Acute and subchronic effects of Org 2305 and diazepam on psychomotor performance in man

M. J. MATTILA, J. KOSKI & C. STRÖMBERG

Department of Pharmacology and Toxicology, University of Helsinki, Helsinki 17, Finland

1 Three doses (15, 30 and 60 mg) of Org 2305 (O 15, O 30 and O 60 respectively), a novel anxiolytic drug chemically related to mianserin, were compared with placebo and 15 mg diazepam (DZ) on human psychomotor performance in a double-blind, cross-over study with 15 healthy volunteers.

2 Objective measurements (choice reaction, tracking, flicker fusion, Maddox wing, digit symbol substitution, memory recall) and subjective assessments (visual analogue scales) were done at baseline and 2 and 13 h after the first dose. This testing procedure was repeated on day 7 when administering the seventh consecutive daily night-time dose.

3 After the first dose O 15 did not differ from placebo and O 30 rarely differed from placebo. O 60 impaired various objective functions similarly to, or less than DZ. Subjectively, DZ and O 60 were felt as sedative.

4 During subchronic treatment, DZ caused some impairment of baseline due to accumulation of bioassayable benzodiazepines, but significant responses to the last DZ dose were less than those to the first dose. DZ but not O 60 was reported to have caused lethargy and clumsiness during subchronic treatment.

5 In the doses used Org 2305 impaired psychomotor performance less than diazepam did. A dose of 60 mg Org 2305 may offer some advantage over 15 mg diazepam, provided that their anxiolytic effects are about similar.

Keywords human trial psychomotor impairment Org 2305 diazepam subchronic study

Introduction

Org 2305 is chemically 1, 2, 3, 4, 4a, 9-hexahydro-2-methyldibenzo [c, f] pyrimido [1, 6-a] azepine (E)-2-butenedioate (1:1). The free base is a tetracyclic compound which is chemically (Figure 1) and functionally (antihistaminergic, antiserotonergic, anti- α -adrenergic) related to the antidepressant mianserin but it has a more pronounced anxiolytic profile of action in animals. After single doses (2.5 to 80 mg) and prolonged administration (60 mg daily for 2 weeks) to healthy male subjects, dose-related drowsiness,

Figure 1 Structural formulas of mianserin and Org 2305.

Correspondence: Professor M. J. Mattila, Department of Pharmacology and Toxicology, University of Helsinki, Siltavuorenpenger 10, SF-00170 Helsinki, Finland

sleepiness and decreased internal tension have been recorded without significant effects on heart rate, blood pressure, ECG or chemical laboratory tests. Transient drowsiness was observed in the subchronic study in all subjects, accompanied in three subjects by mild EEG changes indicative of sedation. The EEG profiles recorded after 40 mg of Org 2305 were qualitatively similar to those previously seen with mianserin, but Org 2305 proved to be less potent (Grünberger & Saletu, 1980; Irwin & Frank, 1983; Fink *et al.*, 1977). After oral administration Org 2305 is well absorbed, the plasma peaks are achieved by 1–2 h, and its mean elimination half-life is about 26 h (N.V. Organon, on file).

Org 2305 is currently being investigated in controlled clinical trials for its anxiolytic effect, the effective daily dose being less than 100 mg. The present trial was conducted in order to compare Org 2305 and diazepam in terms of their effects on performance, as evaluated objectively and subjectively after a single dose and after 1 week treatment with a daily night-time dose. Some of the results have been briefly communicated (Mattila *et al.*, 1986a).

Methods

Subjects

Fifteen healthy students (nine females and six males), aged 20–27 years and weighing 50–88 kg, volunteered for the trial and were paid for their time. None of them suffered from mental or somatic disease, or was on medicines, or used alcohol in excess. They gave their written informed consent, and the trial protocol was approved by the departmental committee of ethics. The volunteers were subjected to clinical laboratory tests and they were trained to the performance tests before entering the trial.

Experimental design

Each subject received for 1 week, in a double-blind and cross-over manner at 2 week intervals, daily night-time doses of 15, 30 and 60 mg of Org 2305 (O 15, O 30 and O 60), 15 mg of diazepam (DZ), or placebo in random (Latin square) order. This subchronic treatment was then followed by a 7-day wash-out period, and the procedure was repeated until each volunteer had received each treatment once. The drugs were given in tablet form, three tablets (active and/or placebo) being administered at each dosing occasion. Vital signs and extensive laboratory safety data were monitored regularly (after each

7-day treatment period). Psychometric measurements and visual analogue mood ratings were done on the first and last day of each treatment period. The baseline tests were administered in the evening between 19.00 and 20.30 h, the tablets were taken with water, and the tests were repeated 2 and 13 (next morning) h after tablet intake. Each round of the tests lasted for 20 min, the tests were done in the same order, and the 15 subjects began the rounds at 6 min intervals. The tests at 2 h post drug began 1 h 50 min after the drug intake.

Parameters studied

Several psychometric tests were used for objective measurement of performance (Aranko *et al.*, 1983). In the choice reaction test the subjects responded to visual and sound stimuli, the test lasted for 6 min, and reaction errors and cumulative reaction times were recorded. The tracking test was driven at fixed speed for 30 s, the tracking errors and error % (relative length of track driven off the road) being recorded. Critical flicker fusion was measured by using red flickering light watched at the distance of 100 cm. The pupillary size was kept constant with special spectacles, the fusion point was measured twice (upwards and downwards), and their mean was recorded. Maddox wing device was used to reveal changes in heterophoria (imbalance of extraocular muscles) expressed in diopters. This test is very sensitive to benzodiazepines. In the digit symbol substitution test (Stone, 1984) simple figures were substituted by numbers, and the number of correct substitutions in 3 min were recorded. In a simple memory task spans of 10 digits were presented to the subjects who recalled them forwards or backwards. In the digit symbol substitution and memory tests, matched new material was provided for each time of assessment.

As to subjective assessments, several variables related to mood and alertness were measured by using a graded modification of the visual analogue mood rating scales (Bond & Lader, 1974). The subjects had to locate themselves between two extremes, without seeing their previous rating. The pairs of extremes used were alert/drowsy, calm/excited, strong/feeble, muzzy/clear-headed, well coordinated/clumsy, lethargic/energetic, contented/discontented, troubled/tranquil, mentally slow/quick-witted, tense/relaxed, attentive/dreamy, incompetent/proficient, happy/sad, antagonistic/friendly, interested/bored, and withdrawn/sociable. Adverse drug effects were recorded on the Record of Symptoms Emerging (ROSE) form.

The extensive clinical laboratory tests for organ functions covered haematology (haemoglobin, erythrocytes, leucocytes with differential count, thrombocytes, sedimentation rate), hepatic function (alkaline phosphatase, SGOT, bilirubin, γ -GT), renal function (creatinine, routine checks for urine) as well as blood glucose, serum proteins, and serum K^+ , Na^+ , Ca^{++} and inorganic phosphate.

Pharmacokinetics

Venous blood samples were taken before (baseline) and 2 and 13 h (next morning) after tablet intake on the first and the last day of each 7 day treatment period. Plasma was prepared, divided in two parts, and stored at $-20^\circ C$ for several weeks until required for analysis. Diazepam was assayed, without solvent extraction, with a radio-receptor method (Aranko *et al.*, 1984) against diazepam standards (100, 300, 900 and 3000 ng ml^{-1}) made to respective baseline plasma. [3H]-Flunitrazepam (0.9 mM) was used as a radioligand, rat cerebral cortex homogenate (0.25 mg protein) as a receptor preparation, and the incubation on ice lasted for 60 min. Org 2305 was assayed chemically with gas liquid chromatography (N.V. Organon, on file).

Statistical handling

Mean $\pm$ s.e. mean values of the test results were computed separately for absolute values and for Δ -values (changes from respective baselines). With regard to absolute results, a two-way ANOVA for cross-over trial was applied separately for each of the six times of assessment (BL_1 , 2 h, 13 h, BL_2 , 2 h, 13 h), using in the statistical model 'treatment effect' and 'period effect' as sources of variance. Period effects sometimes cloud treatment effects for various reasons (Liljequist & Mattila, 1979; File & Lister, 1983). If, however, treatment effects and period effects are disentangled—this is done in the two-way ANOVA for cross-over trial—problems no longer exist. In this ANOVA test, variance in the dependent variable was considered to be the sum of general mean, treatment effect, period effect, subject effect and random order effect. Pairwise comparisons between the drugs/dosages were made, expressing the results as estimates of treatment effect contrasts, with $P_{2-sided}$ -values. Summarizing, the two-way ANOVA seemed the most sensitive and the most appropriate statistical test for discovering differences in treatment effects.

Apert from the two-way ANOVA, a paired *t*-test was applied for testing the BL_2 values vs the

BL_1 values, separately for each of the five treatments. Furthermore, the results from objective tests were analyzed with one-way ANOVAs for repeated measures and paired *t*-tests, comparing the Δ -values of each active drug with respective placebo Δ -values. Similar comparisons were done for subjective visual analogue results by using the Wilcoxon sign test. The results of these simple analyses reflect the drug responses as they occur after individual doses taken during the treatment. Since the period effect was not included, these mean responses do not represent pure treatment effects, and there were fewer significant differences. However, this kind of simple testing avoids an exaggeration of subtle drug effects which can reach statistical significance, without practical relevance. However, the results in terms of Δ -values are described only briefly.

Side-effects were tested by means of the sign-test, comparing the active drugs pairwise with placebo and taking into account the data of a subject concerning a certain side-effect only in case the answer was 'no' for placebo and 'yes' for the active drug, or vice versa.

Results

In general, the baseline values on day 1 (BL_1) of the various objective tests remained stable and comparable during different treatment periods. However, the period effects including learning component were assessed in the two-way ANOVA by calculating the estimate for the period effect contrast '1 minus 5' with a $P_{2-sided}$ -value. Some clearly significant results occurred, for instance for reaction time to mixed stimuli and for digit symbol substitution, in the sense that learning effects were present.

Acute effects

Objective measurements Considering obvious diurnal variation of alertness, response variation to placebo remained negligible in all tests. The two lowest doses of Org 2305 (O 15 and O 30) failed to differ significantly from placebo on any test variable except that O 30 increased exophoria (Maddox wing) 2 h after tablet intake. A simple comparison of drug effects shows that in general O 60 impaired various functions similarly to, or less than DZ did (Table 1).

The two-way ANOVA revealed at 2 h the following statistically significant differences between treatment groups. Tracking errors and tracking error % were increased in response to DZ. In tracking error %, DZ differed signifi-

Table 1 Mean $\pm$ s.e. mean values of some objective psychomotor and cognitive tests in 15 healthy subjects after treatment with placebo, 60 mg Org 2305 (O 60) and 15 mg diazepam (DZ). BL₁ and BL₂ refer to baseline tests before dosing on day 1 and 7 respectively; 2 h and 13 h refer to tests done 2 and 13 h after dosing. For more details see text.

Test/Drug	Acute treatment			Subchronic treatment		
	BL ₁	2 h	13 h	BL ₂	2 h	13 h
<i>Tracking error %</i>						
Placebo	19.4 $\pm$ 2.2	21.5 $\pm$ 2.2	18.5 $\pm$ 2.1	19.3 $\pm$ 2.7	19.3 $\pm$ 1.8	17.0 $\pm$ 2.1
O 60	20.9 $\pm$ 2.3	24.2 $\pm$ 2.6	19.6 $\pm$ 2.7	19.5 $\pm$ 2.4	20.4 $\pm$ 2.4	20.4 $\pm$ 2.6
DZ	20.7 $\pm$ 2.0	31.5 $\pm$ 2.4***	19.3 $\pm$ 1.5	20.3 $\pm$ 2.2	28.1 $\pm$ 2.6***	20.7 $\pm$ 2.4
<i>Reaction times to mixed stimuli (s)</i>						
Placebo	39.3 $\pm$ 2.0	39.6 $\pm$ 1.9	38.9 $\pm$ 2.0	38.3 $\pm$ 2.0	37.9 $\pm$ 1.8	39.0 $\pm$ 1.9
O 60	38.3 $\pm$ 1.6	39.2 $\pm$ 1.6	39.1 $\pm$ 1.7	37.5 $\pm$ 1.3	38.3 $\pm$ 1.3	37.0 $\pm$ 1.3**
DZ	39.5 $\pm$ 2.0	41.9 $\pm$ 2.3*	40.1 $\pm$ 2.1	39.6 $\pm$ 1.9	40.6 $\pm$ 2.0**	39.5 $\pm$ 1.8
<i>Critical flicker fusion frequency (Hz)</i>						
Placebo	22.4 $\pm$ 0.6	21.5 $\pm$ 0.6	21.6 $\pm$ 0.6	22.4 $\pm$ 0.6	21.5 $\pm$ 0.6	22.9 $\pm$ 0.7
O 60	22.8 $\pm$ 0.7	20.3 $\pm$ 0.7**	22.6 $\pm$ 0.8	22.3 $\pm$ 0.5	20.5 $\pm$ 0.6*	22.7 $\pm$ 0.8
DZ	22.5 $\pm$ 0.7	20.4 $\pm$ 0.5*	22.2 $\pm$ 0.6	22.4 $\pm$ 0.4	21.0 $\pm$ 0.5	22.6 $\pm$ 0.5
<i>Digits substituted</i>						
Placebo	156 $\pm$ 6	156 $\pm$ 8	148 $\pm$ 5	155 $\pm$ 6	148 $\pm$ 5	151 $\pm$ 4
O 60	155 $\pm$ 7	142 $\pm$ 4*	145 $\pm$ 6	148 $\pm$ 5*	145 $\pm$ 5	147 $\pm$ 5
DZ	155 $\pm$ 7	140 $\pm$ 5**	147 $\pm$ 4	150 $\pm$ 8 ^a	138 $\pm$ 4**	143 $\pm$ 4**
<i>Maddox wing</i>						
Placebo	3.3 $\pm$ 1.3	3.1 $\pm$ 1.3	3.7 $\pm$ 1.3	2.9 $\pm$ 1.1	3.2 $\pm$ 1.0	3.3 $\pm$ 1.0
O 60	3.4 $\pm$ 1.1	5.1 $\pm$ 1.1**	4.2 $\pm$ 1.2	3.5 $\pm$ 1.1	4.6 $\pm$ 1.1***	3.7 $\pm$ 1.0
DZ	3.0 $\pm$ 1.0	5.0 $\pm$ 1.4**	3.9 $\pm$ 1.0	3.9 $\pm$ 1.1 ^{a*}	6.1 $\pm$ 1.1***	5.1 $\pm$ 1.1***

Paired *t*-test a = vs BL₁ ($P < 0.05$); two-way ANOVA vs placebo: * $P < 0.05$; ** $P < 0.01$ and *** $P < 0.001$

cantly not only from placebo but also from all doses of Org 2305. DZ increased reaction times to mixed stimuli and differed significantly from placebo and O 60 (Figure 2). In the digit symbol substitution test, DZ and O 60 differed significantly from placebo in reducing the number of digits substituted (Table 1). In the Maddox wing test, DZ, O 60 and also O 30 caused exophoria, being significantly different from placebo and 0.15. In the critical flicker fusion test, DZ and O 60 differed significantly from placebo in lowering the fusion frequency. DZ tended to lower systolic blood pressure and differed significantly from O 60 which tended to increase blood pressure.

The two-way ANOVA revealed only a few significant differences between treatment groups at 13 h. O 60 caused tracking errors significantly more than placebo did and DZ impaired backward recall in the memory test differing significantly from placebo, O 15 and O 30.

When the Δ -values (changes from BL) were analyzed with one-way ANOVA and paired *t*-test, excluding period effect, significant differences were fewer. However, responses to DZ at 2 h differed from placebo responses on tracking error %, flicker fusion, digit symbol substitution

and Maddox wing. O 60 differed from placebo on flicker fusion and Maddox wing.

Subjective assessments When analyzing subjective sedation at 2 h on visual analogue scales (VAS), DZ and O 60 altered the assessments on various VAS variables related to sedation (Table 2). In addition DZ and O 60 rendered the subjects muzzy, mentally slow, dreamy, and incompetent. O 60 increased calmness more clearly than O 15, O 30 and even DZ did, although all four differed significantly from placebo. O 15, O 30, O 60 and DZ increased drowsiness differing significantly from placebo. DZ tended to increase happiness while Org 2305 did not. O 30 tended to render the subjects friendly and differed significantly from O 15 and O 60 in this respect.

The two-way ANOVA revealed only a few significant differences between treatment groups at 13 h. DZ caused some happiness in comparison with placebo whereas O 30 and O 60 showed a tendency towards sedation on several VAS variables.

Simple comparison (Wilcoxon test) of post-drug Δ -values vs respective placebo responses at 2 h gave about similar results as presented

Figure 2 Mean choice reaction times of 15 subjects before and after the first and last doses of diazepam 15 mg (DZ) and 15 mg, 30 mg and 60 mg of Org 2305 (O). Statistical differences (two-way ANOVA): *vs placebo; • vs O 60 mg. 1 Placebo, 2 O 15, 3 O 30, 4 O 60 and 5 diazepam.

above. However, O 15 did not cause drowsiness, and O 60 rendered the subjects discontented, lethargic and mentally slow but not clumsy. O 30 and O 60 caused muzziness more than DZ did while DZ caused more relaxation and clumsiness. No significant changes were found on happy/sad and friendly/antagonistic scales. No clear drug effects were found at 13 h.

Subchronic effects

When on placebo, no major learning effects were encountered although some improvement of complex performances (tracking, choice reaction) was seen at the end of the one-week maintenance. When on active drugs, a statistically significant difference between BL₁ and BL₂ values was uncommon, but it was found with DZ on some objective skills (Table 1), probably due to a cumulative drug effect. As to subjective assessments of VAS variables statistically significant differences between BL₁ and BL₂ values were occasionally seen (Table 2), probably due to fluctuations of the initial acute baselines (BL₁).

The two-way ANOVA failed to reveal statistically significant differences between the BL₂ values of the different treatment groups on tracking skills, flicker fusion, and memory. Reaction times to mixed stimuli at BL₂ were significantly longer after DZ than after O 60 whereas O 60 impaired digit symbol substitution in comparison with placebo (Table 1). In Maddox wing test, not only DZ but also O 30 increased exophoria in comparison with placebo. DZ lowered systolic blood pressure in comparison with placebo and O 60. When comparing the BL₂ values of various VAS variables, both DZ and O 60 shifted the assessment towards calmness and relaxation when compared to placebo (Table 2).

Objective measurements The two-way ANOVA revealed the following significant differences between treatment groups at 2 h (Table 1). DZ impaired coordinative skills by increasing tracking errors and tracking error % in comparison with placebo, O 30 and O 60. DZ prolonged reaction times to mixed stimuli, and impaired digit symbol substitution. In the flicker fusion

Table 2 Mean $\pm$ s.e. mean values of some visual analogue scale assessments (mm) by 15 healthy subjects after treatment with placebo, 60 mg Org 2305 (O 60) and 15 mg diazepam (DZ). The first mentioned extreme of each pair refers to zero and the latter to 100. For more details see text and Table 1.

Test/Drug	Acute treatment			Subchronic treatment		
	BL ₁	2 h	13 h	BL ₂	2 h	13 h
<i>Alert/Drowsy</i>						
Placebo	46 $\pm$ 5	56 $\pm$ 5	54 $\pm$ 4	48 $\pm$ 5	55 $\pm$ 5	55 $\pm$ 4
O 60	48 $\pm$ 5	77 $\pm$ 3***	59 $\pm$ 4	56 $\pm$ 4	75 $\pm$ 3***	58 $\pm$ 6
DZ	48 $\pm$ 6	72 $\pm$ 5**	52 $\pm$ 5	56 $\pm$ 6	72 $\pm$ 4***	65 $\pm$ 4*
<i>Calm/Excited</i>						
Placebo	40 $\pm$ 4	47 $\pm$ 4	44 $\pm$ 2	51 $\pm$ 3 ^a	45 $\pm$ 3	42 $\pm$ 4
O 60	45 $\pm$ 5	31 $\pm$ 4***	35 $\pm$ 4**	37 $\pm$ 5*	31 $\pm$ 4***	39 $\pm$ 4
DZ	48 $\pm$ 4	36 $\pm$ 4**	39 $\pm$ 4	45 $\pm$ 5	36 $\pm$ 5*	45 $\pm$ 5
<i>Tense/Relaxed</i>						
Placebo	58 $\pm$ 5	54 $\pm$ 4	55 $\pm$ 3	48 $\pm$ 3	55 $\pm$ 4	57 $\pm$ 4
O 60	55 $\pm$ 4	59 $\pm$ 4	63 $\pm$ 3	60 $\pm$ 4**	61 $\pm$ 3	57 $\pm$ 3
DZ	51 $\pm$ 5	67 $\pm$ 3***	55 $\pm$ 4	61 $\pm$ 4**	65 $\pm$ 4*	59 $\pm$ 4
<i>Muzzy/Clearheaded</i>						
Placebo	54 $\pm$ 5	53 $\pm$ 4	52 $\pm$ 4	54 $\pm$ 4	53 $\pm$ 3	54 $\pm$ 4
O 60	55 $\pm$ 4	40 $\pm$ 5*	52 $\pm$ 4	55 $\pm$ 4	41 $\pm$ 4*	49 $\pm$ 4
DZ	52 $\pm$ 4	39 $\pm$ 5**	53 $\pm$ 5	46 $\pm$ 4	40 $\pm$ 5**	46 $\pm$ 5
<i>Attentive/Dreamy</i>						
Placebo	45 $\pm$ 4	49 $\pm$ 4	52 $\pm$ 4	51 $\pm$ 3	55 $\pm$ 3	57 $\pm$ 4
O 60	49 $\pm$ 4	64 $\pm$ 5**	51 $\pm$ 4	51 $\pm$ 3	63 $\pm$ 3*	54 $\pm$ 5
DZ	47 $\pm$ 5	65 $\pm$ 4**	48 $\pm$ 5	55 $\pm$ 4	64 $\pm$ 4**	59 $\pm$ 4

Paired *t*-test: a = vs BL₁ ($P < 0.05$); two-way ANOVA vs placebo: * $P < 0.05$; ** $P < 0.01$ and *** $P < 0.001$

test, the critical fusion frequency with O 60 was significantly lower than that with placebo. In the Maddox wing test, DZ, O 30 and O 60 differed significantly from placebo while DZ still differed significantly from all individual doses of Org 2305.

When the performances at 13 h after the last dose were handled with the two-way ANOVA, O 60 shortened reaction times to mixed stimuli and DZ increased exophoria (Maddox wing) in comparison with all other treatments. DZ also reduced the number of digits substituted. In the reaction times to mixed stimuli and Maddox wing test O 30 proved occasionally stronger than O 60. DZ lowered systolic blood pressure in comparison with placebo, O 30 and O 60.

Simple comparison (paired *t*-test) of post-drug Δ -values vs respective placebo responses revealed that DZ increased tracking error % at 2 h. No significant differences were found at 13 h.

Subjective assessments The two-way ANOVA revealed the following statistically significant differences between treatments at 2 h. All active drugs differed from placebo in causing drowsi-

ness, and DZ as well as O 60 and O 30 increased calmness and rendered the subjects feeble, muzzy and dreamy. DZ also caused clumsiness, relaxation and lethargy, while all doses of ORG 2305 differed from placebo in rendering the subjects bored. At 13 h after the last dose, DZ caused drowsiness while O 60 shifted the assessments towards well coordinated and O 30 towards antagonistic.

Simple comparison of post-drug Δ -values vs placebo responses (Wilcoxon test) at 2 h revealed that DZ rendered the subjects drowsy and feeble, O 15 muzzy, and O 30 dreamy and bored. O 60 rendered the subjects drowsy at 2 h and tense at 13 h.

Side-effects

In the *acute* phase statistically significant effects appeared only when on DZ: the items clumsiness and slowed thinking differed significantly from respective records when on placebo (Table 3). In the *subchronic* phase DZ caused tiredness significantly more than placebo did. The subjects slept better on O 60 and O 30 than on

Table 3 Some subjective side-effects recorded on questionnaire after the first dose (= acute) and after 1 week treatment (= subchronic). O 15, O 30, O 60 and DZ refer to treatment with 15 mg, 30 mg, 60 mg Org 2305 and 15 mg diazepam respectively. Statistical significances (* $P \leq 0.05$) refer to respective values when on placebo. For more details see text.

Side-effects	Treatment				
	Placebo	O 15	O 30	O 60	DZ
<i>Acute side-effects</i>					
Apathetic	0	1	2	2	4
Clumsiness	0	1	3	3	8*
Poor concentration	1	0	4	3	1
Tired	2	2	6	5	7
Loss of energy	1	1	6	3	3
Sedated	1	1	3	4	3
Sleepy, drowsy	2	2	7	5	7
Slowed thinking	1	1	4	5	8*
<i>Subchronic side effects</i>					
Poor concentration	1	2	3	3	4
Tired	1	3	2	4	8*
Loss of energy	1	1	1	3	4
Sedated	0	2	2	2	4
Sleepy, drowsy	2	1	1	1	5
Difficulty waking	0	1	5	3	4
Dreams, nightmares	1	3	1	1	4
Dull in the morning	0	0	3	2	5
Sleeping better	1	6	8*	9*	7

placebo, but DZ did not share this effect at a significant level although the differences with O 30 and O 60 were small.

No clinically significant deviations from reference limits were encountered in laboratory tests on haematology, blood chemistry, hepatic function and renal function.

Pharmacokinetics

As can be seen in Table 4, the plasma levels of Org 2305 increased in relation to the doses given

in acute experiments. The steady state concentrations at BL₂ were negligible with O 15 and O 30, and after O 60 they were definitely lower than the levels produced by acute single 15 mg dose of Org 2305. After the administration of the last dose of Org 2305 the plasma levels were again related to the doses given, yet slightly higher than the respective levels after acute single doses. When the plasma levels of DZ were bioassayed against diazepam standards made to the respective BL₁ plasmas, acute post-drug concentrations represent mainly parent drug

Table 4 Mean $\pm$ s.e. mean plasma concentrations of Org 2305 (O) and diazepam (DZ) in ng ml⁻¹ after the first and last dose during the daily night-time treatment of 15 healthy subjects for one week with 15, 30 and 60 mg of Org 2305 and 15 mg of diazepam (O 15, O 30, O 60 and DZ respectively). BL₂ refers to the baseline value on day 7 before dosing. Bioassayed DZ concentrations include its active metabolite nordiazepam; the ranges of plasma benzodiazepine concentrations are given in parentheses.

Time after dosing	Treatment			
	O 15	O 30	O 60	DZ (range)
2 h	5.1 $\pm$ 2.7	8.6 $\pm$ 8.3	22.6 $\pm$ 17.8	395 $\pm$ 44 (65–790)
13 h	0.9 $\pm$ 0.4	1.5 $\pm$ 0.4	3.5 $\pm$ 1.8	133 $\pm$ 29 (0–370)
BL ₂	0.6 $\pm$ 0.3	1.0 $\pm$ 0.3	2.2 $\pm$ 1.3	786 $\pm$ 59 (520–1250)
2 h	6.1 $\pm$ 3.3	10.8 $\pm$ 7.7	22.7 $\pm$ 15.0	1178 $\pm$ 81 (620–1550)
13 h	1.1 $\pm$ 0.6	1.7 $\pm$ 0.5	5.3 $\pm$ 2.5	926 $\pm$ 75 (350–1550)

diazepam whereas subchronic DZ concentration (from BL₂ on) represent both diazepam and its active metabolite nordiazepam. The concentrations achieved are about similar to, or lower than, those we have previously assayed after comparable daily doses of diazepam.

Discussion

Various test performances after placebo and diazepam proved to be similar to those we have previously recorded in several acute and subchronic trials (Aranko *et al.*, 1983, 1984, 1985). The bioassayed plasma concentrations of diazepam in the present trial were similar to those measured after diazepam 5 mg three times daily for 1 week (Aranko *et al.*, 1985) but slightly higher than those after diazepam 5 mg twice daily for 1 week (Aranko *et al.*, 1984). Against this background of reproducible methods and positive diazepam control, it seems justified to conclude that 15 mg Org 2305 (O 15) proved inert on performance whereas 60 mg Org 2305 (O 60) caused objective and subjective impairment similar to, or less than diazepam did. 30 mg Org 2305 (O 30) proved to be active on some variables but remained mostly inactive.

The psychomotor and sedative side-effects of Org 2305 and benzodiazepines cannot be really compared until the anxiolytic doses of Org 2305 have been established. The present results with O 60 resemble those we have recorded with acute (10 mg) and subchronic (30 mg daily) doses of mianserin (Seppälä, 1977; Mattila *et al.*, 1978); in these studies, a tolerance towards the psychomotor side-effects of mianserin developed in 2 weeks. Our recent experiments with an acute single dose of mianserin 20 mg (Strömberg & Mattila, unpublished, 1985) have shown that it is about equipotent to O 60, a result tallying with previous EEG data (Irwin & Fink, 1983). Considering these observations and pharmacokinetics together, it seems as if Org 2305 would have some advantage over accumulating benzodiazepines diazepam included.

The anxiolytic profile of Org 2305, both in animals and in man, may sound surprising because it is closely related to antidepressant mianserin (Figure 1). However, the antidepressant

mechanisms are poorly understood (e.g. Hollister, 1986), anxiety and depression may often co-exist, and tricyclic antidepressants and sedative drugs such as buspirone and histamine H₁-antihistamines have been used to treat neurotic symptoms. Enantiomers and metabolites of mianserin are less sedative than the racemic parent compound although they share its antidepressant profile of action (Pinder & Van Delft, 1983). On these grounds the different profiles of action of mianserin and Org 2305 are not extraordinary.

The mechanisms of O 60 induced impairment of performance can only be speculated. The nonspecific sedation after various psychotropic drugs has been attributed e.g. to their central H₁-antagonistic and anti- α -adrenergic properties (Gordon, 1974), and traditional H₁-receptor antagonists do impair performance objectively and subjectively (Clarke & Nicholson, 1978; Nicholson & Stone, 1986). In our hands, diphenhydramine 100 mg has proved to be subjectively sedative and distressing, it impairs objective performance less than 0.3 mg kg⁻¹ of diazepam does, and it alters the balance of extraocular muscles (Maddox wing) in the opposite direction as compared to Org 2305 and diazepam (Mattila *et al.*, 1986b). The sedation caused by H₁-antihistamines can be non-specific (Nicholson & Stone, 1986). The antiserotonin effect of Org 2305 could contribute to the O 60 induced impairment of performance, because an antiserotonergic component is included even in benzodiazepine action (Stein, 1980). The α -adrenergic blocking property of Org 2305 is relatively weak and hardly explains its psychomotor effects. At the moment we are inclined to suggest an anti-5-HT effect in combination with a non-specific membrane effect being responsible for the impairment of performance by various psychotropic drugs. Buspirone is a novel anxiolytic drug which acts in a dose dependent way on dopaminergic mechanisms and also may activate 5-HT and block adrenergic receptors (McMillen & McDonald, 1983; Hjorth & Carlsson, 1983). It is subjectively sedative but does not impair performance objectively (Seppälä *et al.*, 1982).

The authors owe Dr Jan Voerman their thanks for his great help in the statistical handling of the data.

References

- Aranko, K., Mattila, M. J. & Seppälä, T. (1983). Development of tolerance and cross-tolerance to psychomotor actions of lorazepam and diazepam in man. *Br. J. clin. Pharmacol.*, **15**, 545-552.
- Aranko, K., Mattila, M. J., Seppälä, T. & Aranko, S. (1984). The contribution of active metabolites to the tolerance developing to diazepam in man: relationship to bioassayed serum benzodiazepine levels. *Med. Biol.*, **62**, 277-284.
- Aranko, K., Mattila, M. J. & Bordignon, D. (1985).

- Psychomotor effects of alprazolam and diazepam during acute and subacute treatment, and during the followup phase. *Acta Pharmac. Tox.*, **56**, 364–372.
- Bond, A. & Lader, M. (1974). The use of analogue scales in rating subjective feelings. *Br. J. med. Psychol.*, **47**, 211–218.
- Clarke, C. H. & Nicholson, A. N. (1978). Performance studies with antihistamines. *Br. J. clin. Pharmac.*, **6**, 31–36.
- File, S. E. & Lister, R. G. (1983). Does tolerance to lorazepam develop with once weekly dosing? *Br. J. clin. Pharmac.*, **16**, 645–650.
- Fink, M., Irwin, P., Gasper, M. & DeRidder, J. J. (1977). EEG, blood level, and behavioral effects of the antidepressant mianserin (Org GB-94). *Psychopharmacology*, **54**, 249–254.
- Gordon, M. (1974). *Psychopharmacological agents, vols II and III*. New York: Academic Press Inc.
- Grünberger, J. & Saletu, B. (1980). Determination of pharmacodynamics of psychotropic drugs by psychometric analysis. *Progr. Neuropsychopharmac.*, **5**, 417–434.
- Hjorth, S. & Carlson, A. (1983). Buspirone effects on central monoaminergic transmission—possible relevance to animal experimental and clinical findings. *Eur. J. Pharmac.*, **83**, 299–303.
- Hollister, L. (1986). Current antidepressants. *Ann. Rev. Pharmac. Tox.*, **26**, 23–27.
- Irwin, P. & Fink, M. (1983). Familiarization session and placebo control in EEG studies of drug effects. *Neuropsychobiology*, **10**, 173–177.
- Liljequist, R. & Mattila, M. J. (1979). Acute effects of temazepam and nitrazepam on psychomotor skills and memory. *Acta Pharmac. Tox.*, **44**, 364–369.
- Mattila, M. J., Liljequist, R. & Seppälä, T. (1978). Effects of amitriptyline and mianserin on psychomotor skills and memory in man. *Br. J. clin. Pharmac.*, **5**, Suppl. 1, 53S–55S.
- Mattila, M. J., Koski, J. & Strömberg, C. (1986a). Acute and subacute effects of Org 2305 and diazepam on psychomotor performance in man. *Br. J. clin. Pharmac.*, **21**, 89P.
- Mattila, M. J., Mattila, M. & Konno, K. (1986b). Acute and subacute actions on human performance and interactions with diazepam of temelastine (SK & F 93944) and diphenhydramine. *Eur. J. clin. Pharmac.*, **31** (in press).
- McMillen, B. A. & McDonald, C. C. (1983). Selective effects of buspirone and malindone on dopamine metabolism and function in the striatum and frontal cortex of the rat. *Neuropharmacology*, **22**, 273–278.
- Nicholson, A. N. & Stone, B. M. (1986). Antihistamines: impaired performance and the tendency to sleep. *Eur. J. clin. Pharmac.*, **30**, 27–32.
- Pinder, R. M. & Van Delft, A. M. L. (1983). The potential therapeutic role of the enantiomers and metabolites of mianserin. *Br. J. clin. Pharmac.*, **15**, 269S–276S.
- Seppälä, T. (1977). Psychomotor skills during acute and two-week treatment with mianserin (Org GB-94) and amitriptyline, and their combined effects with alcohol. *Ann. clin. Res.*, **9**, 66–72.
- Seppälä, T., Aranko, K., Mattila, M. J. & Shrotriya, R. C. (1982). Psychopharmacological effects of buspirone or lorazepam, and alcohol. *Clin. Pharmac. Ther.*, **32**, 201–207.
- Stein, L. (1980). Behavioural neurochemistry of benzodiazepines. *Arzneim. Forsch. (Drug Res.)*, **30**, 868–873.
- Stone, B. I. (1984). Pencil and paper tests—sensitivity to psychotropic drugs. *Br. J. clin. Pharmac.*, **18**, 15S–20S.

(Received 14 July 1986,
accepted 13 October 1986)