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THE RELATIONSHIP BETWEEN PEAK VELOCITY OF SACCADIC EYE MOVEMENTS AND SERUM BENZODIAZEPINE CONCENTRATION

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1 Six healthy male volunteers received single oral doses of 10 mg diazepam, 20 mg temazepam, 15 mg flurazepam, 5 mg nitrazepam, 10 mg desmethyl-diazepam and placebo in a double-blind randomized fashion.

2 Peak velocity of saccadic eye movements, serum benzodiazepine concentration, and subjective ratings of wakefulness and co-ordination were measured at intervals up to 12 h after drug administration.

3 All active treatments produced a statistically significant decrease in peak saccadic velocity. The effect of temazepam and diazepam was generally more pronounced than that of flurazepam, nitrazepam and desmethyl-diazepam.

4 There were log-linear correlations between peak saccadic velocity and serum benzodiazepine concentration after ingestion of temazepam, diazepam and nitrazepam.

5 These results demonstrate a clear relationship between serum benzodiazepine concentration and its effect on a convenient measure of brainstem reticular formation function.

Introduction

Recent reviews of the pharmacological properties of benzodiazepine drugs in man have emphasized the lack of evidence for specific relationships between dose or serum benzodiazepine concentration and the clinical response (Mandelli *et al.*, 1978; Bellantuono *et al.*, 1980). This has been thought to be largely due to kinetic properties such as the existence of active metabolites (Mandelli *et al.*, 1978) and differences in the rate of distribution in different areas of the brain (Morselli *et al.*, 1973). The probable development of tolerance (Greenblatt & Shader, 1978) and the multitude of clinical effects of these compounds may also play a role (Garattini *et al.*, 1973; Lader, 1978).

Many of the methods used to monitor the effects of benzodiazepine therapy, such as subjective analogue rating scales, choice reaction time, finger-tapping rate and visuo-motor co-ordination in highly trained individuals (Bond & Lader, 1972; Clarke & Nicholson, 1978; Nicholson, 1978; Wittenborn, 1979) are to a large extent dependent on the co-operation of the subjects, and assess a combination of the assumed behavioural effects of these drugs. In general they measure complex behavioural patterns which must be controlled by various neurophysiological systems working in conjunction (Ganong, 1975).

We have developed a technique to quantify objectively the peak velocity of horizontal saccadic eye movements in man, based on the technique of Baloh *et al.* (1975) and on evidence suggesting that peak saccadic velocity is an indicator of the function of a reasonably well-defined group of neurones in the pontine reticular formation (Luschei & Fuchs, 1972; Keller, 1974). We explored the effect on peak saccadic velocity of diazepam, temazepam, nitrazepam and flurazepam, all 1,4-benzodiazepines with different pharmacokinetic profiles and different clinical indications (Bellantuono *et al.*, 1980). Desmethyldiazepam was tested in addition because it is an active metabolite of diazepam, clorazepate, prazepam and probably other widely used benzodiazepine compounds (Kaplan *et al.*, 1973; Greenblatt *et al.*, 1978; Mandelli *et al.*, 1978; Breimer, 1979).

Relevant eye movement physiology

In most real-life situations head movement plays a major role, and complex vestibulo-ocular interactions take place (Bittencourt *et al.*, 1980a). The following considerations apply only to conjugate horizontal eye movements and to situations in which

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the head is stationary, the peripheral vestibular system being therefore inactive. There are basically two types of conjugate eye movements used in day to day activities. Smooth-pursuit eye movements are used to track objects moving slowly across the visual fields (Bittencourt *et al.*, 1980), for example a driver waiting at the traffic lights and following a pedestrian crossing the street. Saccades are used to switch the eyes quickly from one point of interest to the next (Weber & Daroff, 1971), for instance from the pedestrian to the light changing to green. With vergence movements the eyes move disconjugately in order to re-fixate from near to far objects or *vice-versa* (Carpenter, 1977). During smooth-pursuit the eyes can reach velocities of up to 100°/s (Bittencourt *et al.*, 1980a). Saccades are much faster, reaching velocities of up to 700°/s (Baloh *et al.*, 1975), but contrary to smooth-pursuit, vision is not perfect during a saccade (Gresty *et al.*, 1976).

In order to make a saccade, the eyes take off from one position, accelerate rapidly to achieve maximum velocity early in the saccade and decelerate to reach the desired target. As the peak velocity reached during a saccade increases exponentially with the amplitude of the eye movement, the only reliable way of assessing peak saccadic velocity accurately is by studying the velocity-amplitude relationship. A further advantage of such an approach is that it does not depend on subject co-operation. If he tries to slow his saccades the amplitude will be decreased, but the velocity-amplitude relationship is maintained (Becker & Fuchs, 1970).

Although areas in the cerebral hemispheres may be involved in generating the motor command for saccade initiation (Hoyt & Frisén, 1975), and the cerebellum and hemispheres are involved in providing saccade accuracy (Baloh *et al.*, 1978), the peak velocity of saccadic eye movements, as well as their duration, are the function of the so-called saccade pulse-generator, localizing mostly in the pontine reticular formation (Hoyt & Frisén, 1975). Perhaps the most lucid demonstration of this proposed organization was that of Keller (1974), who performed single-unit recordings of the firing-rate of cells in the pontine reticular formation. A sub-group of these, the BURST units, discharged at very high frequencies. The firing-rate increased with the amplitude of the eye movement, and was related to saccade velocity. This relationship was maintained during very slow saccades produced by the animals when drowsy because of lack of environmental stimulation.

Methods

Subjects

Six healthy male volunteers, aged 20–32 (mean 22) years, took part in the study. All were free of ocular pathology, as evidenced by thorough history and physical examination. None were smokers, or needed drug therapy during the course of the study. Informed signed consent was obtained. All were acquainted with their tasks and were given practice sessions (2–4) until no further learning effect could be noticed.

Procedure

Treatments were given orally, in a double-blind randomized fashion, and consisted of 10 mg diazepam (Valium®), 2 × 10 mg temazepam (Normison®), 15 mg flurazepam (Dalmane®), 5 mg nitrazepam (Mogadon®), 10 mg desmethyl-diazepam (in capsules, dispersed in 100 mg lactose) or placebo (to match temazepam).

Subjects were requested not to ingest alcohol 12 h before and 24 h after each trial day. They were allowed a breakfast of fruit juice and toast, one hour before arriving at the laboratory at 08.30 h. An indwelling cannula was inserted upon arrival and a 10 ml blood sample was taken. The eye movement tests and analogue rating scales were performed and the oral treatment was then given with 50 ml water. The same sequence (blood sample, eye movement test and analogue rating scales) was repeated 0.5, 1, 1.5. 2, 3, 4, 6, 9 and 12 h after the treatment. Lunch was allowed at 4 h, and caffeinated drinks at 6 h. Subjects were tested at 1–4 week intervals, over a period of 4 months.

The subjective analogue rating scales consisted of a horizontal line, 100 mm in length, with the words 'sleepy' and 'wide-awake' (to measure wakefulness) and 'clumsy' and 'well co-ordinated' (to measure co-ordination) as ratings extremes. Blood (10 ml) was collected in disposable syringes, allowed to clot for at least 1 h in glass tubes and the serum was stored deep-frozen (−20°C) for subsequent benzodiazepine measurement by gas-liquid chromatography, using a Hewlett-Packard 5710 A gas chromatograph fitted with an electron capture detector. Flurazepam and desalkyl-flurazepam were measured following the method of de Silva *et al.* (1974). Nitrazepam, diazepam and desmethyl-diazepam were measured using the method of de Silva *et al.* (1976) while temazepam was measured by the method of Belvedere *et al.* (1972).

Eye movement test

The method described by Baloh *et al.* (1975) was substantially modified in order to decrease test duration to around 3 min and to increase the signal to noise ratio.

Constant ambient illumination of dark red light was maintained in the sound-attenuated laboratory. Stick-on silver-silver chloride electrodes were placed lateral to both outer canthi, with a similar reference

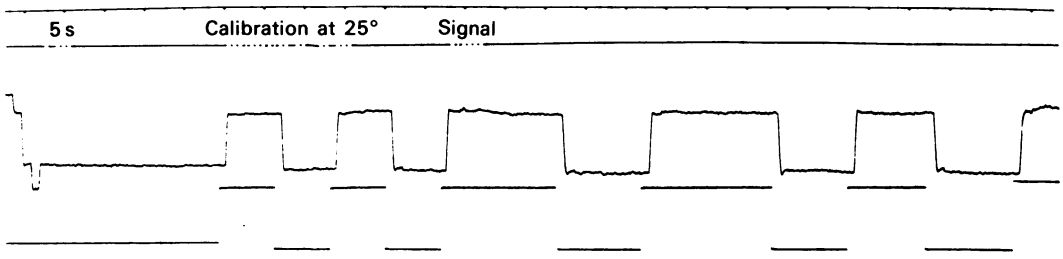

Figure 1 *Eye movement test.* The calibration stimuli (lower trace), placed at fixed intervals, are followed by test stimuli, at random intervals. These produce light spot displacements on the screen subtending an angle of 15° in relation to the nasion. The stimuli then change to 20° excursions. Two other sets of stimuli follow, one for amplitudes of 25° and 30° (shown here), another for 35° and 40°, separated by 30 s intervals. The resulting eye movements (upper trace) are largely saccadic in nature.

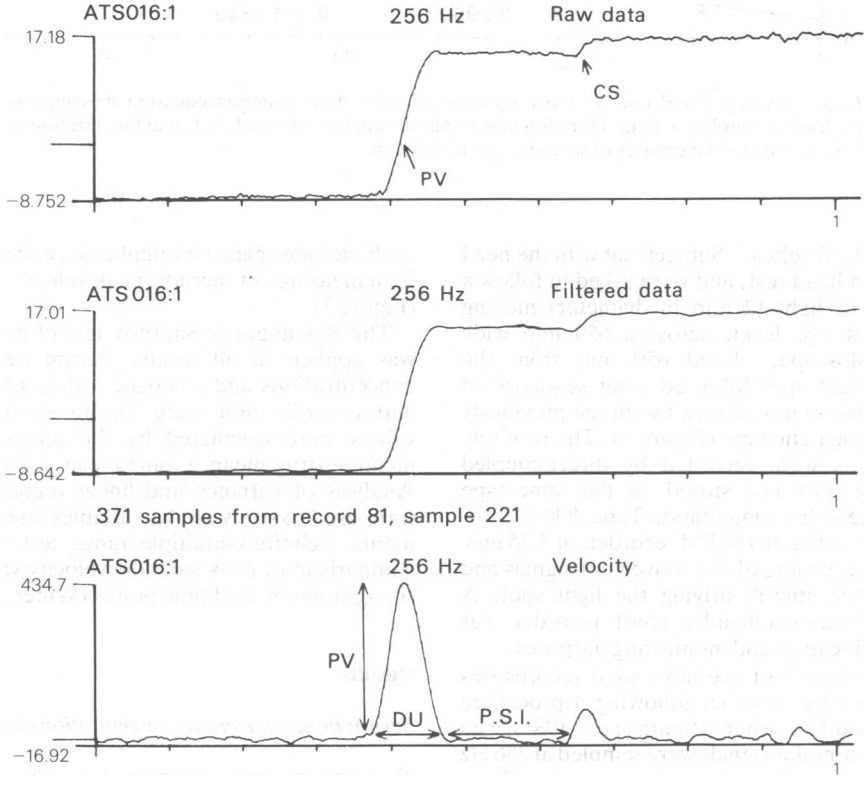

Figure 2 *Computer analysis of a saccade.* The eye position signal (top) is smoothed (middle) and differentiated (bottom). The analysis is carried out in the velocity trace thus obtained. In this case the eyes started off from the left, accelerated to reach peak velocity early in the course of the movement (PV), and slowly decelerated approaching the target. As happens in many normal saccades, the eyes did not quite reach the target, and a corrective saccade (CS) was necessary after about 200 ms. The amplitude, peak velocity and duration of each saccade are measured.

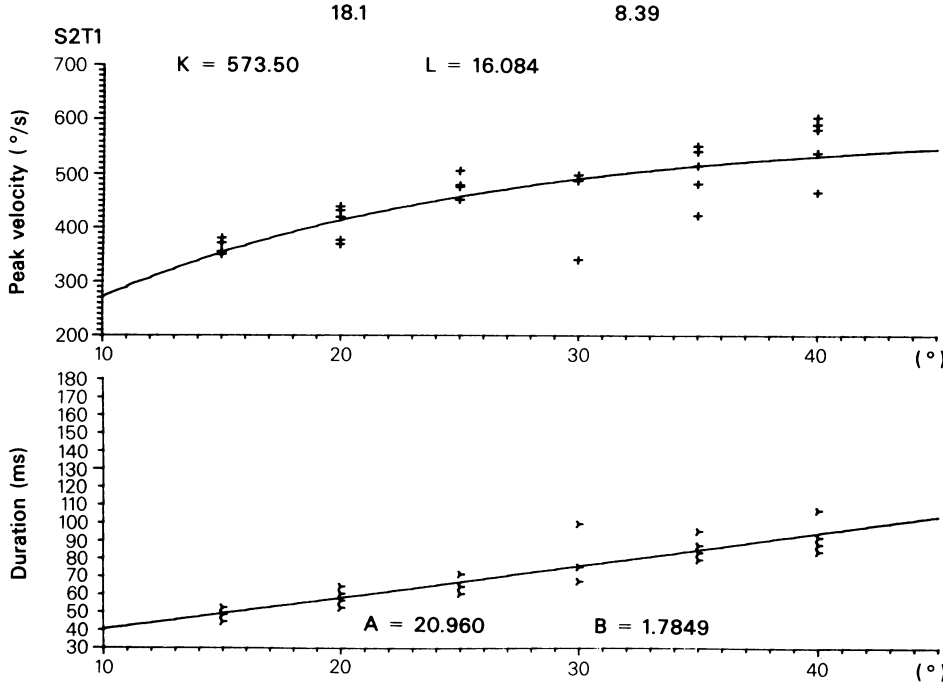

Figure 3 *Final measures of peak velocity and duration of saccades.* An exponential equation of asymptote K is fitted to the peak velocity x amplitude data. Duration and amplitude are linearly related. K and the duration at which the line crosses 30° are the final measures of saccadic eye movements.

electrode on the forehead. Subjects sat with the head restrained by a head rest, and were asked to follow a point source of light (2 mm in diameter) moving horizontally at eye level, across a 650 mm wide Lanelec oscilloscope, placed 650 mm from the nasion. The light spot followed a set sequence of step-like displacements, driven by stimuli previously recorded on magnetic tape (Figure 1). The resulting eye movements were recorded by direct-coupled electro-oculography and stored on the same tape (Scotch Magnetic Instrumentation Tape, 890-0.25 in-2300 PR). An SLE-Labs 84 FM recorder, at 3.75 in/s, was used for recording of eye movement signals and playback of the stimuli driving the light spot. A rectilinear Elema-Schonander chart recorder was used for amplification and monitoring purposes.

Computer analysis of eye movement records was carried out blindly, off-line, following a procedure reported in detail elsewhere (Smith *et al.*, 1981). The EOG and the stimulus signals were sampled at 256 Hz by the A/D convertor of a mini-computer (Data General Nova 3D). The stimulus channel was then scanned to determine the location of all eye movements. The amplitude, duration and peak velocity of the detected saccade were calculated (Figure 2). The computer then plotted peak velocity and duration of each saccade against its amplitude, and calculated the final measures of saccadic peak velocity and duration (Figure 3).

The Kolmogorov-Smirnov test of goodness of fit was applied to all results. Serum benzodiazepine concentrations and analogue ratings of wakefulness and co-ordination were log-transformed. Missing values were calculated by the operation (patient mean + drug mean + time mean − 2 grand mean). Analysis of variance and linear regression analysis were used to study all the variables after all six treatments. Scheffe's multiple range test was used for comparison of peak saccadic velocity values after all six treatments at all time points (Winer, 1972).

Results

Serum benzodiazepine concentrations (Table 1)

Temazepam and diazepam were absorbed rapidly, peak concentrations being reached 90 min after ingestion of both compounds. Temazepam was rapidly eliminated, three out of six subjects having no detectable levels at 12 h. Low desmethyl-diazepam concentrations were present 12 h after diazepam,

Table 1 Number of samples (6 unless otherwise indicated in brackets) and mean ± s.d. serum benzodiazepine concentrations found after oral administration of diazepam (10 mg), temazepam (20 mg), nitrazepam (5 mg) and desmethyl-diazepam (10 mg).

Nine hours after diazepam, 2 samples contained trace amounts, and one 10 ng/ml of desmethyl-diazepam. At 12 h, all samples contained the metabolite (12.5 ± 4 ng/ml).

	Serum concentration (ng/ml)											
	Diazepam		*Temazepam*			*Nitrazepam*			*Desmethyl-diazepam*		
Time (h)	*Mean*	*s.d.*	*Mean*	*s.d.*		*Mean*	*s.d*		*Mean*	*s.d.*	
0.5	98	54	127	60		13		(1)	8	4	(2)
1	182	86	269	197		24	25		6	5	(5)
1.5	238	77	307	232		25	16	(5)*	9	5	
2	223	43	203	103		30	16		11	3	
3	189	63	151	70		26	7		11	2	
4	148	50	119	64		19	6		6	3	
6	94	24	93	62		12	4		6	1	(4)
9	69	27	57	38		11	6	(5)	5		(1)
12	56	26	42	23	(3)	7	2	(4)			(0)

* indicates missing sample.

when the concentration of the parent compound was found to be about five times that of the metabolite. No desmethyl-diazepam or nitrazepam could be detected 30 min after administration of these compounds. Low peak concentrations were found at 2–3 h for both compounds. Desmethyl-diazepam was more rapidly eliminated, so that in only one subject could the drug be detected at 9 h, while 9 h after nitrazepam, serum concentrations were detectable in four out of six subjects. After oral administration of 15 mg flurazepam, measurable concentrations of the parent drug and the desalkyl metabolite were seen in only a few samples. As a result, they will not be used in the following analysis of the results.

Subjective analogue ratings of wakefulness and co-ordination (Figure 4)

Analysis of variance showed significant increases in 'sleepiness' ($P < 0.05$) after all five active treatments, but not placebo. These changes were particularly prominent between 30 min and 3 h after ingestion of the treatment.

There were significant increases in the ratings of 'clumsiness' ($P < 0.05$) after ingestion of diazepam, temazepam, nitrazepam, and desmethyl-diazepam, but not flurazepam or placebo. These changes were more pronounced between 30 min and 3 h after ingestion of the treatment.

Peak saccadic velocity

No significant changes were noticed after placebo. Learning effects could not be observed either by comparison of baseline values in successive days, or within the placebo days (Figure 5).

All five active treatments produced significant

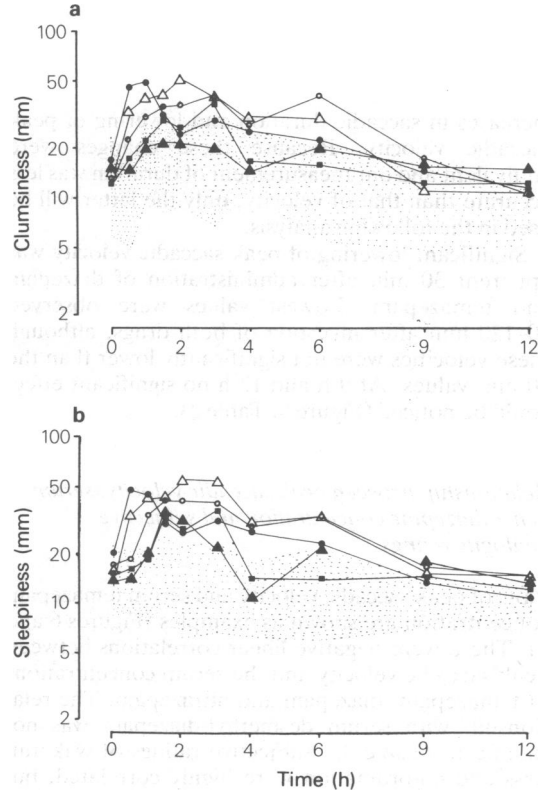

Figure 4 Subjective analogue ratings of clumsiness (a) and sleepiness (b) after oral administration of 10 mg diazepam (△), 20 mg temazepam (●), 15 mg flurazepem (▲), 5 mg nitrazepam (■), 10 mg desmethyl-diazepam (○) and placebo (∴). The shaded area denotes ± 1 s.d. of placebo, while other plotted values are means across the six subjects.

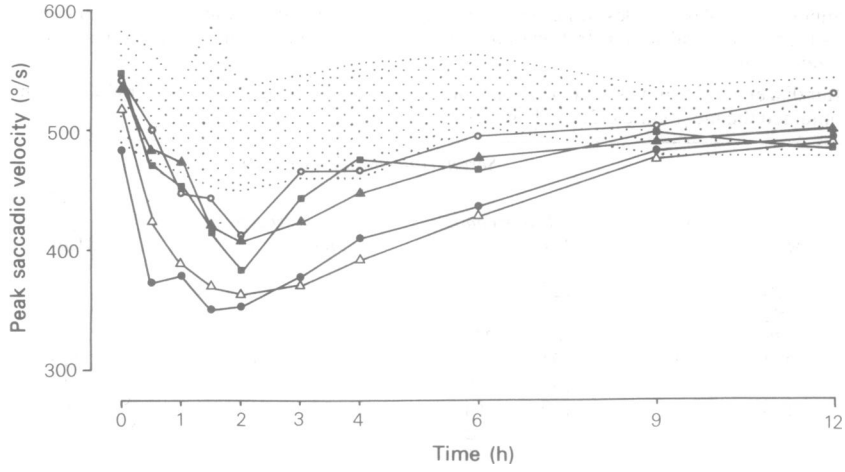

Figure 5 Peak saccadic velocity after administration of 10 mg diazepam (△), 20 mg temazepam (●), 15 mg flurazepam (▲), 5 mg nitrazepam (■), 10 mg desmethyl-diazepam (○), and placebo (∴). The shaded area denotes ± 1 s.d. of placebo. Other values plotted are means across the six subjects.

increases in saccadic duration and lowering of peak saccadic velocity. Because these changes were equivalent and our measurement of duration was less accurate than that of velocity, only the latter will be used in the following analysis.

Significant lowering of peak saccadic velocity was apparent 30 min after administration of diazepam and temazepam. Lowest values were observed 90–120 min after ingestion of both drugs, although these velocities were not significantly lower than the 30 min values. At 9 h and 12 h no significant effect could be noticed (Figure 5, Table 2).

Relationship between peak saccadic velocity, serum benzodiazepine concentration and subjective analogue ratings

Plots of peak saccadic velocity and serum temazepam concentration are shown as examples (Figures 6 and 7). There were negative linear correlations between peak saccadic velocity and the serum concentrations of temazepam, diazepam and nitrazepam. The relationship with serum desmethyl-diazepam was not significant (Table 3). Subjective ratings of wakefulness and co-ordination were highly correlated, but the relationship of each with serum benzodiazepine concentration was poor. After nitrazepam and diazepam there were relatively strong linear correlations between peak saccadic velocity and both subjective ratings, while after placebo, temazepam, flurazepam and desmethyl-diazepam a linear relationship was not observed.

Discussion

Many studies have emphasized the role of the brainstem reticular formation in the regulation of consciousness (Moruzzi & Magoun, 1949) and eye movements (Lorente de Nó, 1933; Luschei & Fuchs, 1972). It has been demonstrated that during periods of drowsiness monkeys make slower saccades, the velocity of which is strongly related to the discharge-rate of a well-defined group of neurones (burst units) in the para-medial pontine reticular formation (Keller, 1974). The same author has been able to monitor the animals' 'level of alertness' using both peak saccadic velocity and the discharge-rate of the burst units (Keller, 1977).

There is a large body of evidence suggesting a close association between the generation of saccadic eye movements and of certain aspects of sleep. Partial lesions of the raphé nuclei abolish slow wave sleep selectively and after their total destruction animals do not sleep (Simon, Gershon & Brooks, 1973). REM sleep has been found to be abolished in the cat after bilateral pontine tegmental lesions (Jones, 1979) and in 'locked-in' patients (Adams, 1959) shown to have lesions of the pontine tegmentum (Cummings & Greenberg, 1977; Markand & Dyken, 1976). Furthermore, neurones in the para-medial and dorsolateral pontine reticular formation have been shown to be responsible for generation of REM sleep (Jouvet 1967). Additional evidence has been provided by a recent report that patients with spinocerebellar degeneration and very slow saccades (10–20°/sec) have absent REM sleep (Osorio &

Table 2 Summary of statistically significant decreases in peak saccadic velocity after all five active treatments (column on the left) when compared against each other and against placebo between 0.5 h and 6 h after ingestion of the treatments (other columns).

There were no significant decreases in peak saccadic velocity at 9 h and 12 h. Scheffe's multiple range test was used at $P < 0.05$.

	Time (h)						
	0.5	*1*	*1.5*	*2*	*3*	*4*	*6*
Temazepam	flurazepam	flurazepam	flurazepam				
	nitrazepam	nitrazepam	nitrazepam		nitrazepam	nitrazepam	
	d-diazepam	d-diazepam	d-diazepam	placebo	d-diazepam	placebo	placebo
	placebo	placebo	placebo		placebo		
Diazepam	d-diazepam	flurazepam	d-diazepam		nitrazepam	nitrazepam	
	placebo	d-diazepam	placebo	placebo	d-diazepam	d-diazepam	placebo
		placebo				placebo	
Flurazepam	NS	NS	placebo	placebo	placebo	NS	NS
Nitrazepam	NS	NS	placebo	placebo	NS	NS	NS
D-diazepam	NS	NS	placebo	placebo	NS	NS	NS

NS = not significant. d-diazepam = desmethyl-diazepam.

Daroff, 1980). These findings taken together strongly suggest that the generation of saccadic eye movements, particularly the control of peak saccadic velocity, shares to a large extent the same pontine circuit with the generation of REM sleep (Osorio & Daroff, 1980).

Suppression of REM sleep is a characteristic effect of hypnotic compounds, particularly benzodiazepines, and is possibly related to their sleep-inducing properties (Feinberg *et al.*, 1979). Accurate assessment of peak saccadic velocity therefore provides reliable information on the function of a well-defined group of cells in the pontine reticular formation (Keller, 1974; Hoyt & Frisén, 1975) which are closely concerned with the desired effect of benzodiazepines, i.e. sleep.

It should be emphasized that saccade generation is bascially a motor function, since the objective is to move the eyes as quickly as possible between two points. In order that velocities as high as 700°/s can be achieved by the eyes in mid-flight, a highly synchronized discharge-rate is required of the 'burst cells' in the pontine reticular formation, which comprise the immediate pre-motor stage of saccadic eye movements (Hoyt & Frisén, 1975). The muscle-relaxant effect of benzodiazepines is mediated via brainstem reticular formation systems as well (Przybila & Wang, 1968).

We have demonstrated that single oral doses of temazepam (20 mg) and diazepam (10 mg) are equivalent as measured by their effect on peak saccadic velocity until 12 h after administration. Flurazepam (15 mg), nitrazepam (5 mg) and desmethyl-diazepam (10 mg) had a less pronounced effect throughout the 12 h of testing. These findings contrast with the generally accepted equivalence of the benzodiazepine doses used in this study (Calne, 1980; Hollister, 1980).

The serum benzodiazepine concentrations observed are in agreement with previous studies of temazepam (Bittencourt *et al.*, 1979), diazepam (Mandelli *et al.*, 1978), and nitrazepam (Rieder & Wendt, 1973). Desmethyl-diazepam has been more frequently studied after administration of its precursor, potassium clorazepate (Shader *et al.*, 1978). The low concentrations of flurazepam and its metabolite observed are to a large extent due to rapid transformation of the parent compound into a variety of active metabolites (Greenblatt, Shader & Koch-Weser, 1975; Mahon, Inabu & Stone, 1977).

We have also demonstrated negative linear correlations between serum benzodiazepine concentration and the peak velocity of saccadic eye movements, after administration of diazepam, temazepam and nitrazepam. The lack of such a relationship after administration of desmethyl-diazepam is possibly due to its relatively modest effect on peak saccadic velocity, associated with low serum concentrations.

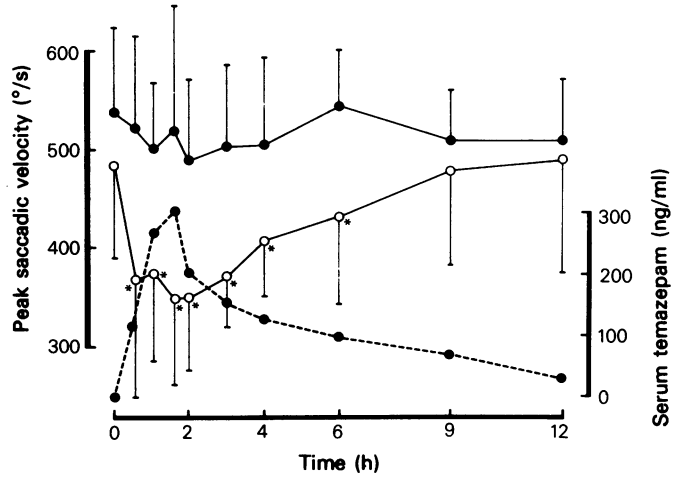

Figure 6 Peak saccadic velocity (○, mean ± s.d.) and serum temazepam concentrations (●---● mean) after oral administration of temazepam 20 mg. Peak saccadic velocity ●, mean ± s.d.) after placebo is also shown. $*P < 0.05$.

These results are qualitatively similar to those observed in relation to smooth-pursuit eye movements (Bittencourt & Richens, 1981) and are further evidence of the lack of discernible pharmacodynamic difference between the compounds studied (Bellantuono *et al.*, 1980). Clinically relevant differences exist in the time-course of the response observed which are to a large extent due to fluctuations in serum concentrations. These results support the view that there is a clear relationship between the serum benzodiazepine concentration and its effect on the brain, when specific aspects of the latter are accurately analysed (Aschoff, 1968; Fink *et al.*, 1976), and conflict with the widely expressed contrary view (Garattini *et al.*, 1973; Mandelli *et al.*, 1978, Bellantuono *et al.*, 1980), for which various pharmacokinetic explanations have been offered (Morselli *et al.*, 1973). Specific pharmacokinetic-dynamic relationships may be difficult to establish, though, in the case of compounds which are extensively metabolized on to various active metabolites even after a single dose, such as flurazepam (Greenblatt *et al.*, 1975).

The effect of diazepam on peak saccadic velocity was studied by Aschoff (1968), who found a direct relationship between the dose of diazepam and the decrease in saccadic velocity. These findings have been partly replicated by other investigators (Gentles & Llwellyn-Thomas, 1971; Frecker & Llewellyn-Thomas, 1972). Although criticisms can now be made of the methodology and conclusions of these studies, it is surprising that they have been ignored by reviewers of the pharmacological properties of benzodiazepines (Mandelli *et al.*, 1978; Bellantuono *et al.*, 1980) and of the effects of benzodiazepines on performance (Wittenborn, 1979), and misplaced as a sensory test in a lengthy review of methods in psychopharmacology (Hindmarch, 1980). In fact, neurophysiological measures have received comparatively little attention in the assessment of drug effects on the human brain, as compared to psychomotor measures (Solomon *et al.*, 1979; Wittenborn, 1979; Hindmarch, 1980), from which serum concentration or dose relationships with the pharmacological response cannot be expected. Measurements are usually of complex behavioural patterns, such as visuo-motor co-ordination (Clarke & Nicholson, 1978), the neurophysiological basis of which is not at all clear

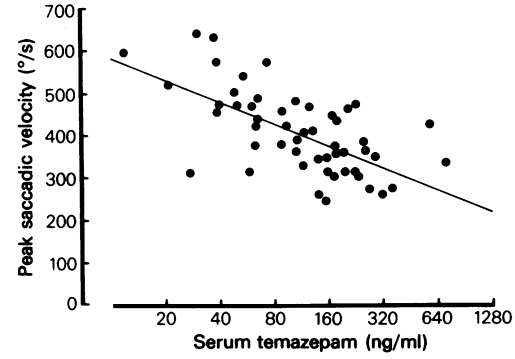

Figure 7 Negative linear correlation between peak saccadic velocity and serum temazepam concentration ($r = -0.631$, $n = 51$, $P < 0.01$). Values plotted are the raw results of Figure 6, irrespective of time.

Table 3 Linear correlation coefficients (r) and number of observations (n) between peak saccadic velocity, serum benzodiazepine concentration and subjective analogue ratings of wakefulness and co-ordination, after all active treatments: (for $n > 42$, $P < 0.01$ when absolute $r < 0.393$).

			Diazepam		*Temazepam*		*Flurazepam*		*Nitrazepam*		*Desmethyl-diazepam*	
			r	n	r	n	r	n	r	n	r	n
		Peak saccadic velocity	−0.49	54	−0.63	51			−0.47	45	−0.27	36
Serum concentration	x	Wakefulness	−0.465	54	−0.405	50			−0.42	44	−0.19	36
		Coordination	−0.495	54	−0.39	50			−0.45	44	−0.32	36
Peak saccadic velocity	x	Wakefulness	0.57	60	0.13	59	0.14	58	0.47	58	0.09	60
		Coordination	0.55	60	0.11	59	0.01	58	0.48	58	0.17	60
Wakefulness	x	Coordination	0.89	60	0.94	59	0.80	59	0.89	59	0.81	60

(Ganong, 1975). These measures may reflect real-life situations more closely, but breaking the behavioural patterns into well-defined neurophysiological measures should provide more reliable assessment of drug effects on CNS pathways, as shown previously (Fink *et al.*, 1976; Bittencourt *et al.*, 1980b). The demonstration of a statistically significant relationship between effect and serum level or dose should be sine qua non for a measure to be considered reliable or specific in term of the effects it is supposed to be measuring.

While there is little doubt that benzodiazepines have, among others, hypnotic effects, many other issues presently under discussion, such as the clinical relevance of their pharmacokinetic properties (Breimer, 1979) and the development of tolerance (Greenblatt & Shader, 1978) can only be studied by accurate measurement of specific aspects of brain function and by comparisons of these measures with pharmacokinetic data.

In the present study, the use of analogue rating scales has been restricted to looking into the relationship between subjective ratings, serum benzodiazepine concentration and peak saccadic velocity. The relationship of peak saccadic velocity to both ratings was contradictory, in that it was relatively strong after diazepam and nitrazepam, and absent after temazepam, flurazepam and desmethyldiazepam. After all treatments, serum benzodiazepine concentration was not clearly related to the subjective ratings, in contrast to the more definite correlation between peak saccadic velocity and serum benzodiazepine concentration. Subjective ratings of wakefulness correlated highly with those of co-ordination, and the correlation coefficient increased with the magnitude of the effect on peak saccadic velocity, suggesting that subjects are not able to differentiate between 'sleepiness' and 'clumsiness' induced by benzodiazepines.

Changes of the visual fixation point are executed by saccadic eye movements (Collewijn, 1977), while fixation itself is carried out by the smooth-pursuit system, which is also impaired by benzodiazepines (Bittencourt & Richens, 1981). Because vision is not perfect during saccades, and slower saccades have a considerably longer duration, the total amount of visual input to the brain is likely to be decreased in subjects whose saccades are slowed down by benzodiazepines. These findings may underlie the association of increased risk of serious road accidents and use of benzodiazepines (Bø *et al.*, 1975; Skegg *et al.*, 1979) and the impairment of real car driving in outpatients on diazepam (de Gier, 1980).

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