areas (spinal cord, brain stem) in which a high affinity uptake for glycine was described (Johnston & Iversen, 1971; Snyder *et al.*, 1973) and where glycine acts as a neurotransmitter (Aprison, Davidoff & Werman, 1970; Curtis & Johnston, 1970).

In conclusion, a process of exchange accounts, at least in large part, for what has been interpreted as high affinity uptake of GABA, and perhaps of other putative neurotransmitter amino acids.

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On the release of accumulated [^{3}H]-γ-aminobutyric acid (GABA) from isolated rat superior cervical ganglia.

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Exogenous GABA is accumulated by glial cells in rat superior cervical ganglia (Bowery & Brown, 1972; Young, Brown, Kelly & Schon, 1973). Some preliminary observations (Bowery & Brown, 1972) suggested that labelled GABA taken up by this tissue could be released by 'depolarizing' stimuli. In the present experiments we have examined this phenomenon more closely.

Superior cervical ganglia (with pre- and post-ganglionic trunks) were excised from male Wistar rats (anaesthetized with urethane 1.5 gm/kg), desheathed, and incubated for 2-3 h in [^{3}H]-GABA (0.1-20 μM) in the presence of amino-oxyacetic acid (AOAA, 10 μM). The ganglia were then superfused at 0.5 ml/min with Krebs solution at ~22° C in such manner that the effluent radioactivity from the entire preparation or from individual regions (pre-ganglionic nerve trunk, post-ganglionic trunk or ganglion soma) could be collected at 2 min intervals and measured.

After 15-20 min superfusion of the entire preparation the release of tritium assumed a single exponential over several hours with a rate coefficient of 0.0014 ± 0.0002/min (mean ± s.e. of 27 ganglia). This value was independent of the ganglionic [^{3}H]-GABA concentration between 2 and 364 μM. In the presence of AOAA, released tritium (resting and stimulated) as well as accumulated tritium (Bowery & Brown, 1972; Walsh, Bowery, Brown & Clark, 1974) corresponded to >95% [^{3}H]-GABA. This rate coefficient for release was increased (a) by raising external [K^+] (the increment in rate coefficient increasing with $[K]_o$ over the range 30-140 mM); and (b) by applying electrical stimuli to the ganglion soma or pre-ganglionic nerve trunk (threshold 0.5 Hz, 0.1 ms duration).

These 'depolarizing' stimuli are comparable to those previously found to release GABA from presumed neural loci in brain tissue (Srinivasan, Neal & Mitchell, 1969; Katz, Chase & Kopin, 1969). Accelerated release following orthodromic stimulation is especially interesting because such stimuli also produce depolarization of ganglionic

satellite cells (Adams & Brown, 1974). This raises the question whether release of GABA from such cells results from neural excitation. Several observations suggest that this may not be so. (a) Release following electrical stimulation was not prevented when transmission and/or conduction were blocked by (i) 0 mM [Ca^{2+}], (ii) 0 [Ca^{2+}] + 10-30 mM [Mg^{2+}], (iii) 0 mM [Ca^{2+}] + 1 mM [EDTA] and (iv) 3 mM procaine or 300 μM amethocaine. (b) Carbachol (550 μM), which depolarizes ganglionic neurones but not glial cells (Adams & Brown, 1974), did not accelerate [^{3}H]-GABA release. (c) When the effluent radioactivity from the trunks and soma was measured separately during pre-ganglionic trunk stimulation, an increased efflux rate from the pre-ganglionic trunk but *not* from the soma was detected. In contrast, accumulated [^{3}H]-choline *was* released from the soma following pre-ganglionic stimulation.

Thus, release of GABA from ganglionic glial cells by 'depolarizing' stimuli appears to result from a direct and local action on the glial cells *per se*

This investigation was supported by a grant from the Medical Research Council.

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The effects of depleting brain amines on the behavioural actions of γ-hydroxybutyric acid

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It has been well-established that γ-hydroxybutyric acid (GHB) produces a dose-dependent increase in the concentration in the brain of dopamine (Gessa, Vargiu, Crabai, Boero, Caboni & Camba, 1966; Roth & Suhr, 1970; Hutchins, Rayevsky & Sharman, 1972; Clifford, Taberner, Tunnicliff, Rick & Kerkut, 1973; Stock, Magnusson & Anden, 1973) and serotonin (Clifford *et al.*, 1973; Spano & Przegalinski, 1973). There is a close correlation between the duration of the sleep-like state induced by GHB and the increase in dopamine level (Gessa *et al.*, 1966; Clifford *et al.*, 1973), although no causal relationship has been

Table 1 Effects of 6-hydroxydopamine on brain amine levels and γ-hydroxybutyric acid (GHB) sleeping time

Dose schedule	*Whole brain amine levels (µg/g wet weight)*			*Sleeping time (min)*
	Dopamine	*Noradrenaline*	*Serotonin*	
Control (saline)	0.936 ± 0.104	0.569 ± 0.042	0.485 ± 0.063	–
GHB (4 mmoles/kg i.p.)	1.23 ± 0.14***	0.446 ± 0.073	0.688 ± 0.151**	58.9 ± 7.8
6-Hydroxydopamine (0.8 µmoles)	0.197 ± 0.130	0.074 ± 0.017	0.573 ± 0.392	–
GHB (4 mmoles/kg i.p.)	0.142 ± 0.054	0.123 ± 0.057	0.676 ± 0.094	24.3 ± 4.9*

* $P < 0.001$; ** $P < 0.05$; *** $P < 0.01$.

Results are the means ± s.d. of at least 5 determinations. Mice were injected intraventricularly with either 6-hydroxydopamine or vehicle in a volume of 5 µl 72 h prior to assay of brain amines or determination of the GHB sleeping time. Amine levels were determined in GHB-treated mice 40 min after the injection of GHB.