CLINICAL ACTIONS OF FENTANYL AND BUPRENORPHINE

The Significance of Receptor Binding

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Opioid analgesic drugs are used with increasing frequency during surgical anaesthesia to control the endocrine and metabolic responses to stress (Hall, 1980). Fentanyl, one of the more effective agents in this respect, is a lipid-soluble agonist of high intrinsic activity with ready access to the brain and opioid receptor sites. It produces a rapid and dose-dependent clinical response which reaches the maximum attainable for this class of drug. However, in the absence of other anaesthetic supplements, the dose of fentanyl required to achieve full analgesia is 25 μg kg⁻¹ or higher and the ensuing postoperative respiratory depression reduces the clinical benefit of its intraoperative use (Cooper et al., 1981). Pharmacological control of this respiratory depression which allows adequate postoperative analgesia would extend the intraoperative use of opioid anaesthesia.

Buprenorphine is a partial agonist, also of high lipid solubility and receptor affinity but of a lower intrinsic activity (Dum and Herz, 1981). This and other features of its actions have restricted its application in intraoperative anaesthesia, although as an analgesic for postoperative use its efficacy is well recognized (Harcus, Ward and Smith, 1980; Ellis et al., 1982). Buprenorphine is also slow to reach its full effect, but once established it has a long duration of effect resistant to antagonism with naloxone (Orwin, 1977; Gibbs, Johnson and Davis, 1982).

We have conducted a series of in vitro experiments to examine individual and interactive receptor binding properties of these two compounds in an attempt to understand possible receptor mechanisms which may mediate the different clinical effects of these drugs.

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SUMMARY

Receptor binding assays were undertaken in an attempt to elucidate the opioid binding characteristics of fentanyl and buprenorphine, and to investigate some of the differences between them. Buprenorphine showed slow receptor association (30 min), but with high affinity to multiple sites from which dissociation was very slow (Tₜ₀ = 166 min) and incomplete (50% binding after 1 h). This contrasted with the receptor binding of fentanyl, which achieved rapid equilibrium (within 10 min) and dissociated equally rapidly (Tₜ₀ = 6.8 min) and completely (100% by 1 h). Competitive displacement showed buprenorphine displacement of fentanyl binding was concentration- and time-dependent over ranges encountered in clinical use, but buprenorphine binding was displaced with only very high concentrations of other opioids. These findings offer pharmacodynamic explanations for the differences in fentanyl and buprenorphine analgesic response profiles and suggest how binding interactions might be applied to therapeutic use.

MATERIALS AND METHODS

Opioid receptor binding assay

Male Wistar rat (250-300 g) forebrain (whole brain minus brainstem and cerebellum) was used as a source of opioid receptors. Rats were decapitated and the tissue rapidly dissected, weighed, placed in 30 volumes of Tris HCl buffer (50 mmol litre⁻¹, pH 7.4 at 20 °C), and homogenized with an Ultra-Turrax superspeed homogenizer. The homogenate was resuspended in 30 volumes of Tris buffer, incubated in a shaking water-bath at 37 °C for 30 min, recentrifuged and resuspended in 50 volumes of Tris...
FENTANYL AND BUPRENORPHINE: RECEPTOR BINDING

buffer. Ligand binding assays were performed as described previously (Villiger and Taylor, 1981, 1982). Tubes contained 1.0 ml of membrane preparation (20 mg wet weight), 20–100 µlitre [3H]buprenorphine (30 Ci mmol⁻¹; Radiochemical Centre, Amersham) or [3H]fentanyl (12.4 Ci mmol⁻¹; Janssen Pharmaceutica, Beerse, Belgium), 20–100 µlitre drug (where applicable) and Tris buffer to give a final incubation volume of 2.0 ml. Following incubation at 25°C for 30 min, bound was separated from free [3H]opioid by rapid filtration through Whatman GF/B glass fibre filters using a Millipore vacuum manifold. The filters were then removed carefully, dried and placed in vials to which 8.0 ml of a toluene–Triton X-100 scintillation cocktail was added, and radioactivity from labelled opioid was estimated by liquid scintillation spectrometry at a counting efficiency of 28–30%.

Non-specific binding was defined as that occurring in the presence of buprenorphine 1.0 µmol litre⁻¹ when [3H]buprenorphine was used, and levorphanol 1.0 µmol litre⁻¹ in [3H]fentanyl binding experiments. Except when saturation isotherms were being constructed, [3H]fentanyl and [3H]buprenorphine binding were assayed at 2 and 1 nmol litre⁻¹ respectively.

RESULTS

Binding affinities and binding sites

Scatchard analysis of [3H]buprenorphine binding suggests that this drug binds with high affinity ($K_D = 1.13$ nmol litre⁻¹) to a large, apparently homogenous number of binding sites (34.0 pmol/g tissue wet weight) (fig. 1). In contrast, [3H]fentanyl binding was characterized by curvilinear Scatchard plots suggesting both high ($K_D = 0.5$ nmol litre⁻¹) and lower ($K_D = 4.3$ nmol litre⁻¹) affinity binding sites. Also, [3H]fentanyl labelled only 24% ($B_{max} = 8.4$ pmol g⁻¹) of the sites labelled by [3H]buprenorphine.

Drug binding kinetics

Binding of [3H]fentanyl is characterized by a comparatively rapid association with the receptor, with equilibrium being achieved by 10 min. [3H]Buprenorphine binding was a slower process, taking 30 min to achieve equilibrium (fig. 2).

Dissociation kinetics were analysed using computerized non-linear regression analysis (Sedman and Wagner, 1974).

![Fig. 1. Scatchard plots of [3H]fentanyl and [3H]buprenorphine binding to homogenates of rat forebrain. Scatchard plots were derived from saturation isotherms which were obtained by assaying binding at increasing concentrations of [3H]ligand (0.25–15 nmol litre⁻¹ for [3H]fentanyl and 0.1–15 nmol litre⁻¹ for [3H]buprenorphine). Values given are the means of four experiments (SD < 15% of the mean at all points).](https://academic.oup.com/bja/article-abstract/57/2/192/248691)

![Fig. 2. Association and dissociation of [3H]fentanyl and [3H]buprenorphine to and from the opioid receptor. Association rates were established by determining specifically bound drug at times from 0 to 30 min. Dissociation rates were determined by adding levorphanol 1 µmol litre⁻¹ (for [3H]fentanyl) or buprenorphine 1 µmol litre⁻¹ (for [3H]buprenorphine) at 30 min and assaying specific binding at the times indicated. Values given represent the means of three experiments (SD < 15% of the mean at all points).](https://academic.oup.com/bja/article-abstract/57/2/192/248691)
Buprenorphine's action on $[^3H]$fentanyl binding

(a) Fentanyl displacement by addition of buprenorphine. After 30 min equilibration, $[^3H]$fentanyl 2 nmol litre$^{-1}$ was equilibrated for 30 min to achieve binding saturation. Buprenorphine 1, 2 or 4 nmol litre$^{-1}$ was then added and specific fentanyl binding assayed at times from 0 to 120 min. Values given are the mean of four experiments (SD < 15% at all points).

(b) Fentanyl binding after membrane pre-incubation with buprenorphine. Simultaneous addition of $[^3H]$fentanyl 2 nmol litre$^{-1}$ and buprenorphine 0.5, 1 or 2 nmol litre$^{-1}$ resulted in 65%, 31% and 20% $[^3H]$fentanyl binding in each of the mixtures, respectively. Pre-incubation of membranes for 45 min with the same buprenorphine solutions enhanced significantly the displacement potency of buprenorphine ($P < 0.01$; paired $t$ test) with displacement of 63%, 88% and 97% of $[^3H]$fentanyl from the receptor sites (i.e., only 37%, 12% and 3% of $[^3H]$fentanyl remained bound) (fig. 4).

(c) Buprenorphine and fentanyl affinities for receptors labelled with $[^3H]$fentanyl. On the basis of these studies, buprenorphine was two to three times more potent than fentanyl in displacing tritiated fentanyl 2.0 nmol litre$^{-1}$ from $\mu$ receptors. The respective molar concentrations giving 50% inhibition of $[^3H]$fentanyl binding were 0.6 ± 0.1 nmol litre$^{-1}$ for buprenorphine and 1.4 ± 0.1 for fentanyl. Calculated on a mass basis, the IC$\textsubscript{50}$ values for buprenorphine and fentanyl then become 281 pg ml$^{-1}$ and 471 pg ml$^{-1}$, respectively, giving a potency ratio of 1.7 for buprenorphine to fentanyl.

DISCUSSION

The distinction between agonist and agonist-antagonist response profiles is not sufficient to explain many of the clinical features of fentanyl and buprenorphine. Buprenorphine has a high lipid solubility (log of the partition coefficient between heptane and 0.1-M phosphate buffer pH 7.4 = 1.78 (Hambrook and Ranee, 1976)), as does fentanyl (log of the partition coefficient between octanol and water at pH 7.4 = 2.98 (Leysen, Gommern and Niemegeers, 1983)), making for ready CNS access, so the slow onset of the action of buprenorphine is attributable probably to pharmacodynamic rather than pharmacokinetic factors. The observed in vitro studies showing that it took 10 min to establish drug–receptor equilibrium with fentanyl, compared with 30 min for buprenorphine could explain, in part, their temporal differences in attaining full analgesic efficacy.

Our results indicate also that buprenorphine binds to four times the number of receptors labelled by fentanyl. This is consistent with the idea that fentanyl has selective affinity for morphine ($\mu$) recep-
ors (Villiger, Ray and Taylor, 1983) while buprenorphine is comparatively non-selective, binding to μ, δ (enkephalin) and κ (benzomorphan) sites (Villiger and Taylor, 1981; Wood, 1982; Villiger, 1984). Analgesia in clinical situations may be mediated by actions at the high affinity μ sites as proposed by Wolozin and Pasternak (1981). Binding of buprenorphine to other opioid receptors may be less important in generating analgesia, but may bring about the observed antagonistic responses, for example actions on the K receptor being antagonistic to μ receptor effects (Sadee, Rosenbaum and Herz, 1982).

Duration of action for most agents is a function of their plasma concentration, response times reflecting their fate in the body as they are distributed, metabolized or excreted. Fentanyl responses parallel plasma concentration (Stoeckel et al., 1982), but this is not so for buprenorphine, the actions of which (6 h or more (Cowan, Lewis and Macfarlane, 1977)) extend well beyond plasma half-life times of 3–5 h (Bullingham, McQuay and Moore, 1983). Although pharmacokinetic factors could account for this in part, the distinction might be explained further by differences observed with in vitro binding. Fentanyl dissociation is both rapid and almost complete, while that for buprenorphine is both very slow and incomplete, 50% of the drug remaining bound 1 h after attempted washout. This sluggish dissociation of buprenorphine from opioid receptors may account for the inability of naloxone to reverse established buprenorphine effects. The IC₅₀ for naloxone displacing [³H]buprenorphine is approximately 100 nmol litre⁻¹ (Villiger and Taylor, 1981), while only 2.5-nmol litre⁻¹ solutions of naloxone are needed to produce 50% inhibition of [³H]fentanyl binding (Villiger, Ray and Taylor, 1983). On this basis, 40 times the dose of naloxone would be needed to reverse comparable concentrations of buprenorphine as opposed to fentanyl receptor binding.

When utilized concurrently, the kinetics of fentanyl dissociation will depend on the dose of buprenorphine introduced and the timing of administration. Based on these in vitro studies, concurrent buprenorphine use at a concentration of 4 nmol litre⁻¹ (or approx. 2 ng ml⁻¹) will effectively displace 90% of bound fentanyl 2 nmol litre⁻¹ or 0.7 ng litre⁻¹ within 2 h. If the buprenorphine precedes fentanyl administration, buprenorphine 2.0 nmol litre⁻¹ can suppress almost completely any subsequent binding of equimolar concentrations of fentanyl. These values are within the ranges of plasma concentrations associated with the use of these drugs in clinical practice. How plasma concentrations relate to concentrations at opioid receptor sites is not known.

Distinctions in receptor binding may potentially explain several aspects of clinical experience with buprenorphine and fentanyl. In particular, their pharmacodynamic differences in in vitro receptor binding offer explanations for temporal differences in onset and duration of action as well as their reversal characteristics with naloxone. Further application in combined sequences may have bearing on the use of buprenorphine for treatment of drug withdrawal effects (Jasinski, 1981) and in the reversal of acute opioid effects, as suggested by De Castro (1979), who proposed buprenorphine as an antagonist to fentanyl anaesthesia.

REFERENCES


