EFFECTS OF MORPHINE ON EVOKE POTENTIALS
RECORDED FROM THE AMYGDALA BY TOOTH
PULP STIMULATION IN CATS*

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Abstract—Effects of intravenously administered morphine on the evoked potentials of
the amygdala elicited by tooth pulp stimulation were examined in cats. The various
evoked potentials were observed in regions of the amygdala such as nucleus
amygdaloideus centralis (pars lateralis), nucleus amygdaloideus basalis (pars magnocellularis),
nucleus amygdaloideus basalis (pars parvocellularis) and nucleus
amygdaloideus lateralis. Evoked potentials were significantly decreased by morphine
in four of the recorded regions. Morphine had no effect on the latency at any site
of the amygdala observed. Depressant effects of morphine on evoked potentials
were antagonized by naloxone in all 14 cats. This study indicates that there is a receptive
field of pain in the amygdala which undoubtedly plays a role in emotion.

Jelasic reported diminished pain sensation and the emotional component of pain after
bilateral amygdalecctomy in a case of intractable trigeminal neuralgia (1). Rodgers has
directly shown that bilateral micro-injection of morphine into the cortico-medial amygdala
produced a dose-dependent increase in aversive threshold (2). Moreover, in recent years,
the greatest degree of opioid receptor binding was found to be localized within the amygdaloid
region of the limbic system while the periaqueductal grey matter exhibits a high level of
opioid receptor binding and is an important site of morphine analgesia and tolerance (3).
These studies suggest that there is a relationship between the local distribution of the
morphine-like factor, enkephalin, in the amygdala and analgesic action of morphine. In
the present investigation, we attempted to elucidate the action of morphine on the evoked
potentials in various nuclei of amygdala produced by tooth pulp.

MATERIALS AND METHODS

Animals and drugs used: Twenty-three adult cats weighing 2.5 to 4.0 kg and the
following drugs were used: morphine hydrochloride (Sankyo) anesthetic ether (Sanraku
Ocean), pentobarbital sodium (Dainippon), atropine sulfate (Tanabe), gallamine triethiodide
(Teikoku), xylocaine jelly (Fuisawa) and naloxone hydrochloride (Endo Laboratories).
Morphine hydrochloride and naloxone hydrochloride were dissolved in Ringer’s solution.

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Electrode implantation: The implantation of bipolar stainless steel electrodes was performed under pentobarbital anesthesia (35 mg/kg, i.p.) and experiments were conducted at least 10 days after this implantation. The electrodes were coated with cashew paint and had a tip diameter of approx. 0.2 mm. The pole distance was 0.5 mm. The electrodes were implanted into the amygdaloid complex according to the stereotaxic coordinates of Jasper and Ajmone-Marsan (4); nucleus amygdaloideus lateralis [Al], nucleus amygdaloideus basalis (pars parvocellularis) [Abp], nucleus amygdaloideus centralis (pars lateralis) [Acl] and nucleus amygdaloideus basalis (pars magnocellularis) [Abm]. Electrodes were placed contralateral to the stimulated tooth pulp.

Acute experiments: Tracheal and vascular cannulations were performed under ether anesthesia, all operating wounds were carefully closed with sutures and xylocaine jelly was applied. The animals were then placed in a stereotaxic apparatus and immobilized with gallamine triethiodide under artificial respiration. Two small operating holes were drilled bilaterally into the dentine of the lower canine teeth and monopolar stainless steel electrodes (0.2 mm diameter) were inserted into the holes. These holes were covered with dental cement together with electrodes. The tooth pulp was electrically stimulated. (Square wave pulses at a frequency of 0.1 per sec, pulse duration of 0.5 msec and supramaximal intensity at intervals of 5 sec were used.) At least, 3 hr after discontinuing the ether inhalation, experiments were begun. The animals were free from pain as waves in the continuously recorded EEG were synchronized (somatic sensory area I). Body temperature was kept at 37.5–38.5°C. Evoked potentials in this experiment were obtained by 40 stimuli and added together by signal processor (San-ei Instrument Co., Ltd.). These potentials were stored in a FM magnetic tape and reproduced for the purpose of data reduction.

Histology: At the end of the experiments, currents of 20 μA were passed through the electrode for 15 sec to mark the position of the electrode tip. After the animals were sacrificed by an intraveous administration of sodium pentobarbital, 10% formalin with about 1% potassium ferrocyanide was injected into the carotid artery for determination of the site of electrode. Frontal serial frozen sections of 25 μm thickness were stained with 0.1% cresyl violet and recording sites were determined.

RESULTS

Field potentials evoked by tooth pulp stimulation: Field potentials generated by lower canine tooth pulp stimulation were systematically recorded in the nucleus of the amygdala complex. Fig. 1 illustrates the distribution of potentials 0.65 mm caudal to obex. The trace of electrode penetration is illustrated on the left. Various potentials were found the medial and lateral amygdala; potentials of high amplitude and reversive polarity were evident in the region of Abm.

Action of morphine on field potentials in the subnucleus of amygdala by tooth pulp stimulation: The latency and the peak to peak amplitude of evoked potentials in the Acl were 21.5 ± 2.6 msec and 44.5 ± 13.6 μV, respectively. The administration of morphine 1 mg/kg i.v. decreased the amplitude of Acl without affecting the latency. To examine the
specificity of the morphine effect, we attempted to reverse its depressive action (lower amplitude) by giving naloxone 0.2 mg/kg i.v. (Fig. 2). In the Acl, the depression of peak to peak amplitude by morphine was observed in all 6 trials, but completely recovered to the control following administration of naloxone in 4 of 6 trials. The average value of 6 trials is shown in the Table 1. The potentials recorded at the Al, which had latencies of 48.4±2.1 msec and amplitude of 58.7±9.1 μV, were affected in 5 of 13 trials by morphine and naloxone. As seen in a typical example illustrated in Fig. 3, the administration failed to decrease the low potential of the early peak, though the second peak was decreased. The decreased potentials

**Fig. 1.** Field potentials generated by lower canine tooth pulp stimulation. Abbreviations in this and succeeding figures are as follows. Put: Putamen; GP: Globus pallidus; Acl: Nucleus amygdaloideus centralis; Abm: Nucleus amygdaloideus basalis (pars magnocellularis); Abp: Nucleus amygdaloideus basalis (pars parvocellularis); Al: Nucleus amygdaloideus lateralis; To: tractus opticus.

**Fig. 2.** Effects of morphine and naloxone on evoked potentials recorded from Acl by tooth pulp stimulation. Acm: Nucleus amygdaloideus centralis.
TABLE 1. Effects of morphine and naloxone on evoked potentials in the amygdala by tooth pulp stimulation.

| Number of electrodes | Amplitude before drug administration | 15 min after morphine | 30 min after morphine | 15 min after naloxone | 30 min after naloxone |
|----------------------|--------------------------------------|-----------------------|-----------------------|-----------------------|-----------------------|
| Acl 6                | 100±0                                | 35.3±5.8*             | 37.6±7.5*             | 72.3±11.8**           | 57.9±6.7**           |
| Abm 4                | 100±0                                | 78.9±11.2             | 66.0±13.2**           | 77.1±5.9             | 67.3±5.8             |
| Abp 5                | 100±0                                | 94.4±13.9             | 69.3±11.2**           | 84.6±4.7**           | 91.7±2.9**           |
| Al 13                | 100±0                                | 50.3±5.6*             | 42.0±5.8*             | 68.2±7.6*            | 69.1±7.6*            |

*P<0.01 when compared with "before". **P<0.05 when compared with "before". 
*p<0.01 when compared with "30 min after morphine". “p<0.05 when compared with "30 min after morphine". Amplitudes are expressed as percentages of the "before drug" amplitudes. Naloxone was administered i.v. 30 min after morphine.

FIG. 3. Effects of morphine and naloxone on evoked potentials recorded from Al by tooth pulp stimulation. Am: Nucleus amygdaloideus medialis.

FIG. 4. Effects of morphine and naloxone on evoked potentials recorded from Abm by tooth pulp stimulation.
produced by morphine returned to nearly the control after administration of naloxone. The late wave was largely unaffected by morphine and naloxone (Fig. 3). The recovery of peak to peak amplitude by naloxone was observed in one of 4 trials in the Abm and 3 of 5 trials in Abp (Figs. 4 and 5, respectively). Twenty-one of 29 evoked potentials indicated a depression of amplitude as induced by morphine. Fourteen of 21 evoked potentials were recovered by naloxone (Fig. 6). The latency and peak to peak amplitude of evoked potentials in Abp were 37.5±2.8 msec and 60.3±11.6 μV, respectively. The decreased amplitude caused by morphine was not recovered by naloxone. These results are summarized in Table 1.
DISCUSSION

Extensive studies on the amygdala have been done not only by stimulation of the amygdala itself (5–8) but also by peripheral stimulation (9–12) in order to elucidate the functional significance. Dell and Olson (9) regularly encountered evoked potentials on vagal stimulation in the basolateral portion. Machne and Segundo (10) reported that a few amygdala neurons were activated by tooth pulp, vagus, sciatic, olfactory bulb or acoustic stimulation, and frequently, no unit activity was observed at sites where an evoked potential was recorded. Conversely, unit activity was observed in the absence of an evoked potential in the amygdala. Despite the discrepancy between unit activity and evoked response, our finding that the amplitude of evoked potential was decreased by morphine and recovered by naloxone means that among the diverse actions of morphine, including analgesia, there are effects on the nuclei of the amygdala complex.

Recently, it was shown that discrete areas of the monkey and human brain such as amygdala, periaqueductal grey matter and medial thalamus, are particularly rich in opiate receptors (13, 14), and a bilateral micro-injection of morphine into the cortico-medial region of the amygdala produced a dose-dependant increase in aversive threshold while similar injection into the basolateral amygdala or caudate-putamen failed to have any consistent effect on the aversive threshold (1, 15). Lico et al. observed that the lateral region elicited predominantly an analgesic-like effect while the medial region yielded preferentially a pain sensation (16). Jelasic reported diminished pain sensation and emotional component of pain after bilateral amygdalectomy in a case of intractable trigeminal neuralgia. He also noted that the patients experienced a sudden ipsilateral pain deep in the frontal area, behind the eye when the tip of the electrode entered the amygdala (1). The regional distribution of substance P concentration in the central nervous system is remarkably similar to that for opiate receptor binding sites (3, 17). Since substance P may play an excitatory role in the spinal pain pathway (18), this substance acts as a transmitter or modulator of excitability at the first afferent synapse, and is particularly abundant in the medial amygdala (15, 19), therefore a correlation between this region and pain can be presumed.

Our study clearly demonstrates that there are sites where the effects of morphine on the evoked potentials are antagonized by naloxone and other sites which are not affected by naloxone. This difference may depend on the distribution of opiate receptors and their sensitivity within the amygdala. Evidence concerning morphine analgesia (3, 15) and pain sensitive neurons in the amygdala (10, 11) has suggested that this area is one of most important nociceptive sites though there are discrepancies between the distribution of enkephalin and opiate receptors in the amygdala. The amygdala contains the highest density of opiate receptor binding in the monkey brain but only moderate to low levels of enkephalin (20).

Our studies suggest that there is a receptive field of pain in the amygdala which plays an important role in emotion.
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