Summary

The aminoacid γ-aminobutyric acid (GABA) occurs throughout the brain and spinal cord and its role as a major inhibitory neurotransmitter is generally accepted. Though several human disease states are claimed to involve a deranged GABA metabolism, the causal relationship between such a dysfunction and these disorders has not been clearly established. The aim of this thesis was to obtain more information about the occurrence of GABA in the brain and about the regulation of GABA activity by other neurotransmitter systems, especially within the nigrostriatal system. In chapter I we describe a simple and rapid semiautomated assay for GABA in central nervous tissue. GABA is isolated from tissue extracts on small CM Sepharose Cl 6B columns and detected fluorimetrically with o-phthalaldehyde using a continuous flow system (Autoanalyser). The GABA content of several macrodissected rat and mouse brain areas was measured. Administration of 3-mercapto-propionic acid (MPA) to the animals proved to be as effective as microwave irradiation to prevent the post mortem rise in cerebral GABA levels. Using the MPA fixation technique and a slightly improved GABA assay, we measured the GABA distribution within the spinal cord and nucleus caudatus and the GABA content of approximately 70 discrete rat brain nuclei (chapter II). The highest concentration of GABA in the caudate nucleus was found in the ventrocaudal region. GABA appeared to be rather uniformly distributed in the rat brain. The concentration varied from 1 to 90 nmol/mg protein. The lowest amounts of GABA were seen in some hypothalamic nuclei, globus pallidus, eminentia mediana and an extremely high concentration in the zona reticulata of the substantia nigra. Microdissection of the latter nucleus after MPA and/or microwave fixation revealed that microwave irradiation results in a artefactual low GABA concentration. To obtain some direct information about the GABA activity we measured the in vivo release of GABA into a perfusion medium that was pumped through the substantia nigra, using the push-pull cannula technique (chapter III). The GABA content of the perfusate was analyzed with high pressure liquid chromatography coupled with fluorimetric detection, that is essentially the same as that described in chapter I. The major part of the GABA found in the nigral perfusate was of neuronal origin, since the release of this aminoacid was enhanced by potassium in a calcium dependent fashion and by electrical stimulation of the stratum. Moreover, the release was decreased after blockade of the GABA synthesis or in the presence of tetrodotoxine and increased after inhibition of the degradation of GABA. Intranigral application of dopamine (DA) or apomorphine produced biphasic changes in the rate of endogenous GABA release from the substantia nigra (chapter IV). Higher concentrations of DA (≥ 50 μM) resulted in a diminished GABA release, while higher concentrations apomorphine (100 μM) produced a strongly increased GABA output. Both these effects were antagonised by the presence of haloperidol (5 μM), which itself had no effect. Both the DA agonist ADTN and dibutyryl cyclic AMP mimicked the inhibitory effect of 50 μM DA. These results suggest that dopaminergic processes in the substantia nigra affect GABAergic neurotransmission and that DA and apomorphine have different effects on GABA release. Application of the push-pull cannula technique to the striatum, that receives a dopaminergic input from the substantia nigra, revealed that the release of endogenous GABA in this nucleus is of neuronal origin too (chapter V). The decreased GABA output after local application of the GABA agonist muscimol, that was antagonised by picrotoxin suggests a local GABAergic feedback control. Though the cholinomimeticum oxotremorine slightly increased the striatal GABA release, this effect was blocked both by atropine and haloperidol. The latter two drugs did not affect the rate of GABA release themselves. The morphine induced inhibition of striatal GABA release was counteracted by nalorphine but not influenced by haloperidol. Serotonin did not result in any change of striatal GABA release. Chapter VI deals with the dopaminergic regulation of GABA release in the striatum. Local application of DA produced an inhibition of the striatal GABA release, that was reversed by addition of haloperidol. Apomorphine decreased the GABA release at low concentrations (10 μM) while at higher concentrations the drug was ineffective. We showed that amphetamine influences GABA release independent of the presence of DA or noradrenaline (NA), since the inhibitory action of amphetamine was not affected by pretreatment of the rats with reserpine and -methyl-p-tyrosine which strongly reduces the levels of DA and NA. In addition to DA,