*Peptides*, Vol. 6, Suppl. 3, pp. 33–46, 1985. © Ankho International Inc. Printed in the U.S.A. 0196-9781/85 $3.00 + .00

# Caerulein and its Analogues: Neuropharmacological Properties

G. ZETLER

*Department of Pharmacology, Medical University of Lübeck, D-2400 Lübeck Federal Republic of Germany*

ZETLER, G. *Caerulein and its analogues: Neuropharmacological properties.* PEPTIDES 6: Suppl. 3, 33–46, 1985.—The decapeptide from the frog *Hyla caerulea*, caerulein (caerulein diethylammonium hydrate, ceruletide, CER) is chemically closely related to the C-terminal octapeptide of cholecystokinin (CCK-8). Like CCK-8, CER and some of its analogues produce many behavioural effects in mammals: inhibition of intake of food and water; antinociception; sedation; catalepsy; ptosis, antistereotypic, anticonvulsive and tremorolytic effects; inhibition of self-stimulation. Effects of CER in man comprise sedation, satiety, changes in mood, analgesia and antipsychotic effects. A modulation of central dopaminergic functions appears to be one possible mechanism of CER and its analogues. A common denominator for all effects of CER is, at present, not evident.

Caerulein Cholecystokinin octapeptide CCK-8 Behaviour Food intake Water intake
Sedation Catalepsy Ptosis Tremor Self-stimulation Analgesia Schizophrenia

THE decapeptide from the skin of the Australian frog *Hyla caerulea*, caerulein (ceruletide, caerulein diethylammonium hydrate, CER) [36] has been widely used in clinical practice to stimulate the gallbladder during cholecystography and to restore intestinal motility in cases of postoperative ileus. The motive for studying possible neuropharmacological effects of CER initially arose from results of experiments on the isolated guinea-pig ileum. It was known that CER and the chemically related C-terminal octapeptide of cholecystokinin (CCK-8, see legend to Table 2) stimulated this preparation indirectly by releasing acetylcholine via a primary excitation of the intramural neuronal plexus; this effect was antagonized by morphine [170,171]. No wonder that opioid peptides likewise inhibited the gut-stimulating effect of CER and CCK-8 which, together with other observations, induced the speculation that opioid and CCK-like peptides interacted by exerting allosteric influences via different but functionally linked receptors located at the intramural neurons of the ileum [175]. Theoretically, allosteric interactions may function in "both directions," so that likewise, CCK-like peptides may influence the opioid receptors. So it was thought to be appropriate to study the question of whether CER (and CCK-8) would exert behavioural effects especially on nociception. This proved right [176] and has induced further studies on CER resulting in an impressive neuropharmacological profile shared in nearly all respects by CCK-8 [189,194]. This is fascinating, because CCK-8, the mammalian analogue of CER, is widely distributed in the peripheral and central nervous system of mammals and man and is thought to play a role as neurotransmitter and neuromodulator [10, 31, 35, 112, 163]. In contrast, CER has not been detected in the brain of the rat [161], *Rana temporaria* and *Xenopus laevis* [30], although it seems to be present in the brain of *Rana esculenta* and the teleost, *Gadus morrhua* [95].

## EFFECTS ON ANIMALS

### *Behavioural Effects*

Research in behavioural neuropharmacology concerns the effects of drugs either in animals that did not receive any other treatment, or in animals showing abnormal behaviour as induced by pretreatment with another drug. Accordingly, the behavioural effects of CER can be subdivided as in Table 1. In all paradigms, irrespective of whether drug-dependent or not, CER produced concurrent effects that can be characterized as sedative, damping, reducing and inhibiting spontaneous activities and reactions to stimuli, or influences from inside or outside the animal. As far as studied, CER produced the same effects after central administration as after peripheral injection, which suggests a central site of mechanism of action. Behavioural or neurological abnormalities were not observed in a study which showed that CER has extremely low acute and chronic toxicity in mice, rats, rabbits, and dogs [18].

When compared with appropriate reference drugs in a quantitative way, CER revealed a surprisingly high potency in the mouse (Table 2). This also applies to the rat, where CER was by far more active than haloperidol and morphine in reducing spontaneous locomotor activity [128] and nociception [145], respectively. CCK-8 was generally less potent than CER, however, in many paradigms still superior to the drug in question. The reference drugs of Table 2 belong to different pharmacological groups such as neuroleptics (chlorpromazine, haloperidol), anxiolytics and anti-

TABLE 1

BEHAVIOURAL EFFECTS OF CAERULEIN IN SEVERAL ANIMAL SPECIES AFTER PERIPHERAL AND CENTRAL ADMINISTRATION

| Paradigm | Route of administration | |
|---|---|---|
| | peripheral* | central* |
| A. Not drug dependent | | |
| 1) Inhibition of eating | M: [125, 158, 196]; R: [55, 125, 150, 155]; C: [107]; F: [140]; P: [4]; RB: [66] | R: [155] |
| 2) Inhibition of drinking | M: [197]; R: [150] | |
| 3) Inhibition of motility | M: [106, 176, 191]; R: [99, 106, 128, 145, 165] | R: [77] |
| 4) Inhibition of exploratory rearing | M: [178, 181, 182, 187, 190, 191]; R: [165] | |
| 5) Catalepsy | M: [176, 179, 180, 183, 191] | |
| 6) Ptosis | M: [176, 182, 184, 185, 190, 191] | |
| 7) Antinociception | M: [176, 185, 186]; R: [15,145] | R: [77,78] |
| 8) Inhibition of self-stimulation | R: [29] | |
| 9) Modification of seizures by electroshock | M: [194] | |
| 10) Modification of passive avoidance | R: [165] | R: [165,166] |
| B. Drug dependent | | |
| 1) Potentiation of barbiturates and ethanol | M: [178,183] | R: [86] |
| 2) Inhibition of excitation by amphetamine | M: [106]; R: [103] | R: [86,106] |
| 3) Inhibition of hypolocomotion by apomorphine | | R: [165,166] |
| 4) Inhibition of chemoconvulsions | M: [81, 82, 177, 179, 181, 194]; R: [82] | R: [82] |
| 5) Antagonism of tremors by harmine and ibogaine | M: [187] | |
| 6) Antagonism of stereotypies by apomorphine, amphetamine, and methylphenidate | M: [179, 180, 182, 193] | R: [34] |
| 7) Inhibition of amphetamine-induced circling | | R: [106] |

*The figures indicate reference number (M, mouse; R, rat; C, cat; F, fowl; P, pig; RB, rabbit).

epileptics (diazepam), and analgesics (morphine). Even more drugs not mentioned in Table 2 (e.g., tremorolytics) were used in order to appraise the significance of the effects produced by CER. Obviously, the paradigms listed in Tables 1 and 2 comprise several neuropharmacological features that have to be commented on separately.

*Inhibition of Food and Water Intake*

CER reduced the food intake in many animal species (Table 1). Peripherally administered CER seems to produce satiety (in rats) by acting at a vagally innervated abdominal site [150] and not via a central site of action as assumed previously [155]. Naloxone, a specific opioid antagonist whose inhibitory influence on eating and drinking is well documented [52,115] increased the effects of CER in mice, whereas the eating-stimulant enkephalin analogue, (D-Ala)$^2$(MePhe)$^4$-(Met(O)-ol)$^5$-enkephalin (FK 33-824), antagonized it [196]. A primary effect on intestinal functions was perhaps not the cause of the reduction in eating, because CER doses necessary to inhibit gastric emptying in mice were 10 times larger and naloxone did not modify the stimulation by CER of the intestinal propulsion [143]. As inhibitors of drinking in the mouse, morphine and FK 33-824 were much less potent than CER and their effect was antagonized by naloxone, whereas that of CER was enhanced; FK 33-824 and CER antagonized each other in this paradigm [197]. The differential potency of CER for inhibition of food and water intake (in mice, Table 2) has also been observed in rats [150]. Since as reviewed [22], benzodiazepines such as diazepam stimulate eating and drinking, the effects of CER on ingestive behaviour are neither diazepam-like nor opioid-like.

*Sedation*

The sedative effect of CER in mice and rats manifested itself as a reduction both in motility (ambulatory activity, running on a rotating drum) and in exploratory rearing activity, and also as a potentiation of central depressant drugs (barbiturates, ethanol) or as an antagonism of amphetamine-induced excitation (Table 1). Sedation also occurred in the rat not only after intracerebroventricular (ICV) administration [86,106], but also after microinjection of a few ng into the periaqueductal grey (PAG) and ventromedial thalamus; injection into the caudate or cuneiform nucleus was ineffective in this respect [77]. Furthermore, injection into the nucleus accumbens of the rat neither altered the spontaneous

TABLE 2

POTENCY IN MALE MICE (NMRI STRAIN) OF CERULETIDE (CER) AS COMPARED WITH CHOLECYSTOKININ OCTAPEPTIDE (CCK-8, C-TERMINAL) AND RELEVANT REFERENCE DRUGS

| Effect | CER* | CCK-8† | Reference drug‡ |
|---|---|---|---|
| 1 Inhibition of food intake [196] | 1.5 | 24 | |
| 2 Inhibition of water intake [197] | 5.5 | | |
| 3 Hypothermia [186] | 5 | 79 | 2817 (C) |
| 4 Catalepsy [179,183] | 13 | 184 | *114* (H) |
| 5 Ptosis [176, 179, 182] | *24* | 50 | *1064* (H) |
| 6 Inhibition of rearing [178, 179, 182, 187] | *14* | 112 | *1098* (H)§ |
| 7 Antinociception (hot plate) [176,182] | 24 | 70 | *1640* (M) |
| 8 Antinociception (writhing, acetic acid) [176] | 83 | 790 | 1200 (M) |
| 9 Potentiation of hexobarbital [178,183] | *30* | *190* | *737* (D) |
| 10 Anticonvulsant vs. picrotoxin [179,182] | *194* | 880 | *1600* (D) |
| 11 Anticonvulsant vs. harman [181,183] | *148* | 700 | *165* (D) |
| 12 Anticovulsant vs. thiosemicarbazide [181] | 900 | 4000 | 900 (D) |
| 13 Antistereotypic vs. methylphenidate [179,182] | *170* | 219 | *55* (H) |
| 14 Antistereotypic vs. apomorphine [193] | 16 | 80 | 730 (H) |
| 15 Tremorolytic vs. harmine [187] | 360 | 450 | 3500 (H) |
| 16 Tremorolytic vs. ibogaine [187] | 480 | 2000 | 4200 (H) |

The potency is expressed as $ED_{50}$ (nmol/kg SC). Italicized figures indicate means from 2–3 estimations. References are mentioned with effects.

*Pyr-Gln-Asp-Tyr ($SO_3H$)-Thr-Gly-Trp-Met-Asp-Phe-$NH_2$

†Asp-Tyr ($SO_3H$)-Met-Gly-Trp-Met-Asp-Phe-$NH_2$

‡C, chlorpromazine; D, diazepam; H, haloperidol; M, morphine.

§The respective dose of diazepam is *1490* nmol/kg [178,182].

locomotor activity [58,165] nor modified the dopamine-induced hypermotility [58]. However, it abolished (a) the hypolocomotion caused by very small doses of apomorphine [165] and (b) the excitation by amphetamine [106], when both drugs were likewise injected into this area. The following observations suggested a stimulatory rather than sedative action: when administered ICV to rats, CER elicited wet-dog shakes (a familiar symptom of withdrawal from opioids) and increased the shaking as elicited by thyrotropin releasing hormone (TRH, ICV) or ice-water immersion, and antagonized the suppressive effect of $\beta$-endorphin (ICV) on shaking caused by TRH or ice water [72]; CER (given ICV) induced in rats increased grooming activity [150], an effect likewise exerted by peptides such as bombesin and eledoisin [87] and ACTH [56] but of unknown mechanism and significance.

The rearing-inhibitory potency of CER in mice was (on a molar basis) many times greater than that of sedative reference drugs such as clonazepam, diazepam, haloperidol, and clonidine [178, 190, 191]. This effect of CER was different from that of familiar drugs, since it was not attenuated by methylphenidate (antagonist to haloperidol and clonazepam) and yohimbine or rauwolscine (antagonists to clonidine) [190,191]. Furthermore, the antirearing action of CER (like that of CCK-8 but unlike that of diazepam) was resistant to the selective benzodiazepine antagonist, Ro 15-1788 [184]. Very important with respect to reported effects of CER in schizophrenic patients (see below) is the following finding in rats [103]: CER and haloperidol, when administered subcutaneously (SC) simultaneously as one single treatment, reduced the susceptibility to the stimulatory action of amphetamine for 2 weeks, whereas each compound alone exerted this effect only for 30–60 min [106].

*Anticonvulsant Effects*

CER and its analogues delayed or prevented convulsions induced by the drugs mentioned in Table 2, whereas they were only weak antagonists or inactive against other convulsants such as bicuculline, pentetrazol and strychnine [80–82, 177, 179, 181, 183, 184]. The antagonism of picrotoxin, harman, thiosemicarbazide, and isoniazid [181] suggested a mechanism of action at the level of the "GABA receptor regulator unit" [23, 57, 136]. However, the inactivity of CER against the convulsants, bicuculline and pentetrazol (to which diazepam is a powerful antagonist) and the resistance of the antiharman effect against the diazepam antagonist, Ro 15-1788, separates the anticonvulsant action of CER (and CCK-8) from that of diazepam [184]. CER also delayed the clonic convulsions by 3-mercaptopropionic acid and the psychomotor seizures by kainic acid [194]. The tonic-clonic convulsions caused by maximal electroshock were not prevented by CER, but the latency till clonic seizures and the duration of postictal motor inactivity were prolonged (effects that were not produced by the anticonvulsant benzodiazepine, clonazepam) [194]. CCK-8 likewise reduced convulsions elicited by drugs [80–82, 177, 179, 181, 184] and by electroshock [80].

The finding *in vitro* that CER ($10^{-9}$ M) and CCK-8 ($10^{-7}$ M) facilitate the release of GABA from rat cerebral cortex slices [144], may have some bearing on the anticonvulsant effects described above.

*Tremorolytic Effects*

The antagonism of tremor as elicited in animals by the cholinergic drug, tremorine, is an important indicator of antiparkinson efficacy in man. In this paradigm, CER (and CCK-8) was inactive, however, it reduced or abolished the existing tremors produced in mice by the hallucinogenic indole alkaloids, harmine and ibogaine [187]. In this respect, CER was much more potent than the reference drugs such as haloperidol, clonazepam and 4 drugs well-known for their usefulness in the treatment of human parkinsonism (biperiden, ethopropazine, methixene, trihexyphenidyl). The pharmacological analysis taught that the tremorolytic effect of CER and related peptides was not the result of a general nonspecific depression of central nervous function. According to Table 2, the tremorolytic potency of CER is weak when compared with that in other paradigms. However, it gains weight in view of the fact that (a) the substantia nigra of Parkinsonian patients contains reduced levels of CCK-8 [156] and (b) receptors for CCK-8 (and CER) are decreased in basal ganglia and cortex of patients suffering from another disturbance of motor functions, Huntington's chorea [60].

*Antinociception*

CER and CCK-8, when injected SC into mice, exerted antinociceptive effects in conventional pharmacological trials such as the acetic-acid writhing test [176] and the hot-plate test [176, 182, 185, 188]. The peptides were many times more potent than morphine (see Table 2). Although antagonized by the specific opioid antagonist, naloxone [176], the peptidic antinociception in mice differed from that by morphine, since it was much more resistant to naloxone [185], completely resistant to apomorphine which abolished the effect of morphine [188], nearly unaltered by tolerance to morphine [176], and showed only a very limited additivity with the effect of morphine [185]. In rats too, SC administration of CER produced antinociception in the hot-plate test and especially in the paw-pressure test [145]. In this study, CER was 477 times more potent than morphine (molar basis), however, its effect was potentiated by a dose of naloxone that antagonized the effect of morphine. The antinociceptive effect of CER (in mice) was greater on jumping up than on paw licking in the hot-plate test [188] and (in rats) greater in the paw-pressure test (endpoint vocalization or escape) than in the hot-plate test (paw licking) [145]; this may reflect a preferential affinity of CER to neuronal mechanisms important for the reaction to pain (suffering) as opposed to the perception (recognition) of the stimulus [51]. The inefficiency of systemically administered CER in the tail-flick test in mice [176] and rats [99,145] likewise points to a predominance of supraspinal mechanisms. When the drop in blood pressure following renal pelvis distension in anesthetized rats was used as a model of renal colic, CER (SC) prevented this reaction in doses 400–500 times smaller than those of morphine (molar basis); naloxone abolished the effect of morphine but not that of CER [15].

As to the site of antinociceptive action, it is important that the tail-flick response (to radiant heat) of the rat was inhibited (for more than 2 hr) by a few pmoles of CER injected into either the PAG or the caudate nucleus, the ventromedial thalamus and the spinal subarachnoid space, but not into the cuneiform nucleus [77]. Morphine showed the same behaviour, however, its potency (on a molar basis) was less than that of CER by a factor of 4500 (PAG) or 6900 (intrathecal). In this study, CCK-8 was 9–10 times less potent than CER, and naloxone (0.5 mg/kg, intraperitoneally, IP) abolished the effect of both peptides. When the inhibition of the C-fibre-evoked ascending nociceptive activity in the spinal cord (instead of the tail-flick response) of the rat was studied, CER and CCK-8 (administered intrathecally) were ineffective but morphine was inhibitory [33]. When the site of injection was the PAG, the ascending nociceptive activity was depressed by morphine, facilitated by naloxone as well as CCK-8, and either depressed or facilitated by CER [78].

Obviously, the antinociceptive effect of CER is not well understood at present. Actually, CER and CCK-8, when injected ICV into rats in doses 45–78 times larger than those found effective after intracerebral and intrathecal administration [77], did not produce an effect in the hot-plate test but antagonized the antinociception by $\beta$-endorphin [74]. A similar observation in rats is the antagonism of morphine-induced antinociception (tail-flick test) by CCK-8 [39] and CER [99]. These findings together raise the pivotal question of whether CER modifies the pain threshold in man (see below).

*Hypothermia*

When administered SC to mice, CER rapidly and dose-dependently lowered the body temperature for 30 to 60 min [181, 184, 186, 187]. On a molar basis, CER was more potent than CCK-8, chlorpromazine and haloperidol by factors of 17, 612 and 1260, respectively. According to the results of pharmacological experimentation, the CER-induced hypothermia is different from that by haloperidol and cannot be the cause of other peptide effects such as antinociception or inhibition of convulsions and tremors [181, 186, 187]. The hypothermic mechanism of CER is unknown, however, it may be central since (a) centrally acting drugs such as phenytoin (anticonvulsant) and desipramine (antidepressant) are antagonists [186], and (b) CER lowers the body temperature of the rat after SC [145] as well as ICV administration [85]; the latter applies also to CCK-8 [84, 113, 114], which has recently been shown to inhibit heat production and facilitate heat loss by mechanisms located in the anterior hypothalamus [146].

*Catalepsy*

During the state of catalepsy, animals are motionless and maintain awkward positions without, however, muscular weakness. Catalepsy is a typical neuroleptic effect and different from sedation such as produced by diazepam-like tranquilizing drugs. CER induces catalepsy in mice and is in this respect 9 times more potent than haloperidol (Table 2) [176, 179, 180, 183, 191]. The cataleptogenic action of CER, like that of haloperidol, was attenuated by atropine and phenytoin and enhanced by the GABA transaminase inhibitor, aminooxyacetic acid [179]. Nevertheless, catalepsy by CER was different from that by haloperidol, because it was naloxone-resistant, antagonized by muscimol, and increased by bicuculline and picrotoxin, whereas that by haloperidol was muscimol-resistant but antagonized by naloxone, bicuculline and picrotoxin; furthermore, the cataleptogenic effects of CER and haloperidol were not additive but rather antagonized each other [180]. Another type of antagonistic interaction is the shortening by CER (ICV) of the $\beta$-endorphin-induced catalepsy in the rat [71]. In this respect, CER was more potent than CCK-8 and the opioid antagonist, naltrexone, by a factor of 19 and 11, respectively.

*Ptosis*

A typical effect of neuroleptics in mice and rats besides catalepsy is ptosis, that is, a centrally mediated narrowing of the eye fissure [62]. This ptosis is not merely a feature of an overall sedation, since it is not caused by depressant doses of diazepam, clonazepam, and clonidine [179, 184, 187, 191]. Hence, the high ptosis-producing potency of CER (and CCK-8) in mice (see Tables 1 and 2) is an important neuropharmacological property which also deserves to be studied in rats. Up to now, there exist only secondary observations indicating that neither CER [106] nor CCK-8 [165] elicit ptosis in the rat.

The CER-induced ptosis (in mice) was not modified by naloxone [176,185], Ro 15-1788 [184], and yohimbine [191], which suggested that neither receptors for opioids and for benzodiazepines nor $\alpha_2$-adrenergic autoreceptors are involved in the primary mechanism resulting in ptosis. Central stimulants such as methylphenidate and picrotoxin exerted only a very weak antiptotic effect that did not differentiate between CCK-peptidergic and haloperidol-induced ptosis

[190]. However, apomorphine in very small doses antagonized the ptosis by CER and CCK-8 but was less active against the haloperidol-induced ptosis by a factor of 27. Low-dose haloperidol abolished the antiptotic action of apomorphine versus the peptides, which suggested the presynaptic dopamine autoreceptors were involved in the antiptotic effect of apomorphine [190].

*Antistereotypic Effects*

Another typical neuroleptic effect in laboratory animals is the antagonism of stereotypies (sniffing, licking, gnawing) produced by either direct dopaminergic stimulants (such as apomorphine) or indirect stimulants (such as methylphenidate and the amphetamines). In mice, the fully developed stereotypic gnawing (of the wires of the cage) due to methylphenidate was rapidly reduced or abolished by CER and related peptides [179, 180, 182]. The antistereotypic effect of CER was about three times weaker than that of haloperidol (Table 2), however, it had the same quality of that of the neuroleptic, that is, it was antagonized by muscimol and diazepam [180]. Nevertheless, CER and haloperidol must owe their antistereotypic efficacy to different mechanisms, because their antignawing effects were not additive but rather antagonized each other [180]. The direct dopaminergic stimulant, apomorphine, only induces stereotypic gnawing in mice that have received a sensitizing pretreatment by either scopolamine [61,141] or teflutixol [111,142] and clonidine [195]. In these paradigms, CER had an antistereotypic potency which was markedly higher than that against methylphenidate and well above that of haloperidol [193,195]; the values presented in Table 2 refer to scopolamine-pretreated mice. There was again a contrast between the peptides and the neuroleptics: the antistereotypic potency of trifluoperazine and teflutixol was greater after clonidine than after scopolamine and teflutixol, whereas this was not so with CER and CCK-8. Furthermore, the antistereotypic effect of trifluoperazine and teflutixol was not additive to that of CER [195].

In rats, the stereotypic activity elicited by a peripheral dose of apomorphine was enhanced by the ICV infusion of a small dose of CER (0.22 nmol) and antagonized by a larger dose (0.6 nmol); in the same study, CER (1.2 nmol ICV) inhibited the potentiation of the apomorphine-induced behaviour by dibuturyl-cAMP [34]. However, when injected into the rat nucleus caudatus, CER did not alter the stereotypies caused by apomorphine or amphetamine, both administered via the same route [106,165].

*Influence on Circling and Climbing*

Rats injected unilaterally with 6-hydroxydopamine into the medial forebrain bundle and one week later challenged by apomorphine (SC) show circling movements contralateral to the lesion, but an ipsilateral circling after challenge by amphetamine (SC); only the amphetamine-induced circling was inhibited by systemically administered CER [106]. Cage-climbing of mice is another effect caused by stimulation of postsynaptic dopamine receptors, e.g., by apomorphine, and all clinically effective neuroleptics have anticlimbing potency [174]. However, CER was very weak, although not completely inactive in this paradigm [106,190].

*Effects on Passive Avoidance Behaviour*

When given SC 1 hr before the retention test, CER (and CCK-8) facilitated in rats passive avoidance, that is, the shock-induced hesitancy to enter a dark compartment [165,166]; this effect of CER was still present 24 hr later. However, after direct injection into the nucleus accumbens, CER (and CCK-8) attenuated passive avoidance (also after 24 hr). These effects were reminiscent of those by $\gamma$-type endorphins which, in contrast, only decreased this behaviour irrespective of the route of administration [54,92]. It was concluded that the CCK-like peptides (a) attenuate the passive avoidance behaviour by a mechanism similar to that of the $\gamma$-type endorphins, and (b) facilitate it by interaction with other brain structures in or outside the nucleus accumbens [165]. Experiments using CCK-8 had concordant results in this paradigm [44–46, 79]. In another type of conditioned behaviour (active avoidance), CCK-8 produced or facilitated extinction [21, 41, 43, 45, 48].

*Inhibition of Self-Stimulation Behaviour*

For more than 25 years it has been known that an experimental animal with electrodes implanted in certain subcortical places will stimulate itself regularly for long times; in animals trained to self-stimulate at stable rates, neuroleptics (e.g., chlorpromazine) suppress self-stimulation, whereas tranquilizing drugs (e.g., meprobamate) fail to do so [126]. In such experiments in rats, CER and CCK-8 produced inhibition (lasting 20 min) and antagonized the stimulatory action of d-amphetamine [29]. The neuroleptics, chlorpromazine and haloperidol, were likewise inhibitory, however, only the effect of haloperidol was potentiated by the peptides. The lowest effective dose (nmol/kg IP) tested was 2 for CER and 18 for CCK-8. On a molar basis, chlorpromazine and haloperidol were less active than CER by factors of 376 and 18, respectively. An inhibitory effect of CCK-8 on self-stimulation has also been observed in rabbits and rats [11, 37, 47, 91, 162].

*CER in the Drug-Discrimination Paradigm*

Intracerebral self-stimulation of rats was also used as reinforcing reward for correct discrimination between treatment (IP) by a drug (CCK-8) or by vehicle [29]. After the conditioned behaviour (lever pressing) had been established, the rats were able to discriminate between CCK-8 and a novel drug. Trained animals did not differentiate CCK-8 from CER and haloperidol, but clearly separated CCK-8 from apomorphine, amphetamine and unsulphated CCK-8; intermediate results were obtained with chlorpromazine and diazepam.

*Electrophysiological Effects*

CER, like CCK-8, applied (by pressure ejection from micropipettes) into the immediate vicinity of rat pyramidal cells (CA1 region of the hippocampus), was excitatory and produced depolarization, an increase in excitability and a decrease in membrane input resistance [32,89]. A stimulatory action of CER was also seen when the peptide (4 $\mu$g/kg) was injected intravenously (IV) into rats and the firing of dopaminergic neurons lying in the CCK-rich zona compacta of the substantia nigra was recorded by intracellular micropipettes [65]. Furthermore, the treatment by CER produced a shift to the left of apomorphine's dose-response curve for the inhibition of firing of these mesencephalic neurones; this suggested that CER induced a supersensitivity of dopamine autoreceptors. Electrophysiological experiments using mi-

croiontophoresis have been performed with CCK-8 more often than with CER [14, 19, 32, 75, 129, 135, 138]. It is desirable to study the question of whether CER exerts the same stimulatory effects as produced by CCK-8 on many, but by no means all, central neurons tested.

*Biochemical Effects*

Preincubation of rat striatal homogenates with CER reduced both the affinity and density of binding sites for [$^3$H]-dopamine; CER was at least 4-fold less active than CCK-8 [119]. In contrast to CCK-8, CER did not displace [$^3$H]-dopamine from its specific binding sites in rat striatal homogenates.

The neuroleptic dopamine antagonist, [$^3$H]-spiperone, binds to brain dopamine receptors of the D2 type. When tested *in vitro*, CER failed to modify the specific binding of [$^3$H]-spiperone to rat striatal homogenates [104]. In contrast, when CER was SC injected into mice, [$^3$H]-spiperone binding in subcortex and in frontal cortex was reduced [168]. However, after a pretreatment with haloperidol for 14 days, CER enhanced [$^3$H]-spiperone binding. The latter finding is in accord with that of a CCK-8-induced increase in affinity of striatal dopamine receptors [53]. Hence, these peptides seem to modulate the interaction of neuroleptics with their specific binding sites at dopaminergic systems. Furthermore, it appears that results obtained *in vitro* are not fully concordant with those seen *in vivo*, and that there may also be species differences between mouse and rat. Systemically administered CER (100–800 μg/kg IP) dose-dependently reduced, in the rat striatum (but not in the nucleus accumbens and 6 other areas), both homovanillic acid (HVA) and 3,4-dihydroxyphenylacetic acid (DOPAC), that is, it attenuated the dopamine turnover [104]; CCK-8 (ICV) exerted the same effect on the rat striatum [102]. After repeated treatment with CER (200 μg/kg IP, daily for 5 days) the acute decreases in striatal HVA and DOPAC disappeared, whereas HVA was enhanced in the nucleus accumbens. It was concluded that CER produces different effects on nigrostriatal and mesolimbic dopaminergic systems [104]. However, these doses of CER are by far larger than those necessary to produce a behavioural effect. Therefore, it is important that CER in a very low, although sedative dose (0.25 μg/kg IP), did not modify the dopamine metabolism in rat striatum and nucleus accumbens; 50 μg/kg reduced DOPAC in both areas [128]. CER and CCK-8 likewise modify rat cortical systems. Low IP doses (up to 5 μg/kg) of CER or CCK-8 stimulated acetylcholine release from the cortex (not antagonized by naloxone), whilst larger doses (10 and 20 μg/kg) reduced it (antagonized by naloxone) [99]. *In vitro*, CER ($10^{-9}$ M) and CCK-8 ($10^{-7}$) facilitated the potassium-evoked release of [$^{14}$C]-GABA from tissue slices of rat parietal cortex [144]. However, CER ($10^{-9}$ – $10^{-4}$ M) did not modulate the [$^{14}$C]-dopamine release from rat nucleus accumbens slices [58].

CCK-like peptides have also been observed to influence the pituitary-adrenal axis. In rats, CER (3 ng and more per animal, IP) produced a marked increase in the plasma corticosterone level; since ICV doses of CER up to 50 ng/rat were ineffective in this respect and since either subdiaphragmatic vagotomy or hypophysectomy reduced or abolished the effect, it was concluded that this action of CER might be mediated via the vagus nerve to the hypothalamo-hypophyseal axis [73]. The same results have been obtained with CCK-8 [70,132]. However, at a large ICV dose (1 μg/rat), CER markedly elevated the plasma corticosterone level, while CCK-8 did not [69]; the latter finding contrasts with a positive report [42]. On the other hand, ICV administered CCK-8 (but not CER) dose-dependently suppressed the elevation of plasma corticosterone level induced by an ICV dose of vasoactive intestinal polypeptide (VIP, 1 μg/rat) [69]. The plasma concentration of ACTH in the rat was decreased by ICV administration of CER (2.5–10 ng/animal) but increased by an IV dose of 0.4 μg/kg [137]; pretreatment by atropine methylbromide (that does not pass the blood-brain barrier) prevented the latter effect, suggesting a peripheral site of primary action. However, CCK-8 was capable of releasing ACTH from rat pituitary halves *in vitro* [132].

*Structure-Activity Relationships*

Ten analogues of CER have been studied in mice concerning the effects mentioned in Table 2 (except for effects No. 1, 2, 8, and 12). As has been reviewed [189,194] the following features are obvious: desulphation, deamidation, and shortening the peptide chain to a C-terminal pentapeptide reduces or abolishes the potency in all respects. Small chemical alterations (such as intercalation of $CH_2$ into the backbone or exchange of Met by Nle) leave some effects unchanged while reducing and enhancing others. Suffice it to mention that the heptapeptide, Boc-Nle$^4$-CER-(4–10), was equal to CER concerning effects No. 10, 11, and 15 (Table 2) but as an antistereotypic vs. methylphenidate 3 times more potent and much less potent than CER in all other effects (including inhibition of apomorphine-induced stereotypies, No. 14). The divergent influences of chemical structure on neuropharmacological effects suggest that some of the multiple actions of CER (Table 2) may be independent rather than epiphenomena secondary to a main effect such as hypothermia or general sedation [186,187]. It may be added that the behavioural effects of CER analogues in the mouse did not run parallel to smooth-muscle stimulatory potencies *in vitro* (mouse gallbladder, ileum, colon) [192].

## EFFECTS IN MAN

Pharmacological findings in man are extremely important because they are touchstones for concepts emerging from studies in laboratory animals or systems *in vitro*. According to a study in 4 individuals treated with intramuscular (IM) dose of 0.3 or 0.6 μg/kg CER, plasma levels of CCK-like immunoreactivity peaked between 20 and 30 min following administration and returned to basal levels by 60 min (Tamminga, Littman, Alphs, Chase, Thaker and Wagman, personal communication).

*Analgesia*

An analgesic effect of CER was first observed in patients suffering from biliary colic [5, 6, 127, 129]. Effective doses were 0.5–1 ng/kg (IV) or 15–75 ng kg (IM); analgesia occurred within a few min and was long-lasting. It was first assumed that the relaxing effect of CER on the sphincter Oddi [1,12] and the resulting decrease in intrabiliary pressure was the main reason for this analgetic effect. However, approximately the same low doses of CER alleviated or abolished cancer pain [130]; rest pain in patients with arterial insufficiency of the lower extremity [9] and pain from renal colics [5,96]—the latter in spite of the fact that CER does not produce relaxation of the ureter or urinary bladder *in vitro* [3,38]. In healthy subjects, CER (either 4.2 and 8.4 μg/hr/70 kg IV, or 5 and 10 μg/70 kg IM) dose-dependently increased

TABLE 3

RESULTS OF CLINICAL TRIALS ON CERULETIDE IN CHRONIC SCHIZOPHRENIC PATIENTS

| Study No. | Type | No. of patients | Patients on neuroleptics? | CER dose (μg/kg)* | No. of doses† | Treatment effective?‡ | Duration of effect (weeks) after end of treatment | References |
|---|---|---|---|---|---|---|---|---|
| 1 | Open | 20 | yes | 0.13; 0.26 | 1 | yes (16) | >3 | [117,118] |
| 2 | Open | 58 | yes | 0.3 or 0.6 | 1 or 2§ | yes (20) | 1–2 | [68] |
| 3 | Open | 295 | yes | 0.6 | 2 (7) | yes (106) | unknown | [93] |
| 4 | Open | 15 | yes | 0.8–2.0 | several | yes (11) | unknown | [157] |
| 5 | Open | 20 | yes | 0.6 | 1 | yes (13) | >4 | [116] |
| 6 | Single-blind | 6 | yes | 0.6 | 2 (7) | yes (3) | >5 | [166,167] |
| 7 | Double-blind | 86 | yes | 0.6 | 2 (7) | doubtful | unknown | [93] |
| 8 | Double-blind | 10 | yes | 0.3 | 3 (7) | no | | [2] |
| 9 | Double-blind | 8 | yes | 0.3–0.6 | 8 (0.5) | no | | [63,64] |
| 10 | Double-blind | 9 | no | 0.04; 0.4 | 2 (16) | no | | [98] |
| 11 | Double-blind | 15 | yes | 0.6 | 7 | yes (8) | >2 | ¶ |
| 12 | Double-blind | unknown | no | 0.6 | 5 (1) | # | | [124] |
| 13 | Double-blind | 5 | no | 0.3–1.5 | 10 (1) | no | | ** |
| 14 | Double-blind | 6 | yes | 0.3 | 1 | no | | ** |

*The route of administration was IM except for study No. 8 (IV).

†In parentheses, time between doses (days).

‡In parentheses, number of patients showing a marked improvement.

§Each patient received 1 or 2 doses one week or more apart: 5 patients received 3 or more doses. Of the 20 improved cases, 6 received a single dose and 14 repeated doses. However, the improvement was evident already with the initial dose in all these 14 cases receiving repeated doses.

¶Verhoeven and Van Ree (personal communication); treatment schedule: 7 injections on day 1, 10, 12, 15, 17, 19 and 22.

#Effect "seemingly in favour of ceruletide."

**Tamminga, C. A., R. L. Littman, L. D. Alphs, T. N. Chase, G. K. Thaker and A. M. Wagman (personal communication).

threshold and tolerance to electrically and threshold to thermally induced pain; however, the effect of CER was inferior to that of the opioid drug, pentazocine, and resistant to naloxone [153,154].

*Antipsychotic Effects*

Nearly all studies performed up to now concern chronic schizoprenics under persistent treatment with neuroleptics to which they were resistant (Table 3); however, only 8 out of 14 studies were double-blind. As to the efficacy of the treatment, an equivocal picture emerges from Table 3. So much more important is the double-blind study (No. 11) which led to virtually the same conclusions as the other positive studies. Remarkable features are the very small doses and the long-lasting amelioration following only one injection of CER. Some of the positive studies pointed out that not all psychotic symptoms were improved equally well. According to study No. 6, CER affected positive symptoms (hostility, suspiciousness, uncooperativeness; hallucinations, conceptual disorganization, unusual thought content, grandiosity) more than negative symptoms (anxiety, depression, anergia). Studies No. 1 and 2 found the same influence on positive symptoms, however, in addition, also an improvement of mood and of negative symptoms such as anxiety, motor retardation, emotional withdrawl, tension and mannerism. Weak side effects were reported in 7 studies (No. 1, 2, 4, 6, 7, 9, 11) and occurred in approximately 25% of the patients; they lasted only a few minutes (never longer than 45 min) and consisted of one or more of the following symptoms: epigastric discomfort, nausea, borborygmus, abdominal cramping, vomiting, diarrhea. Neither extrapyramidal symptoms nor exacerbation of existing extrapyramidal symptoms were observed [68,116]. Regarding the equivocal nature of the results of clinical trials with CER (Table 3), it is worth mentioning that therapeutic efficacy (with quite CER-like features) in neuroleptic-resistant schizophrenics has also been assigned to CCK-33 and CCK-8 [13, 120–122]. A beneficial effect of CER (0.6 μg/kg IM, 4 times, once weekly) has been observed in a long-term marihuana smoker suffering from a neuroleptic-resistant amotivational syndrome [172]. CER was inefficient in parkinsonian patients receiving L-dopa [17].

*Sedation*

In the studies of Table 3 sedation was not observed as a side effect. In normal subjects, however, CER produced sedation and fatigue [63, 152–154] without significantly altering critical flicker-fusion threshold and EEG [152,154] or impairing memory [63]; out of 24 subjects, 8 complained of dizziness, 8 of euphoria, 9 of drunkenness, and 3 reported hallucination [154]. When compared with placebo in 14 patients suffering from psychogenic headache, CER did not alter the subjective pain evaluation, but it modified mood by reducing anxiety and scepticism and augmenting elation, cheerfulness and egotism [7]. Cortical evoked potential testing (using visual stimuli) was carried out in 4 schizophrenic patients and did not reveal an effect of an IM dose of 0.3 μg/kg CER (Tamminga, Littman, Alphs, Chase, Thaker and Wagman, personal communication).

*Sleep*

Six healthy male subjects (20–24 years of age) received

either placebo or CER (0.6 μg/kg IM) at 23:00 and were observed by means of polysomnograms till 06:30 [173]. CER increased the percentage of REM sleep but did not alter many other features, including the score of subjective estimation of sleep. It may be of interest that CCK-8, in the rat, did not modify the normal sleep but normalized the paradoxical sleep when reduced by chloramphenicol [76,133].

*Endocrine Effects*

It is well known that drugs having antipsychotic potency in man stimulate the secretion of prolactin by blocking postsynaptic dopamine receptors within the hypothalamo-pituitary axis. CER did not modify the basal prolactin secretion [7, 9, 94, 131]; CCK-8 likewise was inactive in this respect [94] whereas CCK-33 stimulated the prolactin release [123]. CER did not affect the secretion of growth hormone [7, 9, 131], luteinizing hormone and parathyroid hormone [131]. The concentration of ACTH was enhanced by CER only in the cerebrospinal fluid (CSF) [7] but not in blood [9]. Slightly increased β-endorphin levels in plasma and CSF were observed following an IV infusion of CER, 2 ng/kg/min for 15 min [6, 7, 9].

*Appetite*

In a double-blind study performed in non-obese individuals, CER (1 or 2 ng/kg/min, infused IV for 60 min) reduced both self-ratings of hunger and total food consumption [152]; side effects were sedation and abdominal discomfort as described above. A preceding study using CCK-8 (4.6 or 9.2 ng/kg/min, infused IV for 15 min) described very similar results, however, without CER-like side effects [151].

## DISCUSSION

The far-reaching pharmacological similarity of CER with CCK-8 can be explained by the close chemical relationship (see legend to Table 3) and the high affinity of both peptides to the same specific receptor [59, 64, 90, 164]. As to behavioural effects in mice (Table 2), CER, generally, was more potent than CCK-8, in spite of (a) being of equal stimulatory activity on mouse smooth muscles (gallbladder and intestines) *in vitro* [192] and (b) having equal [60,164] or even somewhat less affinity to the receptor than CCK-8 [67,90]. One reason for the superiority of CER *in vivo* may be its greater resistance to enzymatic destruction [28]. However, this cannot explain why the potency ratio of CER and CCK-8 differed with the paradigm: CCK-8 was virtually equi-active with CER in producing ptosis and antagonizing the harmine-induced tremor, whilst it was less antistereotypic, cataleptogenic and rearing-inhibitory by factors of 4–5, 13 and 17, respectively [194]. There are more inconsistencies: *in vivo*, CER and CCK-8 modified the release of acetylcholine from cat cerebral cortex with equal strength [99], whereas CER was 100 times more potent than CCK-8 in enhancing GABA release from rat cortex *in vitro* [144]; CER was not only less active than CCK-8 in modifying [$^3$H]-dopamine binding by rat striatal homogenates, but was even (in contrast to CCK-8) devoid of [$^3$H]-dopamine displacing efficacy in the same system [119]; when injected into the nucleus accumbens of the rat, CCK-8 decreased locomotor activity and antagonized the apomorphine-induced hyperactivity, whereas CER was ineffective in both respects [165]; in mice, yohimbine and rauwolscine weakly antagonized the rearing-inhibitory effect of CCK-8 but not that of CER [191]. Marked differences between CER and CCK-8 have also been observed concerning (a) the ascending nociceptive activity in the spinal cord of the rat [78] and (b) the VIP-induced elevation of plasma corticosterone level in the rat [69]. Taken together, these differences in pharmacological effects between both peptides suggest that CER does not merely mimic CCK-8 but rather is a CCK-like peptide with its own functional features.

The multifarious effects of CER may not have a common denominator. This can be concluded from the unequal influence of changes in chemical structure on pharmacological efficacy. It is noteworthy that smooth-muscle stimulatory potency *in vitro* and behavioural potency *in vivo* do not run parallel [192]. It also appears that CER exerts several independent neuropharmacological effects. This view is supported by the results of some drug-peptide interactions (in the mouse) such as: (a) naloxone antagonizes some though not all [15] antinociceptive effects [77, 176, 185] without influencing sedation and ptosis [176,185] and (b) apomorphine antagonizes ptosis but not sedation and antinociception [188,190].

The antagonism of some antinociceptive effects of CER by naloxone does not permit the straightforward conclusion that the peptide interacts with opioid receptors [15, 77, 185]. It may be stressed here that naloxone failed to antagonize CER in a study of analgesia in man [154] and in a rat model of renal colic [15]. Naloxone even enhanced in rats the antinociceptive (and hypothermic) effects of CER, which suggests that (in the rat) an endogenous opioid peptide inhibits the action of CCK peptides [145]. Furthermore, CER neither lost its antinociceptive efficacy in morphine-tolerant mice [176] nor substituted morphine in dependent mice or induced physical dependence [49]; in the field-stimulated mouse vas deferens, CER did not exert a morphine-like inhibition either [175]. The finding in man that CER increased the plasma level of β-endorphin-like immunoreactivity (ELI) has led to the speculation that a release of endorphin was the cause of CER-induced analgesia [6, 7, 9]. In fact, CCK-8 has been shown to release in rats ELI from the anterior pituitary gland [105]. However, animal studies did not support the notion that released β-endorphin produces analgesia [97,109] and the inhibition by CCK-like peptides including CER of the degradation of Leu-enkephalin by rat synaptic membranes did not reveal the same order of strength as the antinociceptive effect *in vivo* [27].

A question of general importance concerns the ability of peptides such as CER and CCK-8 to penetrate the blood-brain barrier and to affect the central nervous system directly, rather than via a primary peripheral mechanism. This problem has been discussed repeatedly [83, 108, 147, 194]. It is true that numerous behavioural effects mentioned in Table 1 have been observed after peripheral as well as after central administration. However, pharmacokinetic results were equivocal since after IV administration of [$^{35}$S]-CER, radioactivity occurred in the rat brain [160] but not in the mouse spinal cord and brain [110]; an immunohistochemical study failed to detect CER in the rat brain after IM injection [161], whilst serum levels reached peak values within 5 min and were elevated for 30 to 60 min (following doses of 1 or 5 μg/kg, respectively) [159]. Following ICV administration to rats, CER occurred within 5 min in many cortical and subcortical neurons, reached a maximum after 15 min and disappeared within 30 min [161]. It has been discussed with regard to CCK-8 that circulating peptides could reach access to neural sites without penetrating the blood-brain barrier

[108]. Furthermore, it is possible that very minute amounts of CER, that still escape detection, pass into the brain and suffice to bring about an effect. After all, concentrations of CCK-8 as low as $10^{-14}$ and $10^{-11}$ M reduced dopamine release from slices of cat caudate nucleus [100].

Nevertheless, it is also appropriate to consider a peripheral mechanism, because some behavioural effects of CCK-8 such as satiety and sedation were abolished by either vagotomy [24,149] or lesion of the nucleus tractus solitarii (NTS) [25]. Indeed, vagotomy has been shown to abolish three effects in rats of peripherally administered CER, namely: the inhibition of food intake [150], the increase in plasma corticosterone level [73], and the depression of the crossed extensor reflex [88]. However, the stimulatory action of CCK-8 (given IV) on single units of the substantia nigra (zona compacta) was resistant to acute and chronic vagotomy and reduced by only 50% after lesions of the NTS [148]. Furthermore, the stimulatory effect of CCK-8 (IP) on acetylcholine release from cerebral cortex [99] is not abolished by bilateral vagotomy (Magnani, Mantovani and Pepeu, personal communication). At present time it must remain an open question as to whether *all* neuropharmacological actions of peripherally administered CCK-like peptides are altered by lesions of the vagus or the NTS, and whether or not this also applies to effects produced by peptides after ICV or intracerebral injection.

The reviewed behavioural and biochemical actions of CER (and CCK-8) would permit a prediction about an antipsychotic efficacy in man. The latter would also be plausible because schizophrenic patients show (1) a reduced CCK-like immunoreactivity in the temporal cortex and in the hippocampus and amygdala [16,50] as well as in the cerebrospinal fluid [169] which was not confirmed by Tamminga, Littman, Alphs, Chase, Thaker and Wagman (personal communication), and (2) a decreased high affinity CCK binding in hippocampus and frontal cortex [40]. The inconsistency in the results of clinical studies with CER and CCK-8 (cf. Table 3 and comment) may partially be caused by different approaches (open vs. double-blind studies). However, one has to consider that "schizophrenia is a multifactorial cluster of illnesses" [134] and cannot be explained by a single neurochemical lesion [26,134]. Hence, it is possible that only certain subgroups of schizophrenics respond to CER or CCK-8 [64]. Since CCK-like peptides comprise neuroleptic and anticonvulsive potency (classic neuroleptic drugs are proconvulsive!), one can speculate that a responding subgroup may consist of patients that have a schizophrenia-like psychosis as a result of a temporal lobe epilepsy [26].

In many of the studies in Table 3 the patients were under continuous treatment with neuroleptic drugs. The type of this neuroleptic treatment may also modify the peptide effect. This possibility emerges from the observation (in mice) that the antistereotypic effects of CER and haloperidol were not additive when the stereotypy was methylphenidate-induced [179], but added when the stereotypy was caused by apomorphine [195]. However, CER was not additive with other neuroleptics (trifluoperazine and teflutixol) in the latter paradigm. The long duration of the antipsychotic effect of CER and CCK-8 is a very surprising feature, which is not reconcilable with the peptidic nature of these compounds. However, a similar observation has been made in rats [103]: amphetamine-induced hyperactivity was attenuated for 15 days by one single combined treatment with CER (0.04 mg/kg SC) and haloperidol (0.1 mg/kg SC); when given alone, each of these treatments exerted an effect for 30–60 min [106].

The powerful tremorolytic and anticonvulsive effects of CER and related peptides suggest clinical trials too on patients not responding well to conventional treatment of motor dysregulations. Analogues of CER likewise deserve attention. Examples are $Nle^8$-CER and $Nle^8$-CER-(4–10) with increased antistereotypic potency (vs. methylphenidate) and in the latter peptide, enhanced selectivity. Other candidates are Met $(O)^8$-CER and $(\beta\text{-ASP})^9$-CER, both having selective anti-methylphenidate and strongly reduced smooth-muscle stimulatory potency [194].

After submission of this paper, the author became aware of the publication of A. Dumbrille-Ross and Ph. Seeman (Dopamine receptor elevation by cholecystokinin. *Peptides* **5**: 1207–1212, 1984) whose results are important in two respects discussed above: (1) after IP administration to rats of either CCK-8 or CER (50 $\mu$g/kg each), nanomolar levels of CCK-like substances were measurable in the striatum and the accumbens up to 30 min after injection; (2) following one single dose of either peptide, $^3$H-spiperone binding in both the striatum and accumbens was increased for a period up to two weeks. These findings support those of a penetration of CER through the blood-brain barrier [160] and a 14-days antagonism of amphetamine (combined treatment by CER plus haloperidol) [103].

ACKNOWLEDGEMENTS

The author thanks the following colleagues for sending him manuscripts in press or informing him about results to be published in the near future: J. de Belleroche (London), Ph. de Witte (Louvain-La-Neuve), T. Moroji (Tokyo), G. Pepeu (Florence), C. A. Tamminga (Baltimore), J. M. van Ree (Utrecht). The excellent assistance of Ms. Gerda Mundhenke in the preparation of this manuscript is very much appreciated.

## REFERENCES

1. Agosti, A., P. Mantovani and L. Mori. Action of caerulein and related substances on the sphincter of oddi. *Naunyn Schmiedebergs Arch Pharmacol* **268**: 114–118, 1971.
2. Albus, M., M. Akenheil, D. Munch and D. Naber. Ceruletide: a new drug for treatment of schizophrenic patients? *Arch Gen Psychiatry* **41**: 528, 1984.
3. Angelucci, E., G. Falconieri-Erspamer and L. Negri. Bioassay of vasopressin on the rabbit isolated urinary bladder. *J Pharm Pharmacol* **26**: 193–196, 1974.
4. Anika, S. M., T. R. Houpt and K. A. Houpt. Cholecystokinin and satiety in pigs. *Am J Physiol* **240**: R310–R318, 1981.
5. Basso, N., M. Bagarani, D. Gizzonio, A. Basoli, F. Fiocca, C. De Paolis, C. Praga and V. Speranza. Analgesic effect of ceruletide (CRL) in biliary and renal colic. *Gastroenterology* **80**: 1105, 1981.
6. Basso, N., A. Materia, V. D'Intinosante, E. Bianchi and V. Speranza. Beta-endorphin-like immunoreactivity following scalar doses of ceruletide (CRL) in human plasma. *Regul Pept* **1**: S8, 1980.

7. Basso, N., A. Materia, V. D' Intinosante, A. Ginaldi, V. Pona, P. Reilly, S. Ruggeri and M. Fioravanti. Effect of ceruletide on pituitary-hypothalamic peptides and on emotion in man. *Peptides* **2**: Suppl 2, 71–75, 1981.
8. Basso, N., A. Materia, F. Fiocca, M. Percoco, M. Bagarani, E. Lezoche and V. Speranza. Treatment of biliary colic with ceruletide. *Gastroenterology* **78**: 1137, 1980.
9. Basso, N., V. Pona, V. D'Intinosante, F. Fiocca, C. Desantis, M. Bagarani, D. Gizzonio and F. Ponzielli. Effect of ceruletide on rest pain in patients with arterial insufficiency of the lower extremity. *Eur J Clin Pharmacol* **22**: 531–533, 1982.
10. Beinfeld, M. C. Cholecystokinin in the central nervous system. *Neuropeptides* **3**: 411–428, 1983.
11. Belyi, V. P. and M. Fekete. Comparative study of influence of hormonally active and inactive forms of cholecystokinin octapeptide on self-stimulation reaction in rabbits. *J Higher Nerv Activity IP Pavlov* **32**: 344–346, 1982.
12. Bertaccini, G., G. De Caro, R. Endean, V. Erspamer and M. Impicciatore. The actions of caerulein on the smooth muscle of the gastrointestinal tract and the gall bladder. *Br J Pharmacol* **34**: 291–310, 1968.
13. Bloom, D. M., N. P. V. Nair and G. Schwartz. CCK-8 in the treatment of chronic schizophrenia. *Psychopharmacol Bull* **19**: 361–363, 1983.
14. Bradwejn, J. and C. De Montigny. Benzodiazepines antagonize cholecystokinin-induced activation of rat hippocampal neurones. *Nature* **312**: 363–364, 1984.
15. Brasch, H. and G. Zetler. Caerulein and morphine in a model of visceral pain. Effects on the hypotensive response to renal pelvis distension in the rat. *Naunyn Schmiedebergs Arch Pharmacol* **319**: 161–167, 1982.
16. Carruthers, B., D. Dawbarn, M. De Quidt, P. C. Emson, J. Hunter and G. P. Reynolds. Changes in neuropeptide content of amygdala in schizophrenia. *Br J Pharmacol* **81**: 190P, 1984.
17. Chase, T. N., P. Barone, G. Bruno, S. L. Cohen, T. Juncos, M. Knight, S. Ruggeri, L. Steardo and C. A. Tamminga. Cholecystokinin-mediated synaptic function and the treatment of neuropsychiatric disease. *Ann NY Acad Sci* **448**: 553–561, 1985.
18. Chieli, T., C. Bertazzoli, G. Ferni, I. Dell'Oro, C. Capella and E. Solcia. Experimental toxicology of caerulein. *Toxicol Appl Pharmacol* **23**: 480–491, 1972.
19. Chiodo, L. A. and B. S. Bunney. Proglumide: selective antagonism of excitatory effects of cholecystokinin in central nervous system. *Science* **219**: 1449–1451, 1983.
20. Cohen, S. L., M. Knight, C. A. Tamminga and T. N. Chase. Cholecystokinin-octapeptide effects on conditioned-avoidance behavior, stereotypy and catalepsy. *Eur J Pharmacol* **83**: 213–222, 1982.
21. Cohen, S. L., M. Knight, C. A. Tamminga and T. N. Chase. Tolerance to the anti-avoidance properties of cholecystokinin-octapeptide. *Peptides* **4**: 67–70, 1983.
22. Cooper, S. J. Benzodiazepine-opiate antagonist interactions in relation to anxiety and appetite. *Trends Pharmacol Sci* **4**: 456–458, 1983.
23. Costa, E. and A. Guidotti, Molecular mechanisms in the receptor action of benzodiazepines. *Annu Rev Pharmacol Toxicol* **19**: 531–545, 1979.
24. Crawley, J. N., S. E. Hays and S. M. Paul. Vagotomy abolishes the inhibitory effects of cholecystokinin on rat exploratory behaviors. *Eur J Pharmacol* **73**: 379–380, 1981.
25. Crawley, J. N. and J. S. Schwaber. Nucleus tractus solitarius lesions block the behavioral actions of cholecystokinin. *Peptides* **4**: 743–748, 1983.
26. Csernansky, J. G., C. A. Holman and L. E. Hollister. Variability and the dopamine hypothesis of schizophrenia. *Schizophr Bull* **9**: 325–328, 1983.
27. Deschodt-Lanckman, M. and N. D. Bui. Cholecystokinin octa- and tetrapeptide degradation by synaptic membranes. *Peptides* **2**: Suppl 2, 113–118, 1981.
28. Deschodt-Lanckman, M., N. D. Bui, M. Noyer and J. Christophe. Degradation of cholecystokinin-like peptides by a crude rat brain synaptosomal fraction: A study by high pressure liquid chromatography. *Regul Pept* **2**: 15–30, 1981.
29. De Witte, Ph., E. Swanet, M. Gewiss, S. Goldman, B. Roques and J. J. Vanderhaeghen. Psychopharmacological profile of CCK using the self-stimulation and the drug discrimination paradigms. *Ann NY Acad Sci* **448**: 470–487, 1985.
30. Dimaline, R. Is caerulein amphibian CCK? *Peptides* **4**: 457–462, 1983.
31. Dockray, G. J. The physiology of cholecystokinin in brain and gut. *Br Med Bull* **38**: 253–258, 1982.
32. Dodd, J. and J. S. Kelly. The actions of cholecystokinin and related peptides on pyramidal neurones of the mammalian hippocampus. *Brain Res* **205**: 337–350, 1981.
33. Doi, T. and I. Jurna. Analgesic effect of intrathecal morphine demonstrated in ascending nociceptive activity in the rat spinal cord and ineffectiveness of caerulein and cholecystokinin octapeptide. *Brain Res* **234**: 399–407, 1982.
34. Ellinwood, E. H., W. J. K. Rockwell and N. Wagoner. A caerulein-sensitive potentiation of the behavioral effects of apomorphine by dibutyryl-cAMP. *Pharmacol Biochem Behav* **19**: 969–971, 1983.
35. Emson, P. C. and P. D. Marley. Cholecystokinin and vasoactive intestinal polypeptide. In: *Handbook of Psychopharmacology, Vol 16*, edited by L. L. Iversen, S. D. Iversen and S. H. Snyder. New York: Plenum Publishing Corp., 1983, pp. 255–306.
36. Erspamer, V. and P. Melchiorri. Actions of amphibian skin peptides on the central nervous system and the anterior pituitary. In: *Neuroendocrine Perspectives, Vol 2*, edited by E. E. Müller and R. M. MacLeod. Amsterdam: Elsevier Science Publishers B. V., 1983, pp. 37–106.
37. Ettenberg, A. and G. F. Koob. Different effects of cholecystokinin and satiety on lateral hypothalamic self-stimulation. *Physiol Behav* **32**: 127–130, 1984.
38. Falconieri-Erspamer, G., L. Negri and D. Piccinelli. The use of preparations of urinary bladder smooth muscle for bioassay of and discrimination between polypeptides. *Naunyn Schmiedebergs Arch Pharmacol* **279**: 61–74, 1973.
39. Faris, P. L., B. R. Komisaruk, L. R. Watkins and D. J. Mayer. Evidence for the neuropeptide cholecystokinin as an antagonist of opiate analgesia. *Science* **219**: 310–312, 1983.
40. Farmery, S. M., F. Owen, M. Poulter and T. J. Crow. Reduced high affinity cholecystokinin binding in hippocampus and frontal cortex of schizophrenic patients. *Life Sci* **36**: 473–478, 1985.
41. Fekete, M., M. Balazs and G. Telegdy. Effects of intracerebroventricularly administered cholecystokinin antisera on active avoidance and conditioned feeding behaviour of rats. *Acta Physiol Acad Sci Hung* **60**: 243–249, 1983.
42. Fekete, M., M. Bokor, B. Penke, K. Kovacs and G. Telegdy. Effects of cholecystokinin octapeptide and its fragments on brain monoamines and plasma corticosterone. *Neurochem Int* **3**: 165–169, 1981.
43. Fekete, M., M. Bokor, B. Penke and G. Telegdy. Effects of cholecystokinin octapeptide sulphate ester and unsulphated cholecystokinin octapeptide on active avoidance behaviour in rats. *Acta Physiol Acad Sci Hung* **60**: 57–64, 1983.
44. Fekete, M., T. Kadar, B. Penke and G. Telegdy. Modulation of passive avoidance behaviour by cholecystokinin octapeptides in rats. *Neuropeptides* **1**: 301–307, 1981.
45. Fekete, M., A. Lengyel, B. Hegedüs, B. Penke, M. Zarandy, G. K. Toth and G. Telegdy. Further analysis of the effects of cholecystokinin octapeptide on avoidance behaviour in rats. *Eur J Pharmacol* **98**: 79–91, 1984.
46. Fekete, M., B. Penke and G. Telegdy. Structure-activity studies concerning effect of cholecystokinin octapeptide on passive avoidance behaviour of rats. *Acta Physiol Acad Sci Hung* **58**: 197–200, 1981.
47. Fekete, M., A. Rentzsch, H. Schwarzberg and G. Telegdy. Effect of cholecystokinin on self-stimulation behavior in rats. *Eur J Pharmacol* **91**: 77–82, 1983.

48. Fekete, M., A. Szabo, M. Balazs, B. Penke and G. Telegdy. Effects of intraventricular administration of cholecystokinin octapeptide sulfate ester and unsulfated cholecystokinin octapeptide on active avoidance and conditioned feeding behaviour of rats. *Acta Physiol Acad Sci Hung* **58:** 39–45, 1981.
49. Ferrari, M., E. Oggioni and A. C. Rossi. Some further pharmacological data on ceruletide. *Int Symp Brain-Gut Axis*. Florence, June 29–July 1. Abstracts, p. 60, 1981.
50. Ferrier, I. N., G. W. Roberts, T. J. Crow, E. C. Johnstone, D. G. C. Owens, Y. C. Lee, D. O'Shaughnessy, T. E. Adrian, J. M. Polak and S. R. Bloom. Reduced cholecystokinin-like and somatostatin-like immunoreactivity in limbic lobe is associated with negative symptoms in schizophrenia. *Life Sci* **33:** 475–482, 1983.
51. Frederickson, R. C. A. Enkephalin pentapeptides. Review of current evidence for a physiological role in vertebrate neurotransmission. *Life Sci* **21:** 23–42, 1977.
52. Fuller, L. M. The pharmacology of drinking behavior. *Pharmacol Ther* **24:** 179–206, 1984.
53. Fuxe, K., L. F. Agnati, F. Benfenati, M. Cimmino, S. Algeri, T. Hökfelt and V. Mutt. Modulation by cholecystokinins of $^3$H-spiroperidol binding in rat striatum: evidence for increased affinity and reduction in the number of binding sites. *Acta Physiol Scand* **113:** 567–569, 1981.
54. Gaffori, O. and D. De Wied. Effects of des-Tyr$^1$-γ-endorphin and des-enkephalin-γ-endorphin on active and passive avoidance behavior of rats: a dose-response relationship study. *Eur J Pharmacol* **85:** 115–119, 1982.
55. Gibbs, J., R. C. Young and G. P. Smith. Cholecystokinin decreases food intake in rats. *J Comp Physiol Psychol* **84:** 488–495, 1973.
56. Gispen, W. H. and R. L. Isaacson. ACTH-induced excessive grooming in the rat. *Pharmacol Ther* **12:** 209–246, 1981.
57. Haefely, W., A. Kulcsar, H. Möhler, L. Pieri, P. Polc and R. Schaffner. Possible involvement of GABA in the central actions of benzodiazepines. *Adv Biochem Psychopharmacol* **14:** 131–151, 1975.
58. Hamilton, M., M. J. Sheehan, J. De Belleroche and L. J. Herberg. The cholecystokinin analogue, caerulein, does not modulate release or dopamine-induced locomotor activity in the nucleus accumbens of rat. *Neurosci Lett* **44:** 77–82, 1984.
59. Hays, S. E., M. C. Beinfeld, R. T. Jensen, F. K. Goodwin and S. M. Paul. Demonstration of a putative receptor site for cholecystokinin in rat brain. *Neuropeptides* **1:** 53–62, 1980.
60. Hays, S. E. and S. M. Paul. CCK receptors and human neurological disease. *Life Sci* **31:** 319–322, 1982.
61. Hedley, L. R. and M. B. Wallach. Potentiation of apomorphine-induced gnawing in mice. *Prog Neuropsychopharmacol Biol Psychiatry* **7:** 47–56, 1983.
62. Hill, R. T. and D. H. Tedeschi. Animal testing and screening procedures in evaluating psychotropic drugs. In: *An Introduction to Psychopharmacology*, edited by R. H. Rech and K. E. Moore. New York: Raven Press, 1971, pp. 237–288.
63. Hommer, D. W., D. Pickar, J. N. Crawley, H. Weingartner and S. M. Paul. The effects of CCK-like peptides in schizophrenics and normal human subjects. *Ann NY Acad Sci* **448:** 542–552, 1985.
64. Hommer, D. W., D. Pickar, A. Roy, P. Ninan, J. Boronow and S. M. Paul. The effects of ceruletide in schizophrenia. *Arch Gen Psychiatry* **41:** 617–622, 1984.
65. Hommer, D. W. and L. R. Skirboll. Cholecystokinin-like peptides potentiate apomorphine-induced inhibition of dopamine neurons. *Eur J Pharmacol* **91:** 151–152, 1983.
66. Houpt, T. R., S. M. Anika and N. C. Wolff. Satiety effects of cholecystokinin and caerulein in rabbits. *Am J Physiol* **235:** R23–R28, 1978.
67. Innis, R. B. and S. H. Snyder. Distinct cholecystokinin receptors in brain and pancreas. *Proc Natl Acad Sci USA* **77:** 6917–6921, 1980.
68. Itoh, H., S. Tanoue, G. Yagy, M. Tateyama, M. Kamisada, Y. Fujii, M. Takamiya and S. Nakajima. Clinical study of the psychotropic effects of caerulein—an open clinical trial in chronic schizophrenic patients. *Keio J Med* **31:** 71–95, 1982.
69. Itoh, S. and R. Hirota. Inhibitory effect of cholecystokinin octapeptide on vasoactive intestinal peptide-induced stimulation of adrenocortical secretion. *Jpn J Physiol* **33:** 301–304, 1983.
70. Itoh, S., R. Hirota and G. Katsuura. Effect of cholecystokinin octapeptide and vasoactive intestinal polypeptide on adrenocortical secretion in the rat. *Jpn J Physiol* **32:** 553–560, 1982.
71. Itoh, S. and G. Katsuura. Suppressive effect of cholecystokinin and its related peptides on β-endorphin-induced catalepsy in rats. *Eur J Pharmacol* **74:** 381–384, 1981.
72. Itoh, S. and G. Katsuura. Effects of β-endorphin, thyrotropin-releasing hormone and cholecystokinin on body shaking behavior in rats. *Jpn J Physiol* **32:** 677–675, 1982.
73. Itoh, S., G. Katsuura, R. Hirota and K. Odaguchi. Effect of caerulein on plasma corticosterone concentration in the rat. *Life Sci* **27:** 2205–2210, 1980.
74. Itoh, S., G. Katsuura and Y. Maeda. Caerulein and cholecystokinin suppress β-endorphin-induced analgesia in the rat. *Eur J Pharmacol* **80:** 421–425, 1982.
75. Jeftinija, S., V. Miletic and M. Randic. Cholecystokinin octapeptide excites dorsal horn neurons both in vivo and in vitro. *Brain Res* **213:** 231–236, 1981.
76. Jouvet, M. Neuromediateurs et facteurs hypnogenes. *Rev Neurol (Paris)* **140:** 389–400, 1984.
77. Jurna, I. and G. Zetler. Antinociceptive effect of centrally administered caerulein and cholecystokinin octapeptide (CCK-8). *Eur J Pharmacol* **73:** 323–331, 1981.
78. Jurna, I. and G. Zetler. Effects on ascending nociceptive activity in the rat spinal cord produced by cholecystokinin octapeptide, ceruletide and morphine injected into the periaqueductal grey matter. *Ann NY Acad Sci* **448:** 609–612, 1985.
79. Kadar, T., M. Fekete and G. Telegdy. Modulation of passive avoidance behaviour of rats by intracerebroventricular administration of cholecystokinin octapeptide sulfate ester and non-sulfated cholecystokinin octapeptide. *Acta Physiol Acad Sci Hung* **58:** 269–274, 1982.
80. Kadar, T., A. Pesti, B. Penke and G. Telegdy. Inhibition of seizures induced by picrotoxin and electroshock by cholecystokinin octapeptide and their fragments in rats after intracerebroventricular administration. *Neuropharmacology:* **23:** 955–961, 1984.
81. Kadar, T., A. Pesti, B. Penke, G. Toth, M. Zarandi and G. Telegdy. Structure-activity and dose-effect relationships of the antagonism of picrotoxin-induced seizures by cholecystokinin, fragments and analogues of cholecystokinin in mice. *Neuropharmacology* **22:** 1223–1229, 1983.
82. Kadar, T., A. Pesti, G. Toth, B. Penke and G. Telegdy. Effects of cholecystokinin octapeptides and their fragments on seizures induced by different convulsive drugs. In: *Neuropeptides and Psychosomatic Processes*, edited by E. Endröczi, L. Angelucci, U. Scapagnini and D. De Wied. Budapest: Akademiai Kiado Budapest, 1983, pp. 231–238.
83. Kastin, A. J., R. D. Olson, A. V. Schally and D. H. Coy. CNS effects of peripherally administered brain peptides. *Life Sci* **25:** 401–414, 1979.
84. Katsuura, G., R. Hirota and S. Itoh. Cholecystokinin-induced hypothermia in the rat. *Experientia* **37:** 60, 1981.
85. Katsuura, G. and S. Itoh. Effect of cholecystokinin octapeptide on body temperature in the rat. *Jpn J Physiol* **31:** 849–858, 1981.
86. Katsuura, G. and S. Itoh. Sedative action of cholecystokinin octapeptide on behavioral excitation by thyrotropin releasing hormone and methamphetamine in the rat. *Jpn J Physiol* **32:** 83–91, 1982.
87. Katz, R. Grooming elicited by intracerebroventricular bombesin and eledoisin in the mouse. *Neuropharmacology* **19:** 143–146, 1980.

88. Kawasaki, K., M. Kodama and A. Matsushita. Caerulein, a cholecystokinin-related peptide, depresses somatic function via the vagal afferent system. *Life Sci* **33**: 1045–1050, 1983.
89. Kelly, J. S. and J. Dodd. Cholecystokinin and gastrin as transmitters in the mammalian central nervous system. *Adv Biochem Psychopharmacol* **28**: 133–144, 1981.
90. Knight, M., C. A. Tamminga, L. Steardo, M. E. Beck, P. Barone and T. N. Chase. Cholecystokinin-octapeptide fragments: binding to brain cholecystokinin receptors. *Eur J Pharmacol* **105**: 49–55, 1984.
91. Kornblith, C. L., G. N. Ervin and R. A. King. Hypothalamic and locus coeruleus self-stimulation are decreased by cholecystokinin. *Physiol Behav* **21**: 1037–1041, 1978.
92. Kovacs, G. L., G. Telegdy and D. De Wied. Selective attenuation of passive avoidance behaviour by microinjection of $\beta$-LPH$_{62-77}$ and $\beta$-LPH$_{66-77}$ into the nucleus accumbens in rats. *Neuropharmacology* **21**: 451–454, 1982.
93. Kudo, Y. The effect of ceruletide on chronic schizophrenia. *Jpn J Clin Psychiatr* **12**: 703–710, 1983 (quoted after Moroji [116]).
94. Lal, S., N. P. V. Nair, H. Eugenio, J. Thavundayil, E. Lizondo, P. L. Wood, P. Etienne and H. Guyda. Neuroendocrine evaluation of CCK-peptides on dopaminergic function in man. *Prog Neuropsychopharmacol Biol Psychiatry* **7**: 537–544, 1983.
95. Larsson, L.-I. and J. F. Rehfeld. Evidence for a common evolutionary origin of gastrin and cholecystokinin. *Nature* **269**: 335–338, 1977.
96. Libert, M., W. Stekke, R. Askenasi and C. C. Schulman. Utilisation de la ceruleine comme antalgique dans le traitment de la colique nephretique. *Acta Therapeutica* **8**: 145–150, 1982.
97. Lim, A. T., M. Wallace, T. P. Oei, S. Gibson, N. Romas, W. Pappas, J. Clements and J. W. Funder. Foot shock analgesia. Lack of correlation with pituitary and plasma immunoreactive-$\beta$-endorphin. *Neuroendocrinology* **35**: 236–241, 1982.
98. Lotstra, F., P. Verbanck, J. Mendlewicz and J. J. Vanderhaeghen. No evidence of antipsychotic effect of caerulein in schizophrenic patients free of neuroleptics: a double-blind cross-over study. *Biol Psychol* **19**: 877–882, 1984.
99. Magnani, M., P. Mantovani and G. Pepeu. Effect of cholecystokinin octapeptide and ceruletide on release of acetylcholine from cerebral cortex of the rat in vivo. *Neuropharmacology* **23**: 1305–1309, 1984.
100. Markstein, R. and T. Hökfelt. Effect of cholecystokinin-octapeptide on dopamine released from slices of cat caudate nucleus. *J Neurosci* **4**: 570–575, 1984.
101. Martini, A., L. Bonollo, C. De Paolis and M. Lavezzari. Ceruletide: a potent analgesic peptide. *Abstr of the 8th Int Congress of Pharmacology*. Tokyo, July 19–24, p. 775, 1981.
102. Mashal, R. D., F. Owen, J. F. W. Deakin and M. Poulter. The effects of cholecystokinin on dopaminergic mechanisms in rat striatum. *Brain Res* **277**: 375–376, 1983.
103. Matsubara, K. and A. Matsushita. Long-lasting reduction of amphetamine-induced hyperactivity in rats after combined administration of caerulein with haloperidol. *Eur J Pharmacol* **101**: 157–158, 1984.
104. Matsumoto, T., T. Nakahara, H. Uchimura, M. Hirano, H. Yokoo, K. Nakamura and K. Oomagari. Effect of systemically administered caerulein on dopamine metabolism in rat brain. *Brain Res* **324**: 195–199, 1984.
105. Matsumura, M., A. Yamanoi, S. Yamamoto and S. Saito. In vivo and in vitro effects of cholecystokinin octapeptide on the release of $\beta$-endorphin-like immunoreactivity. *Neuroendocrinology* **36**: 443–448, 1983.
106. Matsushita, A. and S. Itoh. Pharmacological properties of caerulein, a CCK-8 analogous peptide. In: *World Psychiatric Association Regional Symposium*, edited by H. Ohashi, K. Nakayama, M. Saito and B. Saletu. Tokyo: The Japanese Society of Psychiatry and Neurology, 1982, pp. 170–173.
107. Mendel, V. E., R. A. L. Sturdevant and J. Elashoff. The effects of caerulein on food intake in the cat. *Physiol Behav* **24**: 629–632, 1980.
108. Mendelson, S. D. and B. B. Gorzalka. Cholecystokinin-octapeptide produces inhibition of lordosis in the female rat. *Pharmacol Biochem Behav* **21**: 755–760, 1984.
109. Millan, M. J., R. Przewlocki, M. Jerlicz, Ch. Gramsch, V. Höllt and A. Herz. Stress-induced release of brain and pituitary $\beta$-endorphin: major role of endorphins in generation of hypothermia, not analgesia. *Brain Res* **208**: 325–338, 1981.
110. Miyhoshi, K. and T. Tokiwa. Distribution of $^{35}$S-caerulein (F.I. 6934) in mice. *Pharmacometrics* **10**: 465–470, 1975.
111. Møller-Nielsen, I. and A. V. Christensen. Long term effects of neuroleptic drugs. *J Pharmacol* **6**: 277–282, 1975.
112. Morley, J. E. The ascent of cholecystokinin (CCK) from gut to brain. *Life Sci* **30**: 479–493, 1982.
113. Morley, J. E. and A. S. Levine. Intraventricular cholecystokinin-octapeptide (CCK-8) produces hyperglycemia and hypothermia. *Clin Res* **28**: 721A, 1980.
114. Morley, J. E., A. S. Levine and S. Lindblad. Intraventricular cholecystokinin-octapeptide produces hypothermia in rats. *Eur J Pharmacol* **74**: 249–251, 1981.
115. Morley, J. E., A. S. Levine, G. K. Yim and M. T. Lowy. Opioid modulation of appetite. *Neurosci Biobehav Rev* **7**: 227–256, 1983.
116. Moroji, T., K. Itoh and K. Itoh. Antipsychotic effects of ceruletide in chronic schizophrenia. An appraisal of the long-term, intermittent medication of ceruletide in chronic schizophrenia. *Ann NY Acad Sci* **448**: 518–534, 1985.
117. Moroji, T., N. Watanabe, N. Aoki and S. Itoh. Antipsychotic effects of ceruletide (caerulein) on chronic schizophrenia. *Arch Gen Psychiatry* **39**: 485–486, 1982.
118. Moroji, T., N. Watanabe, N. Aoki and S. Itoh. Antipsychotic effects of caerulein, a decapeptide chemically related to cholecystokinin octapeptide, on schizophrenia. *Int Pharmacopsychol* **17**: 255–273, 1983.
119. Murphy, R. B. and D. I. Schuster. Modulation of [$^3$H]-dopamine binding by cholecystokinin octapeptide (CCK-8). *Peptides* **3**: 539–544, 1982.
120. Nair, N. P. V., D. M. Bloom, G. Debonnel, G. Schwartz and S. Mosticyan. Cholecystokinin-octapeptide in chronic schizophrenia: a double-blind placebo-controlled study. *Prog Neuropsychopharmacol Biol Psychiatry* **8**: 711–714, 1985.
121. Nair, N. P. V., D. Bloom and J. N. Nestoros. Cholecystokinin appears to have antipsychotic properties. *Prog Neuropsychopharmacol Biol Psychiatry* **6**: 509–512, 1982.
122. Nair, N. P. V., D. M. Bloom, J. N. Nestoros and G. Schwartz. Therapeutic efficacy of cholecystokinin in neuroleptic-resistant schizophrenic subjects. *Psychopharmacol Bull* **19**: 134–136, 1983.
123. Nair, N. P. V., S. Lal, J. X. Thavundayil, P. L. Wood, P. Etienne and H. Guyda. CCK-33 antagonizes apomorphine-induced growth hormone secretion and increases basal prolactin levels in man. *Neuropeptides* **4**: 281–292, 1984.
124. Nakamura, G., M. Nobuyama and Y. Kudo. Antipsychotic effect of ceruletide as a sole agent. *Abstr 14th C.I.N.P. Congress*, Florence, June 19–23, p. 8, 1984.
125. Newmark, S. R. Caerulein inhibits food intake in the rat and the mouse. *Nutr Rep Int* **23**: 151–154, 1981.
126. Olds, J. Studies of neuropharmacologicals by electrical and chemical manipulation of the brain in animals with chronically implanted electrodes. In: *Neuropharmacology. Proceedings of the First International Congress of Neuro-Pharmacology*, edited by P. B. Bradley, P. Deniker and C. Radouco-Thomas. Amsterdam: Elsevier Publishing Company, 1959, pp. 20–32.
127. Pardo, A., Celotti, F. and C. De Paolis. Ceruletide analgesia in biliary colic. *Clin Pharmacol Ther* **36**: 510–514, 1984.
128. Pasinetti, G., S. Govoni, F. Battaini, P. Locatelli, E. Faccini and M. Trabucchi. Caerulein peripheral injection—a study on the correlation with dopaminergic metabolism. *Pharmacol Res Commun* **16**: 1175–1182, 1984.
129. Phillis, J. W. and J. R. Kirkpatrick. The actions of motilin, luteinizing hormone releasing hormone, cholecystokinin, somatostatin, vasoactive intestinal peptide, and other peptides on rat cerebral cortical neurons. *Can J Physiol Pharmacol* **58**: 612–623, 1980.

130. Piazza, E., M. Brambilla, M. T. Cattaneo, A. Martini and M. A. Pacciarini. Analgesic activity of ceruletide in cancer patients. In: *International Symposium on Brain-Gut Axis: The New Frontier*, Florence, June 29 30–July 1, 1981. Abstracts, edited by N. Basso, E. Lezoche, V. Speranza and J. H. Walsh. Florence, 1981, p. 146.
131. Pontiroli, A. E., M. Alberetto, L. Restelli and A. Facchinetti. Effect of bombesin and ceruletide on prolactin, growth hormone, luteinizing hormone, and parathyroid hormone release in normal human males. *J Clin Endocrinol Metab* **51:** 1303–1305, 1980.
132. Porter, J. R. and L. D. Sander. The effect of cholecystokinin octapeptide on pituitary-adrenal hormone secretion. *Regul Pept* **2:** 245–252, 1981.
133. Riou, F., R. Cespuglio and M. Jouvet. Endogenous peptides and sleep in the rat: III. The hypnogenic properties of vasoactive intestinal polypeptide. *Neuropeptides* **2:** 265–277, 1982.
134. Rodnight, R. Schizophrenia: some current neurochemical approaches. *J Neurochem* **41:** 12–21, 1983.
135. Rogawski, M. A. Cholecystokinin octapeptide: Effects on the excitability of cultured spinal neurons. *Peptides* **3:** 545–552, 1982.
136. Rommelspacher, H., C. Nanz, H. Borbe, K. J. Fehske, W. E. Müller and U. Wollert. 1-Methyl-β-carboline (harmane), a potent endogenous inhibitor of benzodiazepine receptor binding. *Naunyn Schmiedebergs Arch Pharmacol* **314:** 97–100, 1980.
137. Saija, A., E. Rapisarda, I. Padovano, A. Trovato and G. Costa. Ceruletide modifies plasma ACTH levels in the rat. *Acta Pharmacol Toxicol (Copenh)* **53:** Suppl I, 193, 1985.
138. Salt, T. E. and R. G. Hill. The effects of C-terminal fragments of cholecystokinin on the firing of single neurones in the caudal trigeminal nucleus of the rat. *Neuropeptides* **2:** 301–306, 1982.
139. Santamaria, A., G. Lucani, M. Montorsi and C. Praga. Action de la ceruleine sur la colique hepatique. *Nouv Presse Med* **8:** 2482, 1979.
140. Savory, C. J. and M. J. Gentle. Intravenous injections of cholecystokinin and caerulein suppress food intake in domestic fowls. *Experientia* **36:** 1191–1192, 1980.
141. Scheel-Krüger, J. Central effects of anticholinergic drugs measured by the apomorphine gnawing test in mice. *Acta Pharmacol Toxicol (Copenh)* **28:** 1–16, 1970.
142. Scheel-Krüger, J. and A. V. Christensen. The role of gamma-aminobutyric acid in acute and chronic neuroleptic action. *Adv Biochem Psychopharmacol* **24:** 233–243, 1980.
143. Scheufler, E. and G. Zetler. Interactions of caerulein and morphine on gastrointestinal propulsion in the mouse. *Naunyn Schmiedebergs Arch Pharmacol* **318:** 66–69, 1981.
144. Sheehan, M. J. and J. de Belleroche. Facilitation of gaba release by cholecystokinin and caerulein in rat cerebral cortex. *Neuropeptides* **3:** 429–434, 1983.
145. Sheehan, M. J. and J. de Belleroche. Naloxone potentiation of the antinociceptive action of caerulein. *Ann NY Acad Sci* **448:** 642–647, 1985.
146. Shian, L. R. and M. T. Lin. Effects of cholecystokinin octapeptide on thermoregulatory responses and hypothalamic neuronal activity in the rat. *Naunyn Schmiedebergs Arch Pharmacol* **328:** 363–367, 1985.
147. Skirboll, L. R., A. A. Grace, D. W. Hommer, J. Rehfeld, M. Goldstein, T. Hökfelt and B. S. Bunney. Peptide-monamine coexistence: studies of the actions of cholecystokinin-like peptide on the electrical activity of midbrain dopamine neurons. *Neuroscience* **6:** 2111–2124, 1981.
148. Skirboll, L. R. and D. W. Hommer. Electrophysiological studies of the role of cholecystokinin in the substantia nigra and its interactions with dopamine. *Ann NY Acad Sci* **448:** 275–282, 1985.
149. Smith, G. P., C. Jerome, B. J. Cushin, R. Eterno and K. J. Simansky. Abdominal vagotomy blocks the satiety effect of cholecystokinin in the rat. *Science* **213:** 1036–1037, 1981.
150. Smith, G. P., C. Jerome, P. Kulkosky, K. J. Simansky. Ceruletide acts in the abdomen, not in the brain, to produce satiety. *Peptides* **5:** 1149–1158, 1984.
151. Stacher, G., H. Steinringer, G. Schmierer, C. Schneider and S. Winklehner. Cholecystokinin octapeptide decreases intake of solid food in man. *Peptides* **3:** 133–136, 1982.
152. Stacher, G., H. Steinringer, G. Schmierer, C. Schneider and S. Winklehner. Ceruletide decreases food intake in non-obese man. *Peptides* **3:** 607–612, 1982.
153. Stacher, G., H. Steinringer, G. Schmierer, C. Schneider, S. Winklehner, G. Mittelbach, C. de Paolis and C. Praga. Ceruletide increases dose dependently both jejunal motor activity and threshold and tolerance to experimentally induced pain in healthy man. *Gut* **25:** 513–519, 1984.
154. Stacher, G., H. Steinringer, G. Schmierer, S. Winklehner and C. Schneider. Ceruletide increases threshold and tolerance to experimentally induced pain in healthy man. *Peptides* **3:** 955–962, 1983.
155. Stern, J. J., C. A. Cudillo and J. Kruper. Ventromedial hypothalamus and short-term feeding suppression by caerulein in male rats. *J Comp Physiol Psychol* **90:** 484–490, 1976.
156. Studler, J. M., F. Javoy-Agid, F. Cesselin, J. C. Legrand and Y. Agid. CCK-8-immunoreactivity distribution in human brain: selective decrease in the substantia nigra from parkinsonian patients. *Brain Res* **243:** 176–179, 1982.
157. Tanaka, S., T. Nakayama and Y. Kudo. A dose-range study of ceruletide in schizophrenia. *Abstr 14th C.I.N.P. Congress*, Florence, June 19–23, 1984, p. 99.
158. Taylor, I. L. and R. Garcia. Effects of pancreatic polypeptide, caerulein, and bombesin on satiety on obese mice. *Am J Physiol* **248:** G277–G280, 1985.
159. Teitelbaum, D. H., T. M. O'Dorisio, T. S. Gaginella and W. E. Perkins. Effects of caerulein on rat small and large intestinal fluid transport in vivo. *Regul Pept* **4:** 97–106, 1982.
160. Tokiwa, T., I. Uemura, H. Murakami, S. Tsuruoka, M. Kaneko, K. Ogawa and T. Manzai. Studies on caerulein (F.I. 6934). Absorption, distribution, metabolism and excretion of caerulein. *Jpn J Pharmacol* **25:** 747–762, 1975.
161. Ushijima, K., M. Kawata, T. Matsuura, Y. Nojyo and Y. Sano. Uptake of exogenous caerulein in the rat brain: an immunohistochemical study. *Biomed Res* **5:** 117–124, 1984.
162. Vaccarino, F. J. and G. F. Koob. Microinjections of nanogram amounts of sulfated cholecystokinin octapeptide into the rat nucleus accumbens attenuates brain stimulation reward. *Neurosci Lett* **52:** 61–66, 1984.
163. Vanderhaeghen, J. J., J. C. Signeau and W. Gepts. New peptide in the vertebrate CNS reacting with antigastrin antibodies. *Nature* **257:** 604–605, 1975.
164. Van Dijk, A., J. G. Richards, A. Trzeciak, D. Gillesen and H. Möhler. Cholecystokinin receptors: biochemical demonstration and autoradiographical localization in rat brain and pancreas using [$^3$H]cholecystokinin, as radioligand. *J Neurosci* **4:** 1021, 1984.
165. Van Ree, J. M., O. Gaffori and D. de Wied. In rats, the behavioral profile of CCK-8 related peptides resembles that of antipsychotic agents. *Eur J Pharmacol* **93:** 63–78, 1983.
166. Van Ree, J. M. and D. de Wied. Neuroleptic-like activity and antipsychotic action of cholecystokinin-related peptides. *Ann NY Acad Sci* **448:** 671–673, 1985.
167. Van Ree, J. M., W. M. A. Verhoeven, G. J. Brouwer and D. de Wied. Ceruletide resembles antipsychotics in rats and schizophrenic patients. *Neuropsychobiology* **12:** 4–8, 1984.
168. Vasar, E., M. Naimets, A. Nurk and L. Allikmets. Caerulein stimulates [$^3$H]-spiperone binding in vivo after long-term haloperidol administration. *Psychopharmacol Bull* **20:** 691–692, 1984.
169. Verbanck, P. M. P., F. Lotstra, C. Gilles, P. Linkowski, J. Mendlewicz and J. J. Vanderhaeghen. Reduced cholecystokinin immunoreactivity in the cerebrospinal fluid of patients with psychiatric disorders. *Life Sci* **34:** 67–72, 1984.
170. Vizi, E. S., G. Bertaccini, M. Impicciatore and J. Knoll. Acetylcholine-releasing effect of gastrin and related polypeptides. *Eur J Pharmacol* **17:** 175–178, 1972.

171. Vizi, S. E., G. Bertaccini, M. Impicciatore and J. Knoll. Evidence that acetylcholine released by gastrin and related polypeptides contributes to their effect on gastrointestinal motility. *Gastroenterology* **64**: 268–277, 1973.
172. Watanabe, N., T. Moroji, K. Tada and N. Aoki. A therapeutic trial of caerulein to a long-term heavy marihuana user with amotivational syndrome. *Prog Neuropsychopharmacol Biol Psychiatry* **8**: 419–422, 1984.
173. Watanabe, N., T. Sakai, K. Tada, N. Takemura, Y. Nogami and T. Moroji. The effects of caerulein on the nocturnal sleep. *Ann Rep Pharmacopsychiat Res Found* **15**: 63–69, 1984.
174. Worms, P. and K. G. Lloyd. Predictability and specificity of behavioral screening tests for neuroleptics. *Pharmacol Ther* **5**: 445–450, 1979.
175. Zetler, G. Antagonism of cholecystokinin-like peptides by opioid peptides, morphine or tetrodotoxin. *Eur J Pharmacol* **60**: 67–77, 1979.
176. Zetler, G. Analgesia and ptosis caused by caerulein and cholecystokinin octapeptide (CCK-8). *Neuropharmacology* **19**: 415–422, 1980.
177. Zetler, G. Anticonvulsant effects of caerulein and cholecystokinin octapeptide, compared with those of diazepam. *Eur J Pharmacol* **65**: 297–300, 1980.
178. Zetler, G. Effects of cholecystokinin-like peptides on rearing activity and hexobarbital-induced sleep. *Eur J Pharmacol* **66**: 137–139, 1980.
179. Zetler, G. Central depressant effects of caerulein and cholecystokinin octapeptide (CCK-8) differ from those of diazepam and haloperidol. *Neuropharmacology* **20**: 277–283, 1981.
180. Zetler, G. Differential cataleptogenic and antistereotypic effects of caerulein and haloperidol. *Neuropharmacology* **20**: 681–686, 1981.
181. Zetler, G. Anticonvulsant effects of caerulein, cholecystokinin octapeptide (CCK-8) and diazepam against seizures produced in mice by harman, thiosemicarbazide and isoniazid. *Neurosci Lett* **24**: 175–180, 1981.
182. Zetler, G. Central effects of ceruletide analogues. *Peptides* **2**: Suppl 2, 65–69, 1981.
183. Zetler, G. Ceruletide, ceruletide analogues and cholecystokinin octapeptide (CCK-8): Effects on motor behaviour, hexobarbital-induced sleep and harman-induced convulsions. *Peptides* **3**: 701–704, 1982.
184. Zetler, G. Caerulein and cholecystokinin octapeptide (CCK-8): sedative and anticonvulsive effects in mice unaffected by the benzodiazepine antagonist Ro 15-1788. *Neurosci Lett* **28**: 287–290, 1982.
185. Zetler, G. Caerulein and morphine: an attempt to differentiate their antinociceptive effects. *Pharmacology* **25**: 121–129, 1982.
186. Zetler, G. Cholecystokinin octapeptide, caerulein and caerulein analogues: effects on thermoregulation in the mouse. *Neuropharmacology* **21**: 795–801, 1982.
187. Zetler, G. Cholecystokinin octapeptide (CCK-8), ceruletide and analogues of ceruletide: effects on tremor induced by oxotremorine, harmine and ibogaine. A comparison with prolyl-leucylglycine amide (MIF), anti-parkinsonian drugs and clonazepam. *Neuropharmacology* **22**: 757–766, 1983.
188. Zetler, G. Apomorphine separates the antinociceptive effects of cholecystokinin octapeptide and ceruletide from those of morphine. *Eur J Pharmacol* **92**: 151–154, 1983.
189. Zetler, G. Behavioral pharmacology of CCK and analogs. *Psychopharmacol Bull* **19**: 347–351, 1983.
190. Zetler, G. Neuroleptic-like effects of ceruletide and cholecystokinin octapeptide: interactions with apomorphine, methylphenidate and picrotoxin. *Eur J Pharmacol* **94**: 261–270, 1983.
191. Zetler, G. Clonidine and yohimbine separate sedation and ptosis by cholecystokinin octapeptide and ceruletide. *Eur J Pharmacol* **102**: 333–340, 1984.
192. Zetler, G. Ceruletide, ceruletide analogues and cholecystokinin octapeptide (CCK-8): Effects on isolated intestinal preparations and gallbladders of guinea pigs and mice. *Peptides* **5**: 729–736, 1984.
193. Zetler, G. Antistereotypic effects of cholecystokinin octapeptide (CCK-8), ceruletide, and related peptides on apomorphine-induced gnawing in sensitized mice. *Neuropharmacology* **24**: 251–259, 1985.
194. Zetler, G. Neuropharmacological profile of cholecystokinin-like peptides. *Ann NY Acad Sci* **448**: 448–469, 1985.
195. Zetler, G. Clonidine sensitizes mice for apomorphine-induced stereotypic gnawing: antagonism by neuroleptics and cholecystokinin-like peptides. *Eur J Pharmacol* **111**: 309–318, 1985.
196. Zetler, G. and K.-H. Mörsdorf. Effects of ceruletide and cholecystokinin octapeptide on eating in mice. Interactions with naloxone and the enkephalin analogue, FK 33-824. *Naunyn Schmiedebergs Arch Pharmacol* **325**: 209–213, 1984.
197. Zetler, G. and A. Raberg. Ceruletide inhibits water intake in deprived mice: comparison with morphine and the enkephalin analogue, FK 33-824. *Eur J Pharmacol* **114**: 247–251, 1985.