

# NEUROPEPTIDES

# INTERET DES NEUROPEPTIDES

Les neurones disposent de 2 systèmes de communication

- classique
- peptidique

qui leur permettent de largement varier le décours et l'amplitude des réponses postsynaptiques

Le plus souvent les neuropeptides coexistent au sein d'un même neurone, avec un autre neuropeptide ou avec un neurotransmetteur "classique" (ACh, 5-HT...)

# EXAMPLES OF PEPTIDES FOUND IN THE VERTEBRATE NERVOUS SYSTEM

## Opioid peptides

- Met-enkephalin
- Leu-enkephalin
- $\beta$ -Endorphin
- Dynorphin

## Gut-brain peptides

- Substance P (SP)
- Vasoactive intestinal peptide (VIP)
- Cholecystokinin (CCK)
- Neurotensin (NT)
- Neuropeptide Y (NPY)
- Galanin
- Insulin
- Glucagon
- Bombesin
- Gastrin
- Secretin
- Motilin

## Hypothalamic releasing hormones

- Thyrotropin-releasing hormone (TRH)
- Luteinizing hormone-releasing hormone (LHRH), also known as gonadotropin-releasing hormone (GnRH)
- Corticotropin-releasing factor (CRF)
- Growth hormone-releasing hormone
- Somatostatin

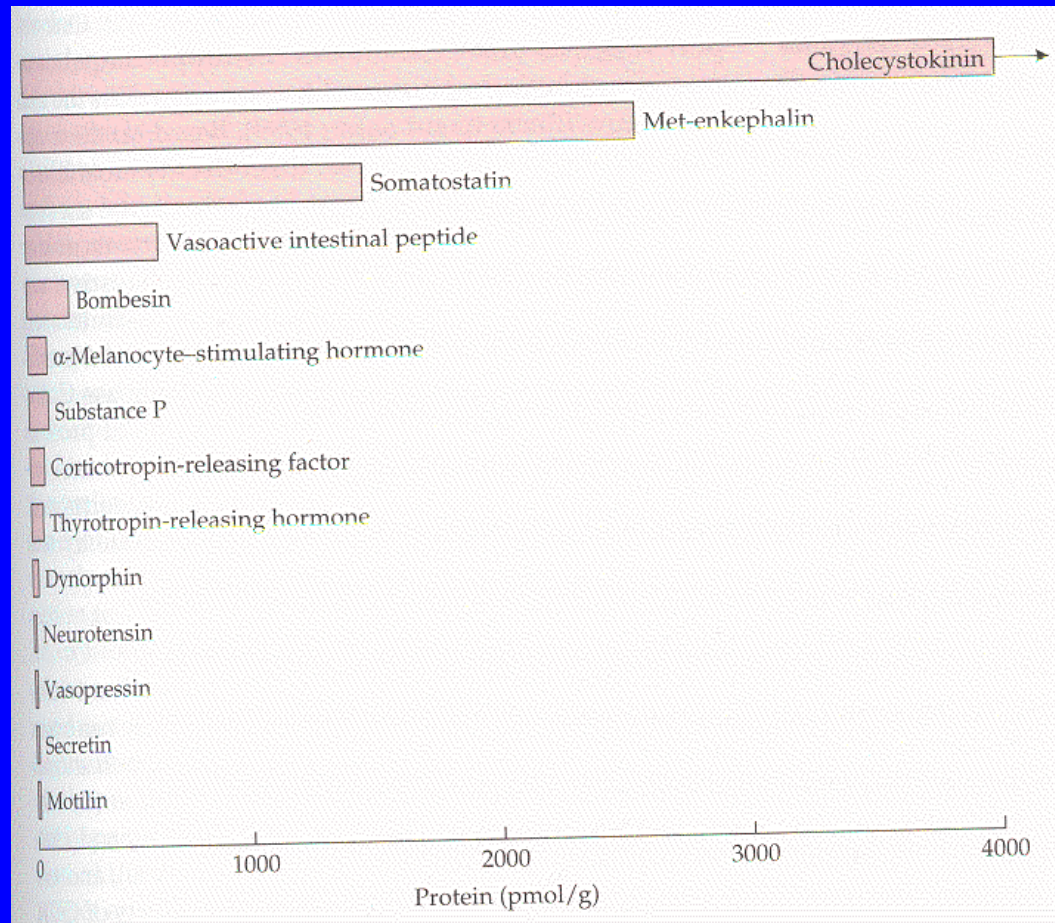
## Pituitary hormones

- Vasopressin
- Oxytocin
- Adrenocorticotrophic hormone (ACTH)
- Growth hormone (GH)
- Thyrotropin, or thyroid-stimulating hormone (TSH)
- Luteinizing hormone (LH)
- Prolactin
- $\alpha$ -Melanocyte-stimulating hormone ( $\alpha$ -MSH)

## Other circulating hormones and miscellaneous peptides

- Angiotensin II
- Bradykinin
- Atrial natriuretic factor (ANF)
- Calcitonin gene-related peptide (CGRP)

# ESTIMATED CONCENTRATIONS OF NEUROPEPTIDES IN THE CEREBRAL CORTEX



# EFFETS DES PRINCIPAUX NEUROPEPTIDES

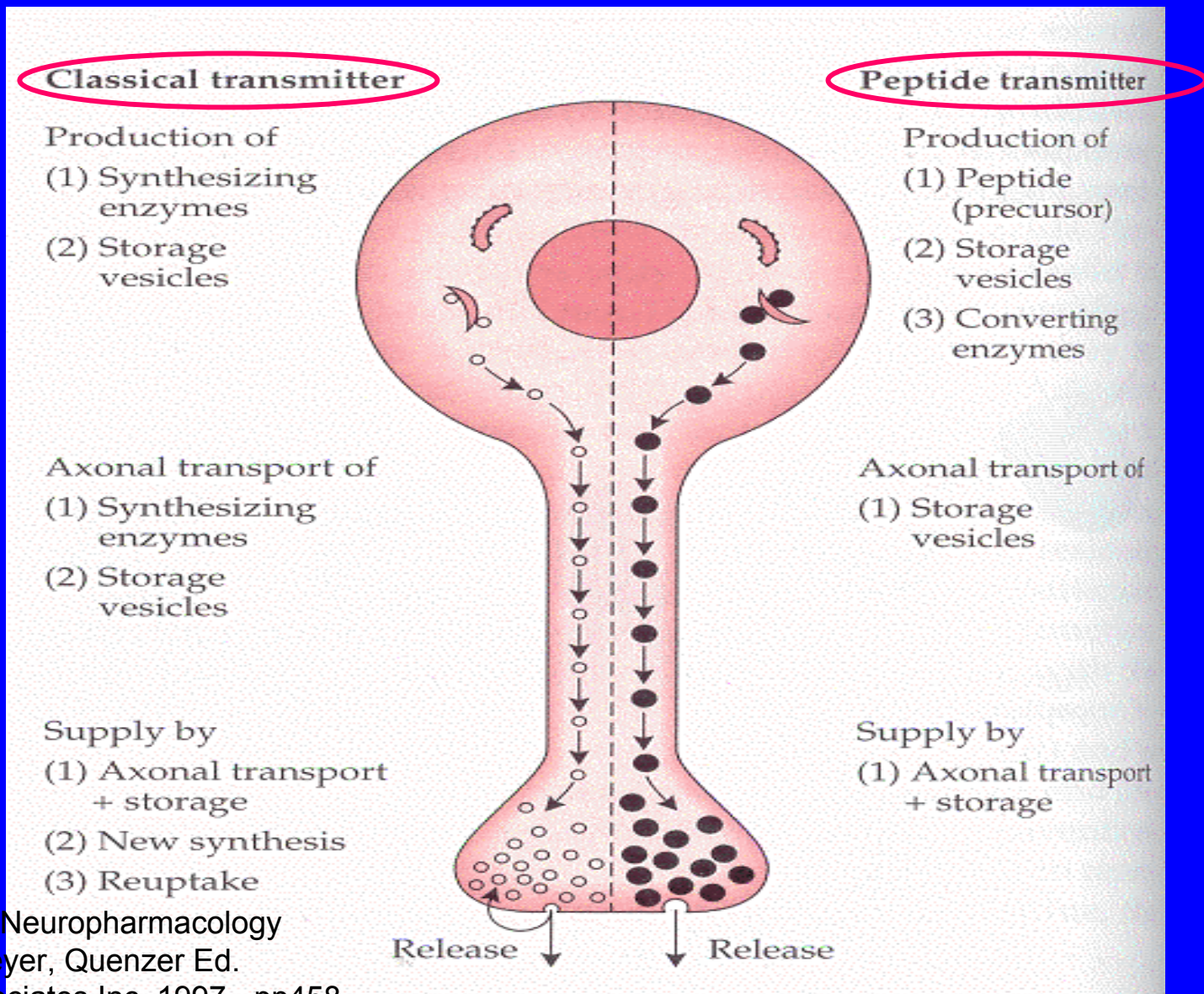
|   | Rôles comme neurotransmetteur                  |
|---|--|
| <b>Vasopressine</b>                           | Apprentissage - mémoire                        |
| <b>Ocytocine</b>                              | Comportement sexuel féminin et maternel        |
| <b>TRH (thyrotropin - releasing hormone)</b>  | Excitateur                                     |
| <b>CRF (corticotropin - releasing factor)</b> | Excitateur<br>Réponses physiologiques ~ stress |
| <b>SP (substance P)</b>                       | Nociception                                    |
| <b>CCK (cholecystokinin)</b>                  | Comportement face à la nourriture              |

# GROWTH-PROMOTING EFFECTS OF NEUROPEPTIDES IN NORMAL AND CANCER CELLS

| Neuropeptide | Normal cells  | Cancer cells   |
|--------------|---|--|
| Angiotensin  | Cardiac and lung fibroblasts; hepatic stellate cells; intestinal epithelial cells; mesangial cells; smooth muscle | Pancreatic; adrenocortical; breast epithelial                        |
| Bombesin     | 3T3 fibroblasts; airway epithelium  | SCLC; prostate; glioblastoma; renal cell carcinoma; colon            |
| Bradykinin   | 3T3 fibroblasts; mesangial cells  | SCLC; prostate   |
| CCK          | Pancreas  | SCLC; pancreatic; colon  |
| Endothelin   | 3T3 and cardiac fibroblasts; endothelial; keratinocytes; astrocytes; smooth muscle                                | Ovarian and cervical carcinoma; Kaposi's sarcoma; prostate; melanoma |
| Galanin      | Sensory neurons   | SCLC   |
| Gastrin      | ECL; gastric mucosa; colon epithelium   | SCLC; colon  |
| Neurotensin  | Adrenal cortex; intestinal epithelial cells   | Pancreatic; SCLC; prostate; astrocytes; colon                        |
| NPY          | Olfactory neurons; smooth muscle  | Breast cancer  |
| Substance P  | Fibroblasts; smooth muscle; endothelial cells; corneal epithelial cells; astrocytes; T lymphocytes                | Astroglomas  |
| Vasopressin  | 3T3 fibroblasts; mesangial cells; hepatic stellate cells  | SCLC; breast; pheochromocytoma                                       |
| VIP/PACAP    | 3T3 fibroblasts; keratinocytes; brain development   | Breast; SCLC; prostate; colon; urinary bladder; neuroblastoma        |



# NEUROPEPTIDES - SYNTHESIS, STOCKAGE ET TRANSPORT



# SYNTHESE DES NEUROPEPTIDES

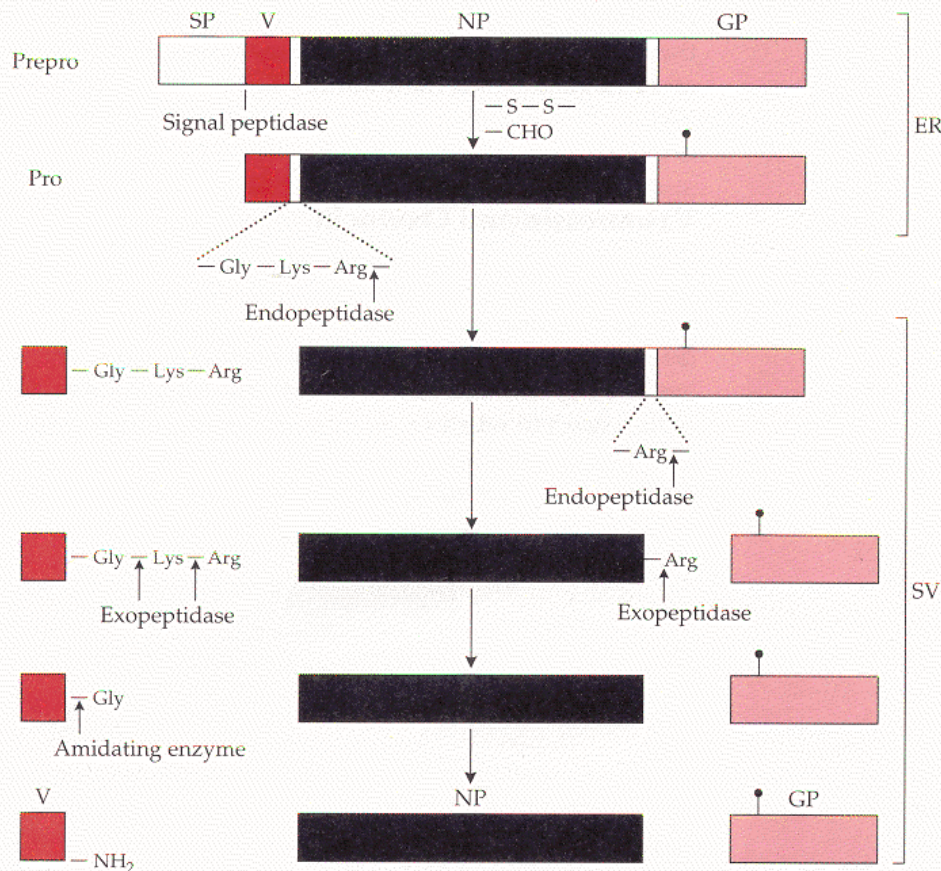
Le chemin entre le lieu de synthèse (soma) et le lieu de sécrétion (terminaisons axonales) est LONG.

- Synthèse dans le corps cellulaire du neurone, au niveau des ribosomes
- Transport dans la lumière du réticulum endoplasmique rugueux.
- Passage dans l'appareil de Golgi puis, de là, dans des vésicules de sécrétion.
- Transport de ces vésicules de sécrétion par le transport axonal antérograde rapide jusque dans les terminaisons de l'axone du neurone peptidergique



# STRUCTURE AND PROCESING OF THE VASOPRESSIN PRECURSOR

SP Signal peptide 19 amino acids  
V Vasopressin 9 amino acids  
NP Neurophysin 95 amino acids  
GP Glycopeptide 39 amino acids



Les peptides sont habituellement formés à partir d'un précurseur plus ou moins long.

Un même précurseur peut donner par clivage plusieurs peptides actifs (Pro-opiomélanocortine POMC donne  $\beta$ -endorphine,  $\alpha$ -MSH et ACTH).

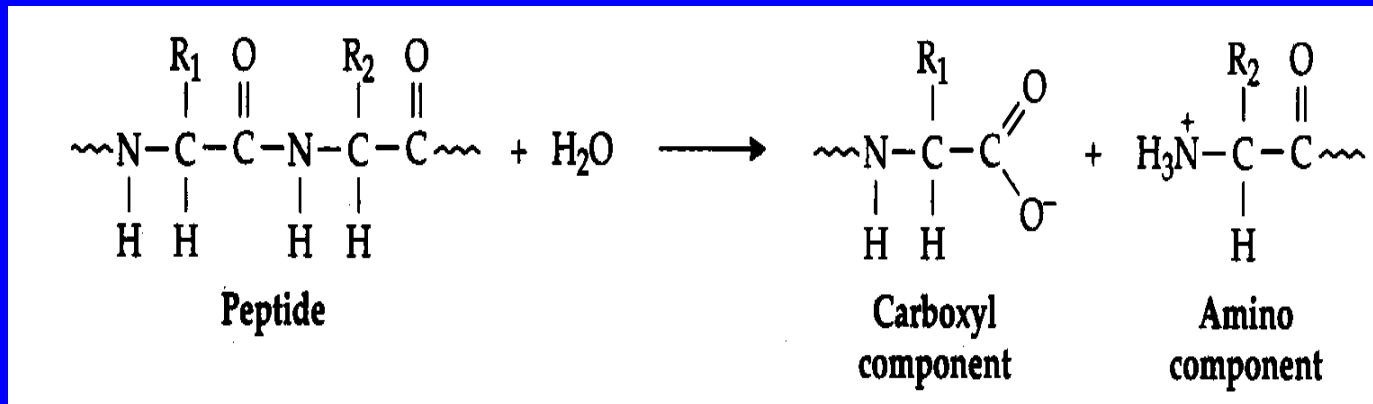
ER: reticulum endoplasmic:  
place of the initial processing

SV: secretory vesicles where  
peptides are packaged

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 464

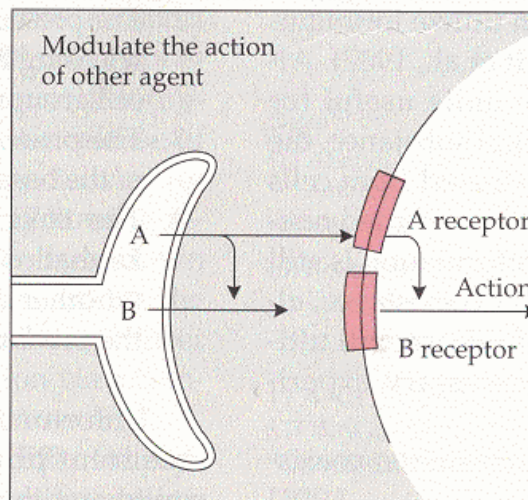
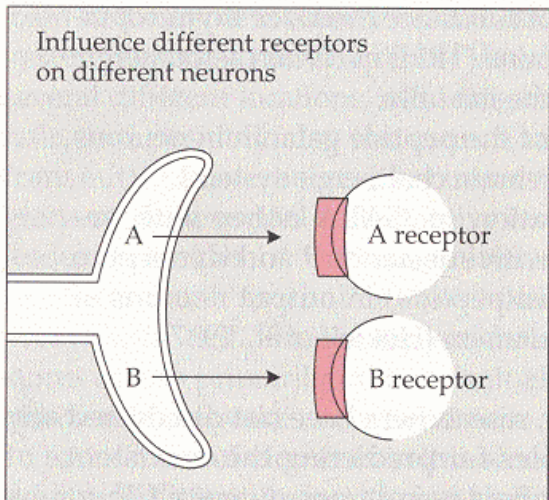
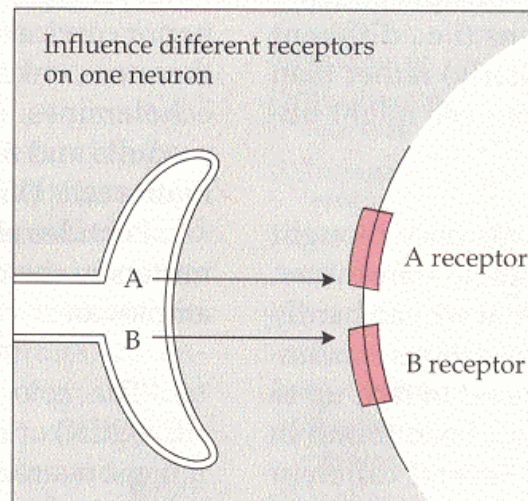
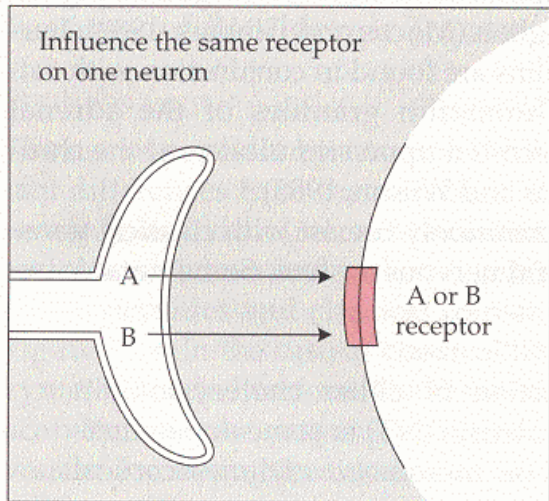
# DEGRADATION ET RECAPTURE DES NEUROPEPTIDES

- Dégradation par les endopeptidases



- Absence de mécanisme de recapture à haute affinité comme ceux existant pour les neurotransmetteurs
- Les peptides ne sont pas dégradés rapidement et persistent dans le milieu extracellulaire. Ils peuvent donc agir pendant des périodes relativement longues (qq secondes à qq minutes)

# POSSIBLE INTERACTIONS OF COEXISTING NEUROTRANSMITTERS OU NEUROMODULATEURS

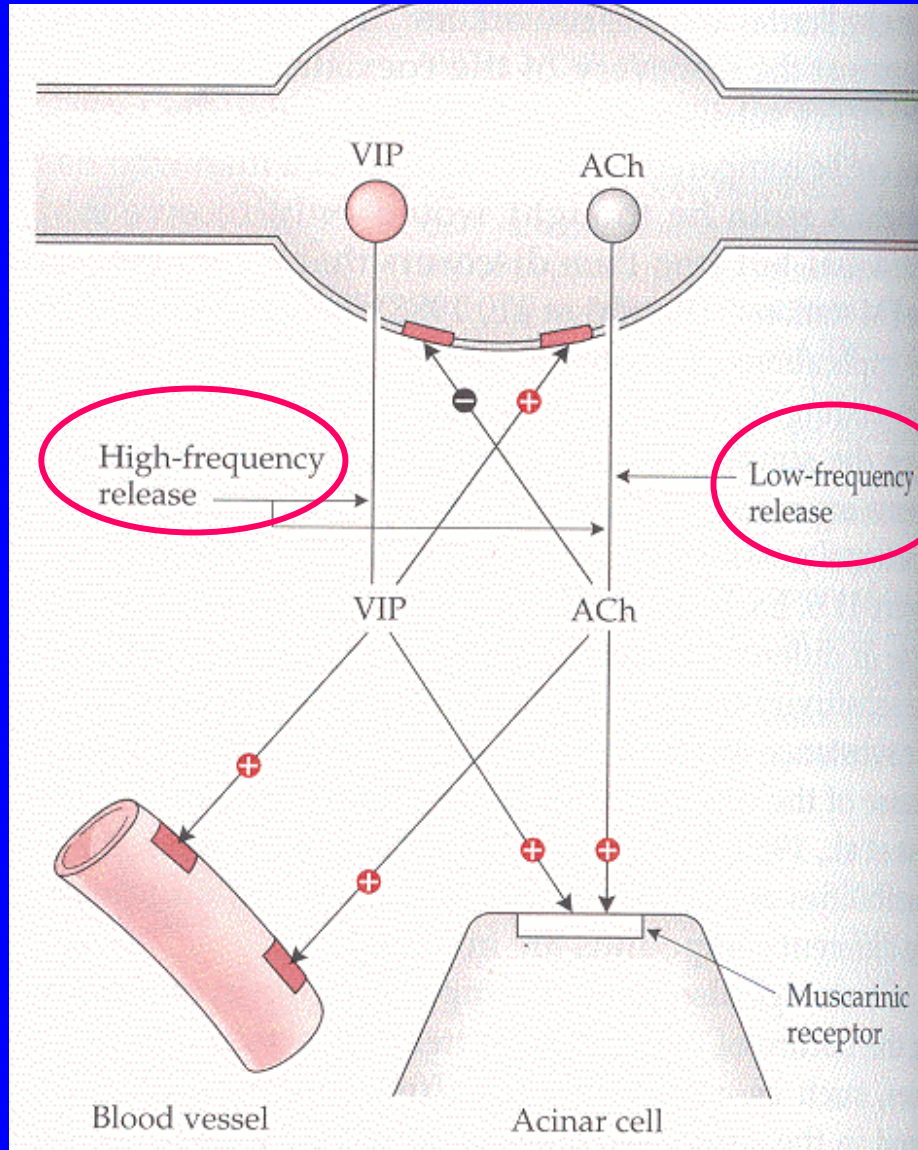


En règle générale, les neuropeptides sont associés à un autre neuromédiateur dit “classique” ou à un autre neuromédiateur dans une même terminaison synaptique

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 460



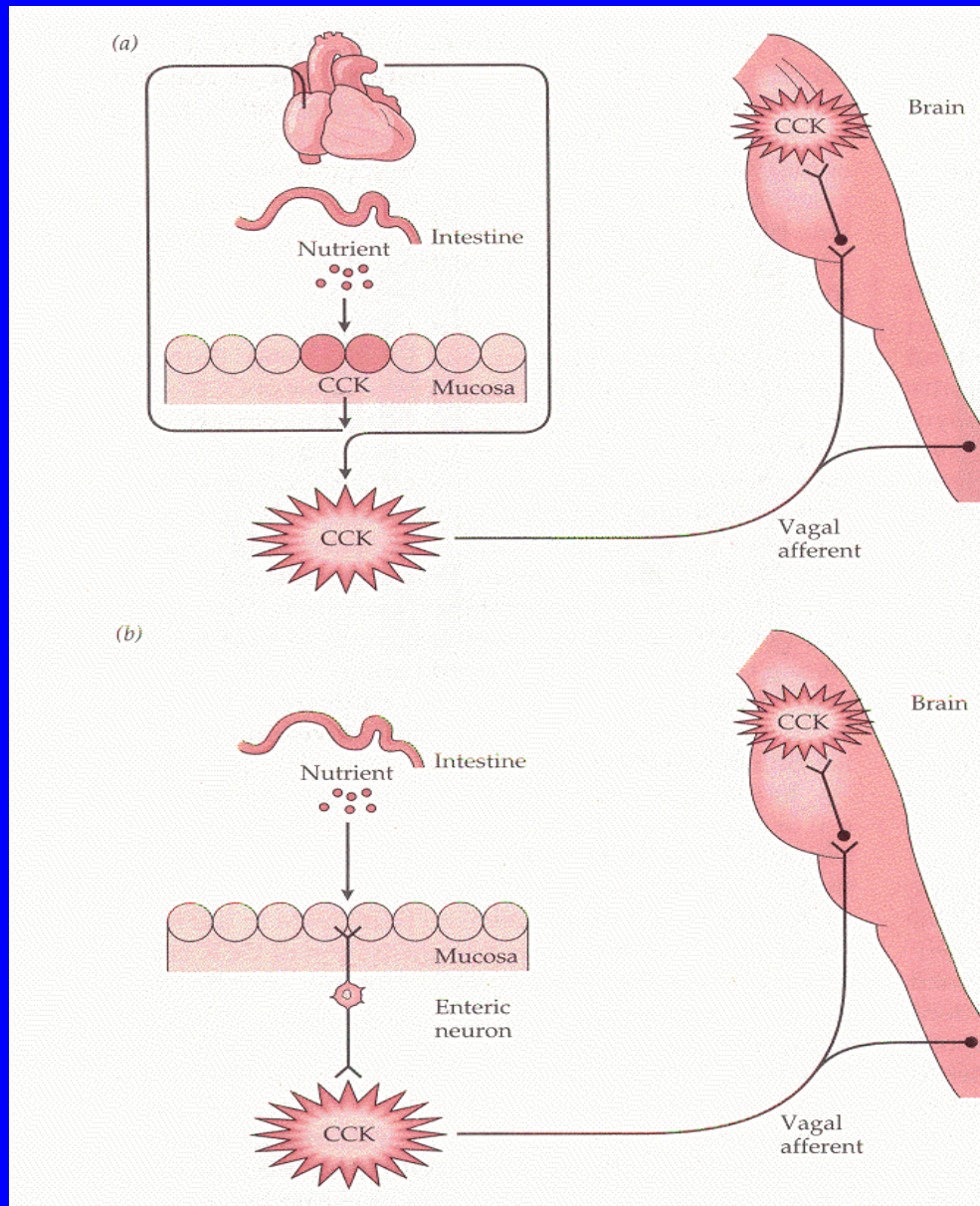
# COEXISTENCE OF ACETYLCHOLINE AND VASOACTIVE INTESTINAL PEPTIDE



Controle de la substance libérée par l'activité de la voie afférente (l'ACh et le VIP -vasoactive intestinal peptide - sont libérés par la même terminaison à des fréquences de stimulation différentes)

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 462

# NEUROPEPTIDES - INTERCONNECTIONS BETWEEN SNC AND SNP EFFETCS

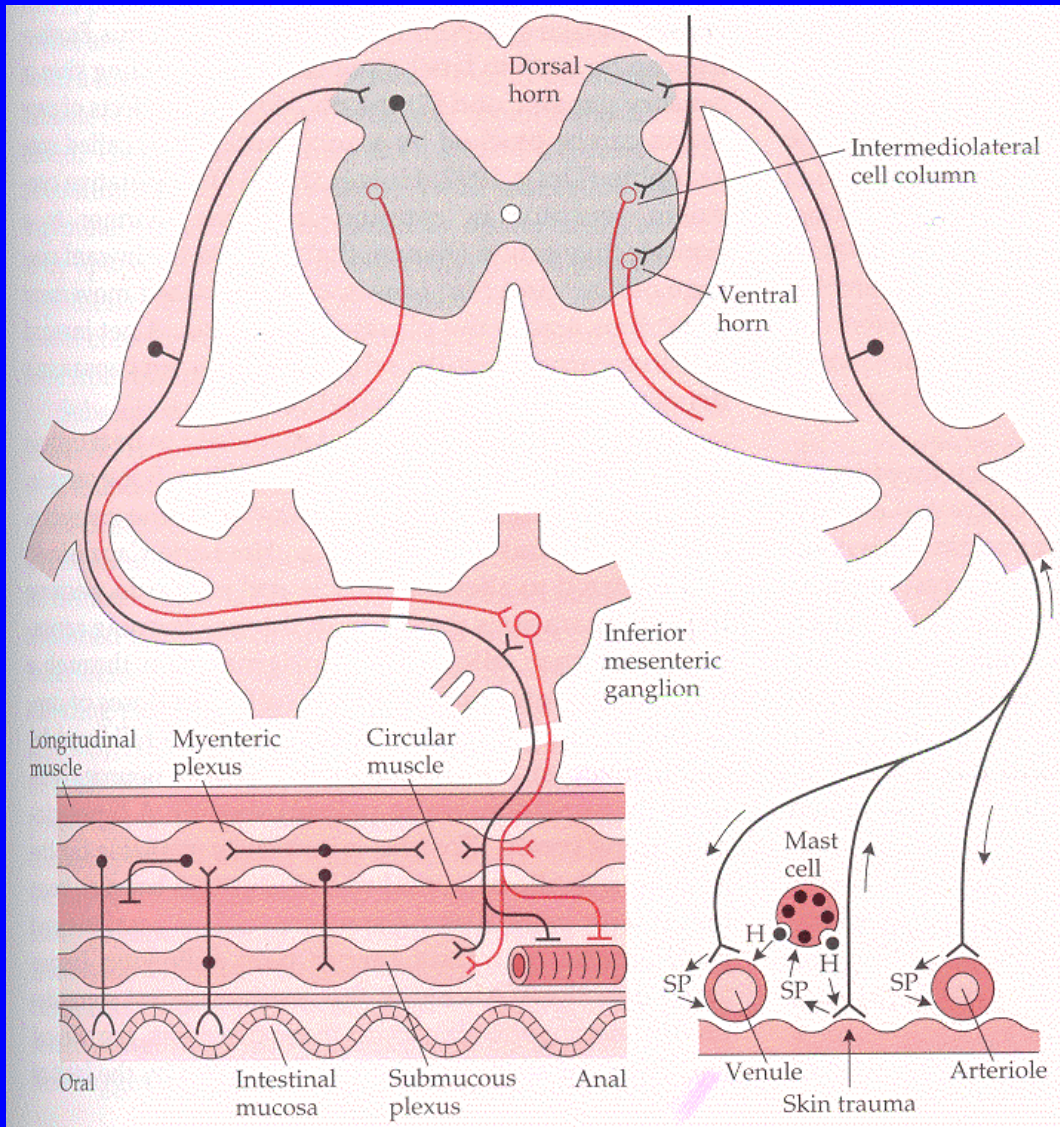


Hypothesized mechanisms by which cholecystikine regulates eating behavior in response to intestinal food stimulation

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 490



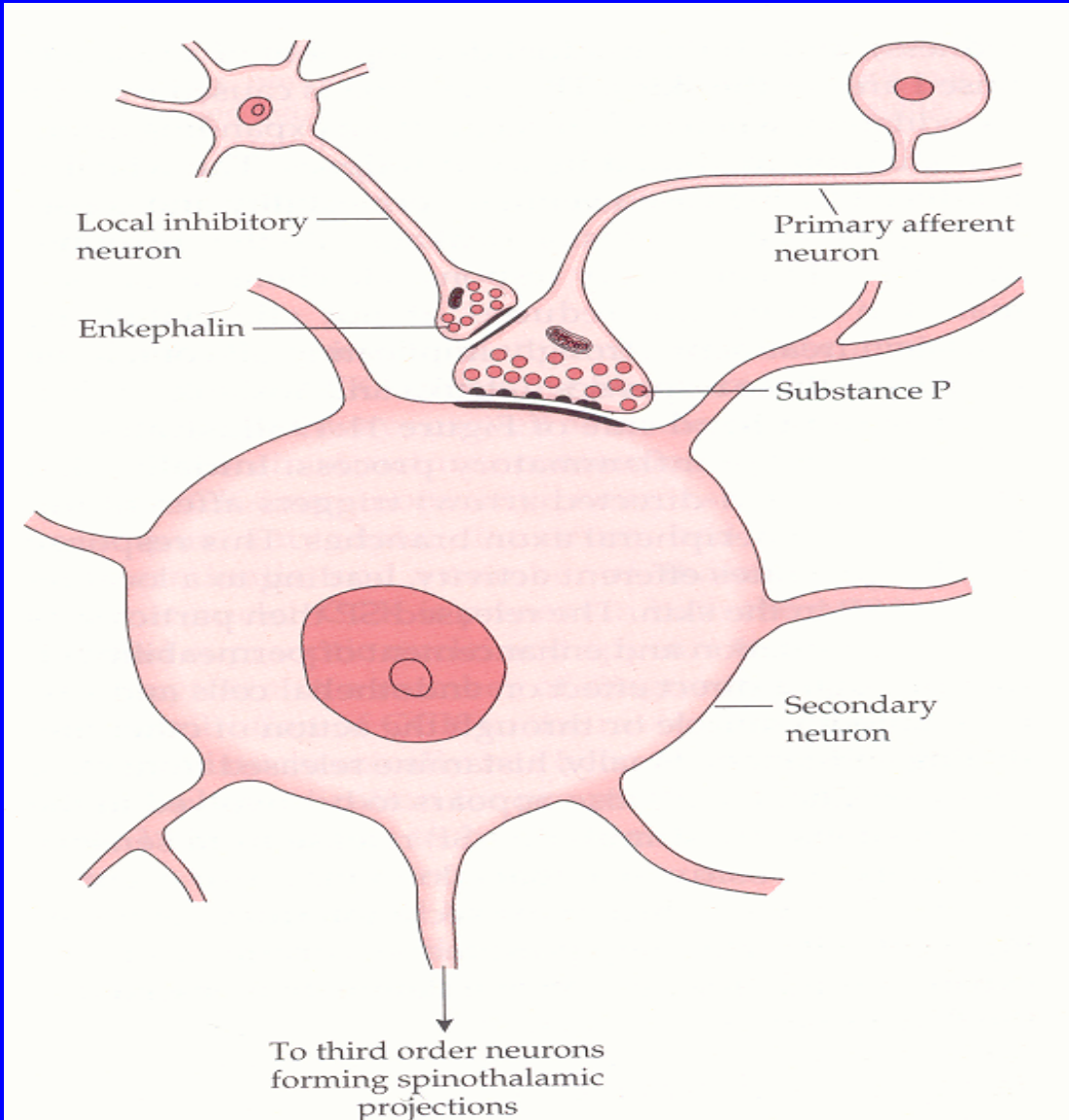
# NEUROPEPTIDES - INTERCONNECTIONS BETWEEN SNC AND SNP EFFETCS



Location of  
substance P-  
containing neurons  
and fibers in the  
spinal cord, skin,  
and intestine

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 481

# NEUROPEPTIDES - INTERMODULATION



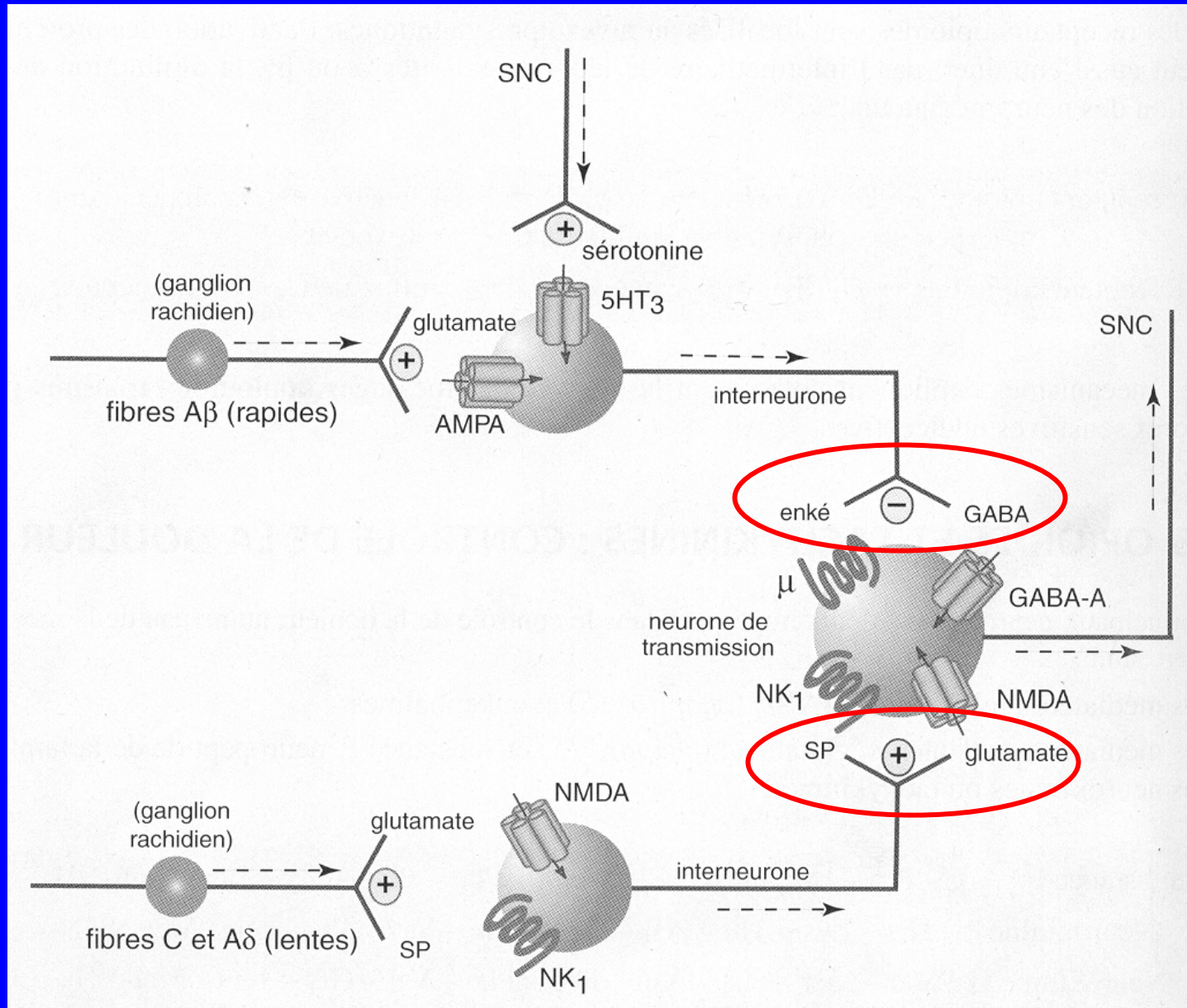
Opiate-induced  
inhibition of substance  
P release

Principles of Neuropharmacology  
Feldman, Meyer, Quenzer Ed.  
Sinauer Associates Inc. 1997 - pp 483

# NEUROPEPIDES OPIOIDES ET CONTRÔLE DE LA DOULEUR



# NEUROPEPTIDES OPIOIDES ET CONTRÔLE DE LA DOULEUR



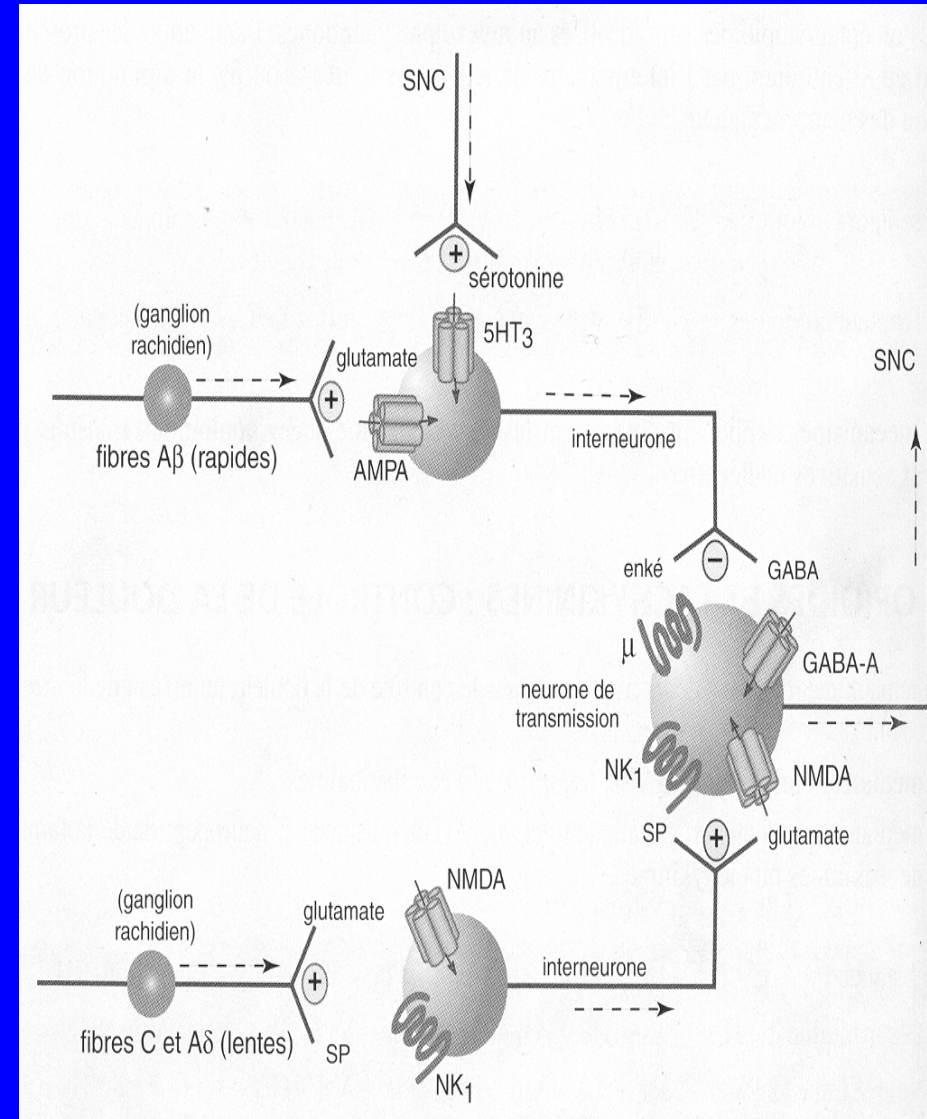
# NEUROPEIDES OPIOIDES ET CONTRÔLE DE LA DOULEUR

Les voies sensibles nociceptives périphériques sont constituées de neurones dont les corps cellulaires sont situés dans les ganglions rachidiens.

Le signal nociceptif issu de la périphérie aboutit au niveau de la moelle épinière.

Les fibres nociceptives A $\delta$  ou C sont dites lentes; elles sont non myélinisées; leur message excitateur en provenance de la périphérie arrive tardivement à la corne postérieure. Il est transmis par la substance P et par le glutamate.

Les fibres nociceptives A $\beta$  sont dites rapides; elles sont myélinisées; leur message excitateur en provenance de la périphérie arrive rapidement à la corne postérieure. Il est transmis par les enképhalines.





# NEUROPEPIDES OPIOIDES ET CONTRÔLE DE LA DOULEUR

Les principaux transmetteurs impliqués dans le contrôle de la douleur au niveau de la moelle épinière sont

- **des médiateurs inhibiteurs:**

- GABA
- enképhalines

Leu-enképhaline: TYR-GLY-GLY-PHE-LEU

Met-enképhaline: TYR-GLY-GLY-PHE-MET

- **des médiateurs excitateurs:**

- Glutamate,
- substance P :

ARG-PRO-LYS-PRO-GLN-GLN-PHE-PHE-GLY-LEU-MET-NH<sub>2</sub>

- neuropeptides de la famille des neurokinines ou tachikinines

- NKA: HIS-LYS-THR-ASP-SER-PHE-VAL-GLY-LEU-MET-NH<sub>2</sub>

- NKB: ASP-MET-HIS-ASP-PHE-PHE-VAL-GLY-LEU-MET-NH<sub>2</sub>

# NEUROPEPTIDES OPIOIDES – ENKEPHALINES, ENDOMORPHINES

## Enképhalines

**Leu-enképhaline:** TYR-GLY-GLY-PHE-LEU

**Met-enképhaline:** TYR-GLY-GLY-PHE-MET

Bulbe, moelle épinière

Transmission du signal nociceptif

Rapidement dégradées par les peptidases

## Endomorphines et nociceptine

**Endomorphine-1:** TYR-PRO-TRP-PHE-NH<sub>2</sub>

**Endomorphine –2:** TYR-PRO-PHE-PHE-NH<sub>2</sub>

**Nociceptine ou orphanine FQ**

# NEUROPEPTIDES OPIOIDES - ENDORPHINES

**$\beta$ -endorphine:** TYR-GLY-GLY-PHE-MET- THR-SER-GLU-  
LYS-SER-GLN-THR-PRO-LEU-VAL-THR-LEU-PHE-LYS-  
ASN-ALA-ILE-ILE-LYS-ASN-ALA-TYR-LYS-LYS-GLY-GLU

Hypophyse, hypothalamus

**$\alpha$ -néoendorphine:** TYR-GLY-GLY-PHE-LEU-ARG-LYS-TYR-  
PRO-LYS

**$\beta$ -néoendorphine:** TYR-GLY-GLY-PHE-LEU-ARG-LYS-TYR-  
PRO

**Dynorphine A:** TYR-GLY-GLY-PHE-LEU-ARG-ARG-ILE-  
ARG-PRO-LYS-LEU-LYS-TRP-ASP-ASN-GLN

**Dynorphine B:** TYR-GLY-GLY-PHE-LEU-ARG-ARG-GLN-  
PHE-LYS-VAL-VAL-THR

# NEUROPEPIDES OPIOIDES - ORIGINE

Proviennent de 3 précurseurs polypeptidiques distincts

- La **proenképhaline A**, précurseur de la met-enképhaline
- La **prodynorphine** ou proenképhaline B, précurseur de la leu-enképhaline, des néoendorphines et des dynorphines
- La **pro-opiomélanocortine** (POMC) qui comprend la séquence de la
  - $\beta$ -endorphine
  - ACTH
  - MSH (melanocyte-stimulating hormone)
  - $\beta$ -LPH (lipotropine)

# NEUROPEPIDES OPIOIDES-RECEPTEURS

|                     | $\delta$   | $\kappa$   | $\mu$  | ORL-1                    |
|---------------------|--|--|--|--------------------------|
| <b>Ki (nM)</b>      |  |  |  |                          |
| Morphine            | >1000  | 163  | 1,4  |                          |
| Naloxone            | 95   | 16   | 4  |                          |
| Dynorphine A        | 45   | 5  | 120  | 110                      |
| Nociceptine         |  |  |  | 0.1                      |
| <b>Localisation</b> | Bulbe olfactif<br>Néocortex,<br>Striatum,<br>Noyau accumbens<br>Intestin | Couche II de la moelle épinière<br>Noyau accumbens<br>Clastrum<br>Intestin | Striatum<br>Noyau caudé<br>Putamen<br>Intestin | SNC<br>SNP               |
| <b>Effet</b>        |  | Régulation du péristaltisme par action centrale                            | Nociception                                    | Transmission nociceptive |



# NEUROPEPIDES OPIOIDES- RECEPTEURS ORPHELINS

**La découverte de la nociceptine:  
exemple de pharmacologie inverse**

**Clonage d'une protéine ORL1**

**De fonction inconnue,**

**Dont la structure présente une homologie important avec  
celle des récepteurs opioïdes  $\delta, \kappa, \mu$**

**= Récepteur orphelin: ne peut être activé par aucun  
médiateur connu avec une bonne affinité**

**Découverte du ligand endogène: la nociceptine ou orphanine FQ**

# NEUROPEPIDES OPIOIDES RECEPTEURS ORPHELINS

autres médiateurs découverts par une démarche de pharmacologie inverse

## **Oréxine ou hypocréline (1998)**

régulation du comportement alimentaire et régulation du sommeil

## **Prolactine-releasing-peptide (1998)**

secrétion de prolactine

## **Ghréline (1999)**

secrétion de l'hormone de croissance

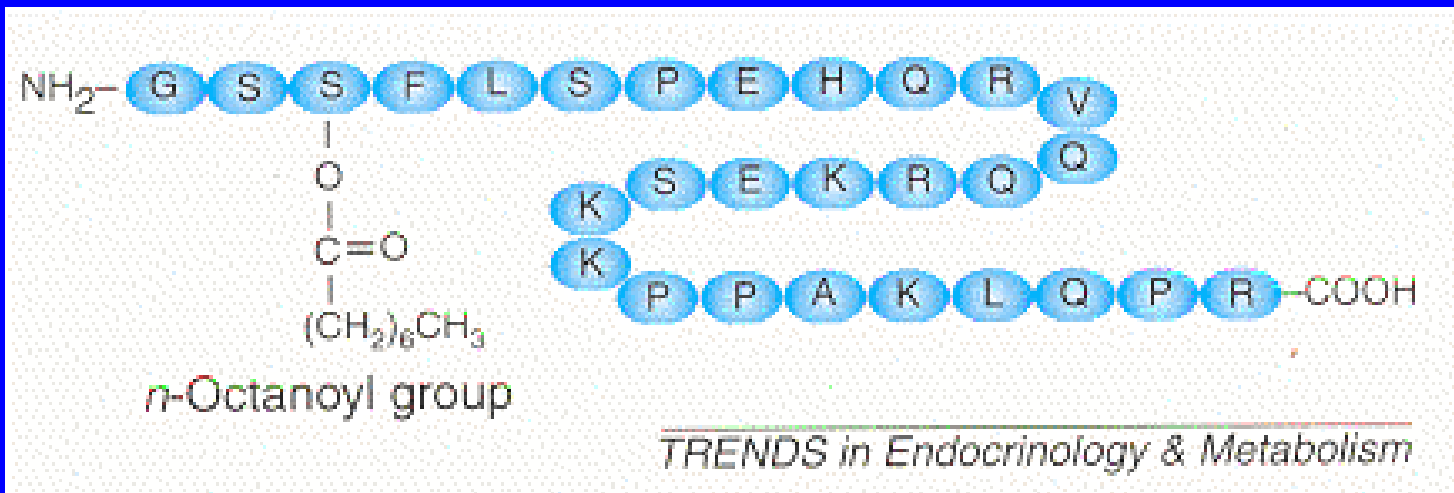
## **Urotensine II (1999)**

vasoconstriction

## **Neuromédine U (2000)**

contraction de l'utérus

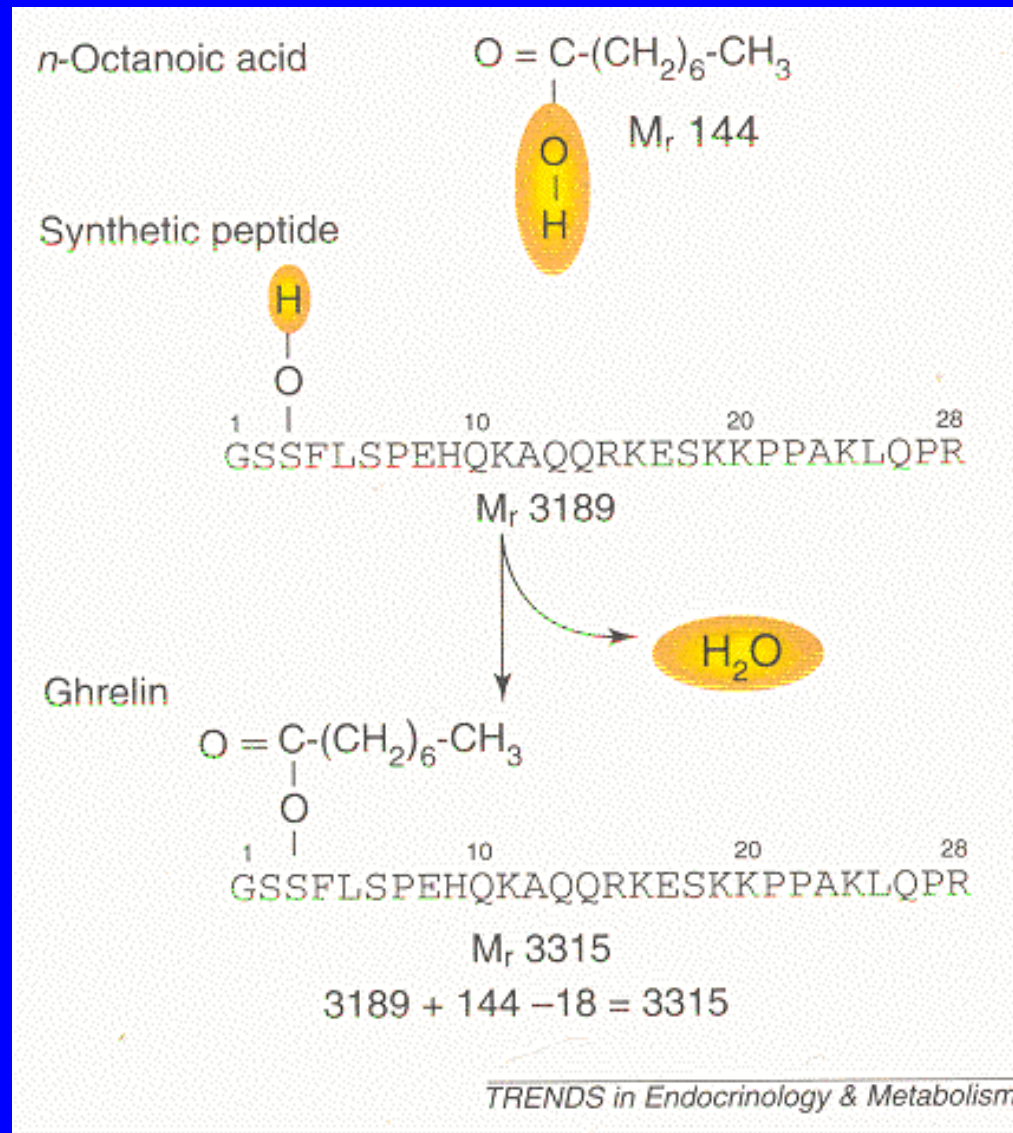
# STRUCTURE OF HUMAN GHRELIN



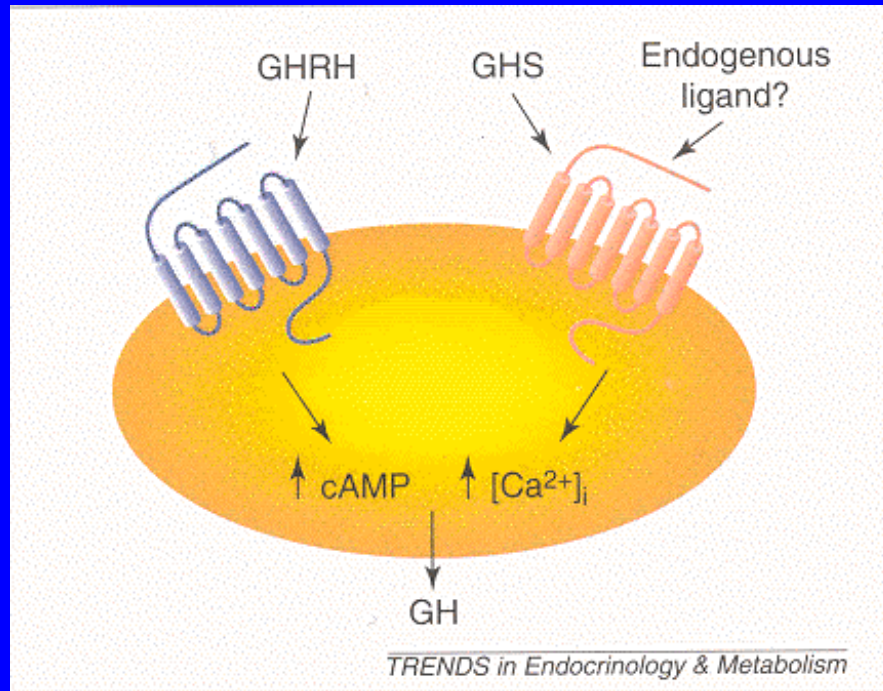
|         |                              |
|---------|------------------------------|
| Ghrelin | GSSFLSPEHQRVQQRKESKKPPAKLQPR |
| Motilin | FVPIFTYGELQRMQE-KERNKGQ      |

*TRENDS in Endocrinology & Metabolism*

# SYNTHESIS OF GHRELIN



# PATHWAYS OF GROWTH HORMONE (GH) RELEASE FROM THE PITUITARY



Kojima et al,  
Trends Endocrinology & Metabolism  
(2001) 12: 118-122

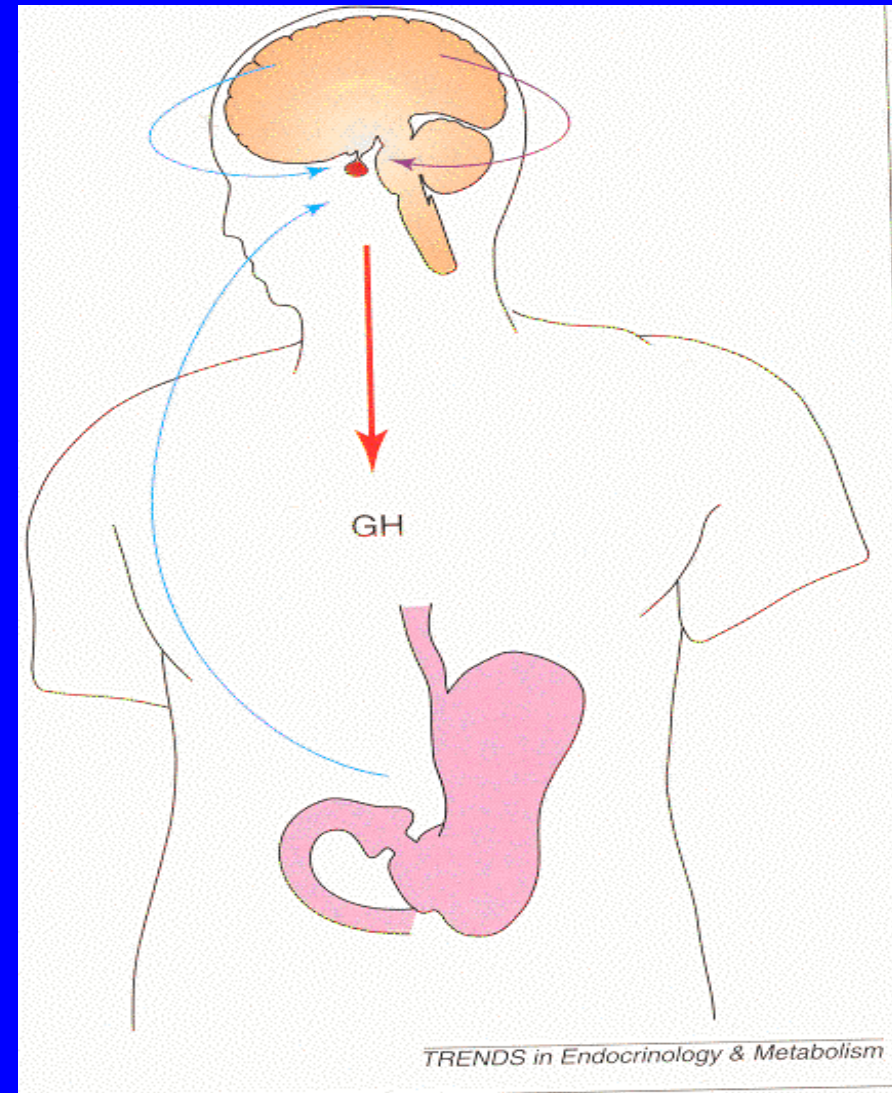
- GH-releasing hormone (GHRH) stimulates GH release through binding to the **GHRH receptor** (blue) and increasing cAMP levels
- GH secretagogues (GHSs) stimulate GH release through the **GHS receptor** (red) to increase intracellular calcium levels. The endogenous ligand for the GHS receptor was not known until the discovery of ghelin



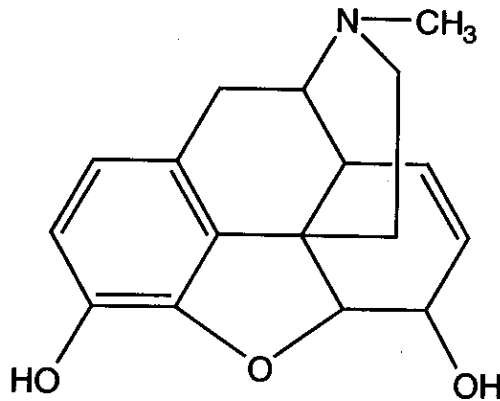
# REGULATION OF GROWTH HORMONE RELEASE FROM THE PITUITARY BY GHRELIN

GH release from the pituitary is controlled by

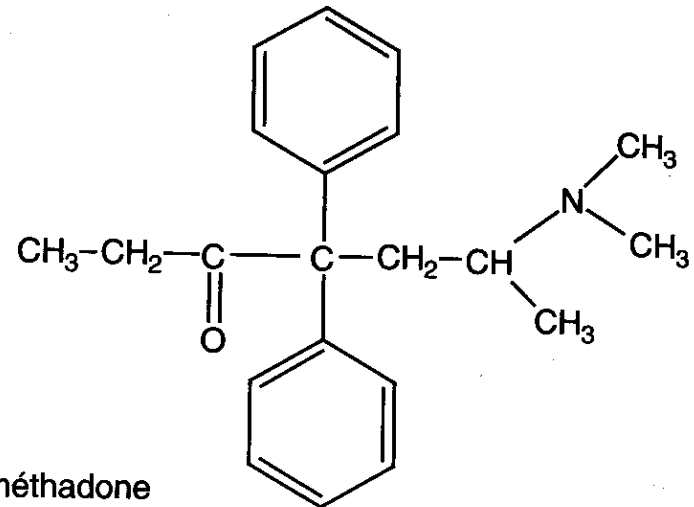
- GH-releasing hormone (purple arrows)
- ghrelin (blue arrows) produced by stomach and hypothalamus



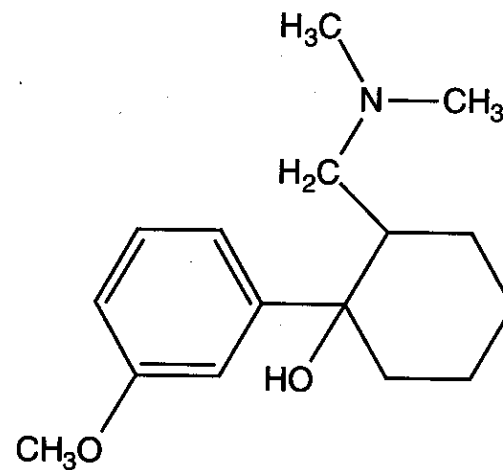
# NEUROPEPIDES OPIOIDES – AGONISTES DES RECEPTEURS OPIOIDES



morphine

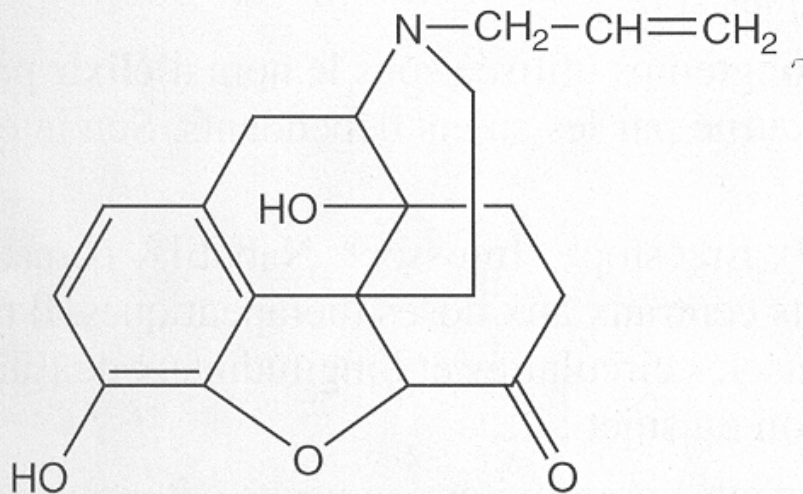


méthadone

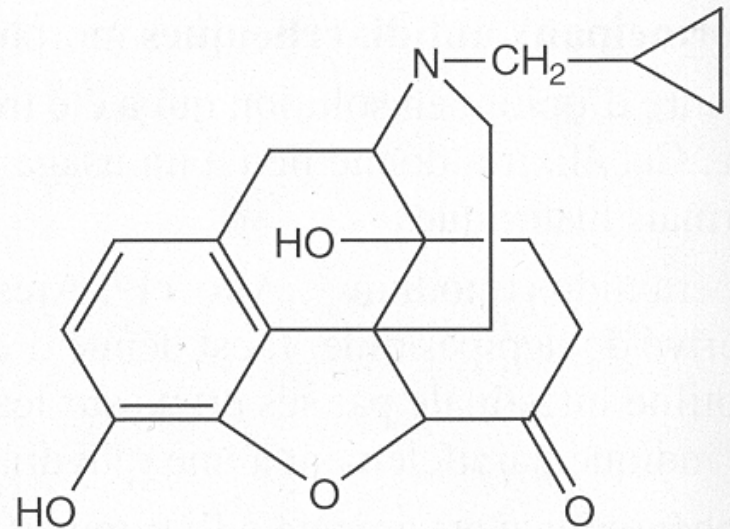


tramadol

# NEUROPEPIDES OPIOIDES – ANTAGONISTES DES RECEPTEURS OPIOIDES

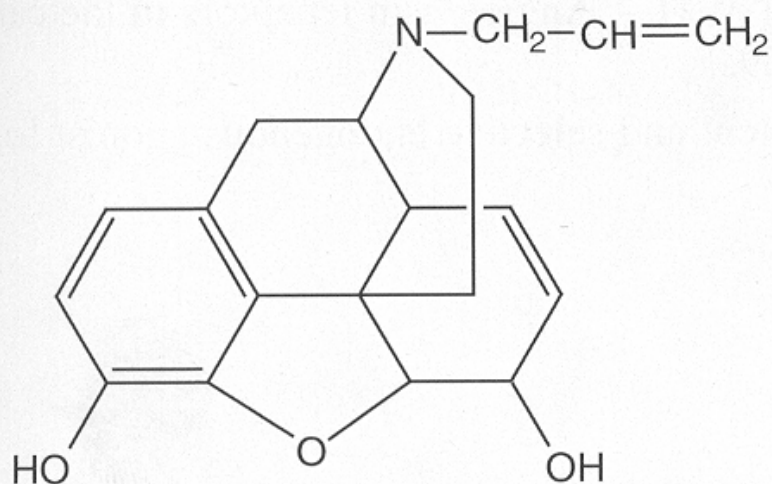


naloxone

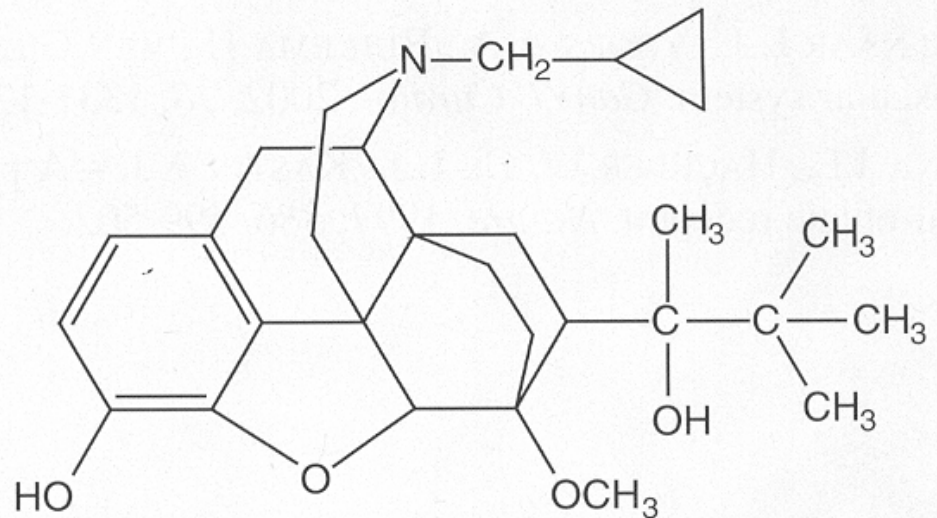


naltrexone

# NEUROPEPIDES OPIOIDES – AGONISTES PARTIELS DES RECEPTEURS OPIOIDES



nalorphine



buprénorphine

# **NEUROPEPTIDE SYSTEMS AS NOVEL THERAPEUTIC TARGETS FOR DEPRESSION AND ANXIETY DISORDERS**

**SUBSTANCE P  
CORTICOTROPIN-RELEASING FACTOR  
VASOPRESSIN  
NEUROPEPTIDE Y  
GALANIN**



# NEUROPEPTIDE SYSTEMS AS NOVEL THERAPEUTIC TARGETS FOR DEPRESSION AND ANXIETY DISORDERS

Pharmacological treatments of mood and anxiety disorders

Monoamine reuptake inhibitors

Benzodiazepine

**BUT...**

- Delayed onset of therapeutic action
- Significant non-responsive patients
- Adverse side-effects
  - nausea, sexual dysfunction, sweating...) with monoamine reuptake inhibitors
  - sedation, cognitive impairment and dependence with benzodiazepine

**NOVEL TREATMENT APPROACHES ~ NEUROPEPTIDES**

# **NEUROPEPTIDE SYSTEMS AS NOVEL THERAPEUTIC TARGETS FOR DEPRESSION AND ANXIETY DISORDERS**

**Neuropeptides demonstrate important functional interactions with monoamines but their effects go beyond modulation of these neurotransmitters**

**Neuropeptides possess a more discrete neuroanatomical localization than monoamines and GABA, and thus might be expected to produce relatively little disruption of normal physiology**

**Neuronal release of neuropeptides requires higher stimulation frequencies than that required by monoamine transmitters colocalized in the same neuron**

# SUBSTANCE P

## Characterization

- 11 amino acid peptide belonging to the tachykinin family
- Mediates its biological activity through G-protein-coupled tachykinin receptors

## Neuropeptide receptor targets for stress-related disorders

NK1 receptor antagonists

    MK0869 Aprepitant (phase III clinical trials)

NK2 receptor antagonists

# SUBSTANCE P

## Discovery of its importance in stress-related disorders

### Neuroanatomical studies

- The concentration of substance P is altered in neural circuits involving the amygdala, hypothalamus, hippocampus and periaqueductal gray
- Substance P has a unique anatomical relationship with monoamine neurotransmitters through which clinically used antidepressant drugs mediate their therapeutic effects

### Pharmacological blockade or genetic deletion

### Pharmacological effect

Central injection of substance P elicits a range of behavioral and cardiovascular stress responses in rats and guinea-pigs

# CORTICOTROPIN-RELEASING FACTOR

- **41 amino acid neuropeptide**
- **Initiates the hypothalamic-pituitary-adrenal axis response to stress**
- **Four CRF-related peptides**
- **Two G-protein-coupled CRF receptor subtypes (CRF 1/2)**
- **CRF1 receptor antagonists might be capable of blocking pathological CRF-mediated stress responses without producing unwanted side effects caused by a general suppression of hypothalamic-pituitary-adrenal axis activity**



# VASOPRESSIN

- Nonapeptide
- Synthesized in the PVN and supraoptic nucleus
- Exerts its effect via a dense localization of vasopressin receptors expressed mainly in limbic areas and in the hypothalamus suggesting that vasopressin might exert a modulatory role in responses to stress
- Vasopressin receptor antagonists might represent potential agents for the treatment of depression and anxiety disorders

# NEUROPEPTIDE Y

- Abundantly expressed in numerous brain areas, including the locus coeruleus, hypothalamus, amygdala, hippocampus, nucleus accumbens, and neocortex
- colocalizes with noradrenaline, GABA
- Actions mediated through heterogeneous G-protein coupled receptors, among which Y1, Y2 and Y5 receptor subtypes mediate CNS effects
- Y1 and Y5 receptor activation is anxiolytic
- Y2 receptor activation is anxiogenic like

# **GALANIN**

**29-30 amino acid neuropeptide**

**Coexists with noradrenaline in locus coeruleus and with 5-HT in the DRN**

**Found (with its receptors) in limbic regions**

**Three known G-protein-coupled galanin receptor subtypes (GAL1, GAL2, GAL3)**

**Blocking the inhibitory effects of galanin on monoamin neurotransmission with galanin receptor antagonists would be predicted to mimic or augment the action of antidepressants**

# BOMBESIN AND SIGNAL TRANSDUCTION

