marczynski, 1971). Originally, it was thought that a total dissociation of ECoG patterns from behavior could be achieved by appropriate doses of anticholinergics; later studies, however, have clearly shown that there are very strict limits to such dissociation since animals trained to perform conditioned responses either fail to respond or cannot learn to respond when the ECoG background reaches a certain level of synchronization induced by anticholinergic drugs (cf. Longo, 1966). The primary action of the anticholinergics is the reduction or blockade of the ascending desynchronizing influences of the ARAS, as demonstrated by atropine- or scopolamine-induced prevention of ECoG desynchronization normally observed in response to sensory stimulation or direct electrical stimulation of the ARAS (Rinaldi & Himwich, 1955; Bradley & Elkes, 1957; Bradley & Key, 1958; Paul *et al.*, 1959; cited by Longo, 1966). It has also been shown that single doses of atropine sulfate given to healthy volunteers clearly have a dual effect: (1) They markedly reduce the duration of ECoG desynchronization induced by photic stimuli, thus confirming the results from animals; and (2) They simultaneously block the alpha-type synchronization reducing the alpha index by an average of 60% (Ostfeld *et al.*, 1960; cf. Longo, 1966).

In cats the anticholinergics slow the rate of lever pressing and, with higher doses (atrophine sulfate 0.8 mg/kg i.m.; scopolamine hydrobromide 0.07 mg/kg i.m.) during the peak effect, the animals perform only sporadically in periods of 2–3 min during which the rate of performances is often comparable to that of control, but the PRS–RCPV responses are blocked or become "choppy", as shown in Fig. 3 (top, middle), in which the PRS shows random changes in amplitude and frequency of oscillations, and the 7–9 Hz

rhythm never occurs in trains longer than two to three consecutive waves (Marczynski, 1971). Hence, in response to antimuscarinic drugs the PRS behaves in the same manner as the reward-induced "masticatory waves" in rabbits (cf. Longo, 1966) or the human alpha activity, despite the fact that the latter does not depend on photic input.

(b) *Duration of PRS–RCPV.* When the duration of the "abortive" choppy PRS associated with small amplitude RCPV is measured, and the mean values compared with control values in the same animal, no significant differences are found. Properly adjusted doses of atropine sulfate (0.5–0.8 mg/kg i.m.) are more suitable for such study because, in contrast to scopolamine, atropine appears to have a weaker synchronizing effect on the ECoG background and less suppressant action on the PRS, and this makes it easier to decide when a PRS burst begins and ends (Marczynski, 1971). This "immunity" of the duration of the choppy PRS indicates that: (1) the antimuscarinic drugs do not interfere with the "primary" effect of reward on substrates that set the specific internal state and background for the emergence of PRS–RCPV but, instead, they interfere only with the mechanism responsible for the "execution" of the PRS response, most likely at the thalamocortical level (see Hypothesis of Cholinergic Mechanism).

(c) *Dual action of physostigmine.* Physostigmine salicylate (0.06–0.08 mg/kg i.m.) administered to cats in whom the PRS–RCPV had been blocked by atropine or scopolamine promptly restores the PRS–RCPV, as well as the background low voltage desynchronized patterns and the normal rate of lever pressing. On the other hand, the same doses of physostigmine salicylate that restore the PRS–RCPV in atropinized animals, if administered prior to anticholinergics, block the PRS–RCPV without prior enhancement (Fig. 3 bottom). During the peak action, a variant of PRS emerges characterized by higher frequency (13–16 Hz) oscillations organized in spindle bursts, which are not associated with any significant RCPV. This effect is centrally mediated since it occurs in animals pretreated with peripherally acting scopolamine methyl bromide. The mechanism of action of physostigmine appears to be two-pronged: (1) this anticholinesterase stimulates the cholinergic component of the ARAS; and (2) most likely it increases the tonus of the ascending catecholaminergic system projecting to the forebrain and responsible for ECoG and behavioral arousal. The presence of the cholinergic link in catecholaminergic arousal and ECoG desynchronization had been postulated some time ago as an extrapolation of the "peripheral" Burn–Rand hypothesis on the basis of similar EEG and behavioral effects of microinjections of norepinephrine and physostigmine into subcortical regions involved in the regulation of sleep–waking cycle (Marczynski, 1967). More recently, this suggestion has been supported by a series of studies on the cholinergic link in adrenergic suppression of the synchronizing recruiting system in the non-specific thalamic nuclei of the rabbit (cf. van Meter & Karczmar, 1971) and in the emergence of paradoxical or REM sleep (Karczmar *et al.*, 1970). In either case the depletion of norepinephrine prevented the effect of physostigmine.

The physostigmine—or nicotine-induced suppression of the PRS–RCPV—can be more successfully reversed by chlorpromazine than by antimuscarinic drugs (Marczynski, unpublished). This emphasizes the complexity of transmitter interactions in the central nervous system and suggests, as already mentioned, the possibility of catecholamine involvement in desynchronizing influences of the ARAS on the thalamocortical relationships, most likely via depression of the inhibitory interneurons whose rate of firing appears to be directly affected by the ARAS (cf. Steriade & Deschenes, 1973).

C. A NEW INTERPRETATION OF THE CHOLINERGIC MECHANISM IN PRS AND HUMAN ALPHA ACTIVITY

To account for the suppression of PRS as well as for the simultaneous increase in background ECoG synchronization after antimuscarinic drugs, we have previously postulated two pharmacologically identical but functionally opposing cholinergic systems: one responsible for desynchronizing influences of the ARAS—and another indirectly involved in the function of the inhibitory phasing circuits (Marczynski, 1971), whose presence in the thalamus and cortex is well established (cf. Curtis, 1969). Although it would appear premature to absolutely rule out the inhibitory and hyperpolarizing action of ACh (Phillis, 1971), an overwhelming amount of evidence indicates that ACh is primarily an excitatory transmitter at the thalamic and cortical levels and that, instead, GABA plays the role of the hyperpolarizing transmitter in the recurrent inhibitory pathways (cf. Curtis, 1973; Curtis & Johnston, 1974). The release of GABA from the cortex is proportional to the level of ECoG synchronization and inversely proportional to the level of ACh release and the tonus of the ARAS (cf. Curtis, 1969).

After reconsidering our own data and those obtained by others, we now propose a more plausible hypothesis of the PRS mechanism and the role of the cholinergic system which, *mutatis mutandis*, can be extrapolated to the human alpha activity. We postulate that: (a) it is not necessary to assume the presence of two functionally opposed cholinergic systems to explain the blocking action of antimuscarinic drugs on PRS and the human alpha activity; (b) a spontaneous slackening of the tonus of the cholinergic component of the ARAS and/or pharmacologic blockade of its facilitatory influences on input to the thalamic relay nuclei and cortex, are sufficient to explain the suppression of PRS and the human alpha activity; and (c) the choppy PRS, as well as the irregular ECoG slow wave activity, characteristic of central antimuscarinic action and spontaneous slow wave sleep, reflect the irregular "idling" of the recurrent

inhibitory phasing mechanisms caused by insufficient synaptic pressure on the thalamic relay neurons. In support of our hypothesis the following evidence can be summarized:

(1) The cholinergic component of the ARAS originates in the reticular formation and the tegmental nuclei, and via numerous cholinergic-cholinoceptive relays, projects to the specific thalamic nuclei, neocortex and the limbic system (Shute & Lewis, 1967).

(2) Stimulation of a specific sensory pathway enhances ACh release from both the primary receiving cortical region, and to a lesser extent, from other cortical areas. This is particularly true for the visual modality (cf. Curtis, 1969).

(3) Considering the physiologic role of subcortical and cortical cholinergic projections of the ARAS, overwhelming evidence indicates their principal role is facilitation of synaptic transmission in the specific thalamic relay nuclei and cortex. The lateral geniculate neurons appear to be particularly sensitive to visual input after "priming" with electrophoretically applied ACh (Steiner, 1968). The same is true for the ventrobasal nuclear complex (McCance *et al.*, 1968) and the medial geniculate (Tebecis, 1970; Phillis, 1971). In all instances, electrical stimulation of the ARAS mimicked the effort of ACh, and atropine or scopolamine blocked both the ARAS influence and the effect of iontophoretic application of ACh.

(4) The inhibitory phasing theory of alpha activity (Andersen & Andersson, 1968) implies that a minimum input to the thalamic relay neurons is necessary to initiate and drive the recurrent inhibitory circuits.

(5) The PRS depends on unpatterned photic input, which can be substituted with electric stimulation of the optic nerve or the lateral geniculate nucleus, and administration of LSD-25 which seems to mimick the photic input. Thus, a "raw" electromotive energy is required and utilized by the recurrent inhibitory phasing system, and not the fine abstracted transforms of a specific visual information.

(6) The best developed PRS-RCPV responses are observed when the animal has to earn the reward, i.e. when a high tonus of the ARAS is suddenly reduced or actively inhibited by influences originating in the forebrain, as during a lever press and subsequent consummatory response, the first phase being associated with ARAS facilitation of sensory input, and the second phase with utilization of input in the recurrent inhibitory pathways.

(7) The duration of the "abortive" or choppy PRS after antimuscarinic drugs is not significantly different from normal PRS, thus indicating that, due to insufficient synaptic pressure, irregular "idling" of the phasing mechanisms is the main cause of the appearance of these choppy waves. In many instances, the "abortive" PRS could be restored to normal PRS simply by increasing the intensity of ambient light in the test chamber (Marczynski, unpublished), although the most reliable effects are obtained with moderate doses of physostigmine.

(8) Physiologic utilization of unpatterned visual input in internal inhibitory processes is exemplified by the demonstration that flash stimuli presented at the PRS rhythm to satiated animals have a strong hypnogenic effect (Marczynski & Sherry, 1972) which can be antagonized by antimuscarinic drugs (Marczynski, unpublished).

(9) In slow wave sleep, the irregular delta waves that emerge always after alpha-like activity can be conceptualized as a state in which the reduced "electromotive" energy in thalamic and cortical recurrent inhibitory circuits is insufficient to produce long trains of alpha waves. This view is supported by the observations that during delta slow wave sleep in the cat, presentation of mild auditory, somatosensory or visual stimuli cause bursts of PRS-like activity in a stimulus-bound manner. The phenomenon is associated with a large positive epicortical steady potential shift whose topographic distribution over occipital cortex is virtually identical to the RCPV and patterns illustrated in Fig. 1 (bottom) during grooming. Moreover, antimuscarinic drugs block these responses (Marczynski, unpublished).

It is true that in approx 3% of cats the PRS–RCPVs are equally well developed both in the presence and absence of light (Rick & Marczynski, in preparation), and ECoG phenomena virtually identical with PRS–RCPV occur during grooming and sleep onset and are independent of light input (Marczynski *et al.*, 1971a). Unpatterned light input is not necessary for and has no effect on the occurrence of the human alpha activity (Adrian & Matthiews, 1934; Bridgwater *et al.*, 1975) which, in most instances, is enhanced by eye closure. All these observations do not contradict our hypothesis on the mode of action of antimuscarinic drugs in blocking the thalamocortical ECoG synchronization, since visual input is not the only modality that can provide synaptic drive for the recurrent inhibitory phasing circuitry. There is ample evidence for somatosensory, auditory and even vestibular input to the striate and parastriate cortex (cf. Morrell, 1972) where PRS and human alpha originate. The calming and hypnogenic effect of cradle rocking, coupled with the singing of monotonous lullabies, are well known to all civilizations, and these rituals testify to the electrophysiologic principles discussed above.

D. BEHAVIORAL "DISINHIBITION" CAUSED BY ANTICHOLINERGIC DRUGS

Characteristic of the central anticholinergic syndrome, various forms of behavioral "disinhibition" are observed, such as the inability to suppress previously learned responses that become inappropriate in a changed set of circumstances, and other forms of perseverative behavior (cf. Longo, 1966). We propose that they are not caused by blockade of hypothetical cholinergic inhibitory synapses but, instead, by removal of the moderate facilitatory cholinergic

mechanisms in the thalamus and cortex, necessary for normal functioning of the recurrent inhibitory phasing mechanisms. The resulting alpha-like activity subserves the internal inhibitory processes responsible for shaping behavioral patterns. There is ample evidence that the thalamo-cortical synchronization of 6–9 c/sec exerts a strong modulating influence on the brain stem ARAS neurons (Schlag *et al.*; cf. Marczynski, 1967) which are regarded as main determinants of general behavioral modes (Scheibel; cf. Smithies & Adey, 1970).

E. NEW VISTAS: POSTREINFORCEMENT AND SLOW WAVE SLEEP "ENTROPY"

There is a general consensus among neurophysiologists that the "frequency code" of neuronal firing is too simple and inefficient to handle the wealth of information carried and processed by the polymodality systems of the mammalian brain (Segundo, 1970; Perkel, 1970). Statistically significant and complex temporal patterns, present in long trains of single unit spike intervals recorded from the thalamus and the basal ganglia, tend to disappear during bursts of ECoG synchronization and particularly during slow wave sleep, and their probabilities of occurrence fall to the theoretical chance level (Marczynski & Sherry, 1971; Sherry & Marczynski, 1972; cf. Marczynski, in press). This indicates that bursts of ECoG synchronization are associated with relaxation of constraints normally imposed on neuronal activity during a state of arousal. One can postulate that drugs which cause perseverative behavior, such as antimuscarinics or stimulants of the ARAS (amphetamine or physostigmine) will block the dynamic shifts in the emergence and suppression of patterns. Studies of complex man-made self-organizing systems clearly show that a phasic introduction of pseudorandom noise, resulting in a transient increase in "entropy" of the system, is necessary to counteract the persistent tendency toward perseveration of certain operational modes, and to preserve the changes for emergence of new functional modes (cf. von Foerster, 1960; Nicolis *et al.*, 1975). One can further postulate that slow wave sleep and associated "entropy" of neuronal connectivity, reflected as an almost random and/or independent neuronal output, have a recuperative role, by "erasing" some of the plastic and nonsignificant changes that took place during the waking state accompanied by patterned neuronal activity. Pharmacologic studies of patterns in neuronal output may add a new dimension to the understanding of brain function. These and related theoretical matters have been discussed elsewhere (cf. Marczynski, in press).

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