

The placebo effect in healthy volunteers: influence of experimental conditions on physiological parameters during phase I studies

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- 1 Although placebo administration is now commonly used as a control condition during clinical pharmacology studies conducted in healthy volunteers, data in placebo-treated subjects usually receive little attention.
- 2 The profile of several physiological and hormonal parameters was reviewed during the placebo sessions of five double-blind phase I studies involving hospitalisation of healthy volunteers for 2 weeks or more.
- 3 A clear trend towards an increase in heart rate, which culminated at about 10 beats min^{-1} at the end of the placebo period was observed, whereas both systolic and diastolic blood pressures remained globally unchanged.
- 4 Increases in the time to sleep initiation or in asthenia self-ratings during the placebo sessions suggested poor neuropsychiatric tolerability of experimental conditions in some of the studies.
- 5 In conclusion, all these data confirm that subjects change during the course of these studies. This is an important reason for conducting phase I studies under double-blind placebo-controlled conditions and to refrain from within-group comparisons (*vs* baseline, first-day effect).

Keywords placebo healthy volunteers phase I studies

Introduction

Placebo, a Latin word from the Roman Catholic liturgy, can be literally translated to 'I shall please'. The first article on the placebo effect was published in 1945 [1]. Although use of a placebo in clinical pharmacology trials is now the gold standard approach, very little attention is paid to the data gathered in placebo-treated healthy volunteers. We recently reviewed 109 double-blind, placebo-controlled, Synthélabo-sponsored studies conducted in 1228 volunteers over the last 10 years [2]. Overall, 19% of the subjects on placebo complained of at least one adverse event (most frequently headache, drowsiness, and asthenia), with some variation depending on study design and population. In order to shed new light on the impact of experimental conditions on the results of clinical pharmacology studies conducted in healthy volunteers, we reviewed the data collected in placebo-treated subjects during five different repeated-dose phase I studies.

Methods

Subjects

All the volunteers who participated in these studies were found to be healthy after careful physical examination and standard haematological and biochemical screening tests. In accordance with the principles of good clinical practice, the studies were approved by local ethics committees, and all the volunteers gave their written informed consent before inclusion in the studies. Volunteers were aged 18 to 40 years and their weight and height were within normal limits. Most of the young volunteers were medical students and were recruited by word of mouth. Financial compensation, which varied significantly across studies, was based mainly on duration of the hospitalisation. In all the studies, the volunteers were informed before entering the study that they might receive a placebo.

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Methods

The five studies were all non-therapeutic trials conducted between 1983 and 1993 in Europe under double-blind placebo-controlled conditions in healthy volunteers given repeated doses of a placebo or active drug (for 13 to 19 days using a parallel-group design). All the studies were aimed at assessing the clinical and laboratory safety of new chemical compounds. The studies were conducted either in the clinical pharmacology department of a university teaching hospital or in facilities belonging to contract research organizations in Europe. All five studies used a parallel-group design and involved hospitalisation of the volunteers throughout the study period.

Evaluation criteria

Study A Haemodynamic parameters were assessed using an automated apparatus (Dinamap®). During the same study, the Leeds Sleep Evaluation Questionnaire (LSEQ) [3, 4] was used to rate subjective perceptions of ease of sleep initiation, quality of sleep, and coordination of behaviour following waking, on 10 cm line analogue rating scales. Subjects completed the LSEQ forms every morning, 30 min after awakening on days 0, 1, 5, 10 and 14 of hospitalisation, from the day after the first dose to the day after the last dose. Finally, on day 1 and day 12 or 13 of treatment, subjects underwent a glucose infusion test (GIT) in which blood glucose was rapidly brought up to, then maintained at about 12 mmol l⁻¹ by administering an i.v. bolus of glucose (0.25 g kg⁻¹) followed by a 1 h i.v. infusion of glucose (10 mg kg⁻¹ min⁻¹).

Data were analysed according to a split plot Anova model for repeated measurements.

Study B Haemodynamic parameters were assessed using the same automated apparatus (Dinamap®). During study B, the sleep latency was self-evaluated on the morning after dosing and the critical flicker fusion frequency threshold (CFF) was assessed as an index of overall CNS activity or cortical arousal [5]. Subjects were required to discriminate flicker from fusion of four red light-emitting diodes held in foveal fixation at a viewing distance of 1 m, using the Leeds psychomotor tester. Individual thresholds were determined by the psychophysical method of limits as the mean of three ascending and three descending

measurements. Furthermore, during this study, sensory-motor performance was assessed on the basis of the choice reaction time (CRT), which measures both motor and recognition reaction times (MRT and RRT, respectively), as well as total reaction time (TRT). Subjects were required to turn off one of six red-lights, lit at random, by touching the appropriate response button using the Leeds psychomotor tester. The recorded reaction time was the mean latency of 50 stimuli presentations [6].

Data were analysed using a split plot Anova model for repeated measurements.

Studies C and D Haemodynamic parameters were assessed using the same automated apparatus (Dinamap®).

Study E This tyramine interaction study of befloxtone, a selective, reversible mono-amine oxidase-A (MAO-A) inhibitor, was based on determination of the TYR 30 (amount of tyramine which induces a greater than 30 mm Hg increase in systolic blood pressure), as follows. The TYR 30 was assessed on three occasions: under fasting conditions (selection criterion: TYR 30 between 400 and 600 mg), after a placebo run-in period, and after repeated doses of befloxtone. During the third period, treatments were administered and tyramine tests were performed (once a day) with increasing doses of tyramine from 50 mg to 300 mg by 50 mg increments, until the TYR 30 was reached. Befloxtone was then discontinued, but a tyramine test was done each day using the same tyramine dose until this dose no longer met the TYR 30 criterion. Time to recovery was defined as the interval between the day of befloxtone discontinuation and the day of failure to meet the TYR 30 criterion.

Results

Haemodynamic parameters on placebo

Four studies (A, B, C, D; see Table 1) in a total of 26 healthy young volunteers were reviewed. In three of the four studies, a clinically significant increase in supine heart rate (HR) of about 10 beats min⁻¹ was recorded after 13 to 16 days of placebo treatment, whereas both systolic and diastolic blood pressures (BP) remained globally unaffected (Figure 1). Values

Table 1 Haemodynamic parameters on placebo: study characteristics

Study code	Subjects (n)	Duration of placebo exposure (days)	Comparative active drug
A	6	14	α ₂ -adrenoceptor antagonist
B	9	19*	Imidazopyridine anxiolytic
C	5	13	Calcium antagonist
D	6	14	5-HT reuptake inhibitor

*16 days of in-hospital treatment followed by 3 days of ambulatory treatment.

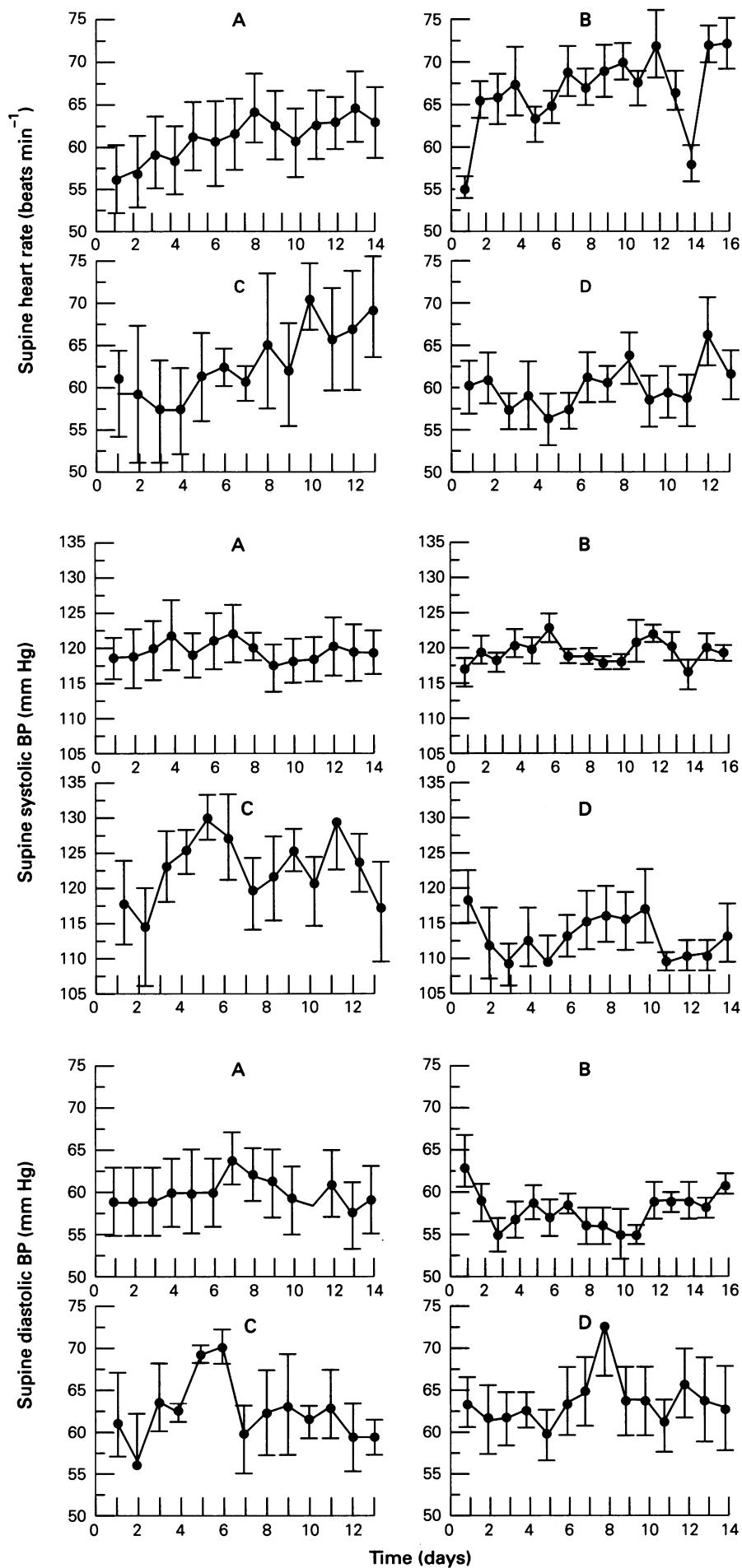


Figure 1 Mean (\pm s.e. mean) daily haemodynamic parameters after repeated doses of placebo in hospitalised healthy subjects (study characteristics are displayed in Table 1).

recorded in the standing position (not shown) showed similar results, i.e. an increase in HR with no change in BP. These observations seemed to be quite consistent from one subject to another as shown by the individual profiles of the supine haemodynamic parameters during study A (Figure 2).

Sleep latency, sleep scale, and psychometrics on placebo

Nine subjects were given a placebo during a 19 day repeated-dose study (study B). During the 16 day hospital stay, there was a progressive increase in mean sleep latency, from less than 20 min on night 1 to more than 40 min on night 16 (Figure 3). On day 17, the subjects were discharged from the hospital but continued to take the placebo. However, their sleep latencies returned to baseline values, demonstrating that prolonged hospitalisation adversely affected self-rated sleep latency. During the same study, CFF and CRT were determined on days 1 and 14 in the nine placebo-treated volunteers.

All the subjects had received intensive training before treatment in order to minimize learning effects during the study. However, a significant increase in the CFF threshold ($P = 0.02$) and a significant decrease in the CRT ($P = 0.001$) were recorded on day 14 vs day 1 (Figure 4). These changes in both psychometric tests indicated enhanced arousal and psychomotor performance after 14 days of hospitalisation in comparison to the first day.

In study A, performed in six subjects hospitalised for 14 days, the LSEQ evidenced an increase in tiredness at awakening and during the morning, which increased with time spent in the hospital, whereas the other parameters (wakefulness, awakenings, ...) remained unaffected (Figure 5).

Placebo conditioning

The tyramine interaction study performed in 10 healthy hospitalised males (study E) confirmed that, in most subjects, the TYR 30 is considerably higher in fed subjects on placebo than in fasted subjects (Table 2). Time to recovery was markedly influenced by experimental conditions. Time to recovery was determined under placebo treatment in the first four subjects and was found to be 3 to 4 days, which was unexpectedly long for such a rapidly reversible MAO-A inhibitor. It was therefore decided to give no drugs at all instead of a placebo during the recovery period in the last six subjects. In all these subjects, time to recovery was 1 day, showing that 'classical conditioning' can occur in healthy volunteers.

Change in metabolism

During Study A in six healthy volunteers given a placebo for 14 days (study A), a GIT was performed on days 1 and 12 (or 13) of treatment. On day 12/13, both insulin and C-peptide levels were significantly ($P < 0.05$) higher than on day 1, while blood glucose remained unchanged (Figure 6). These results provide

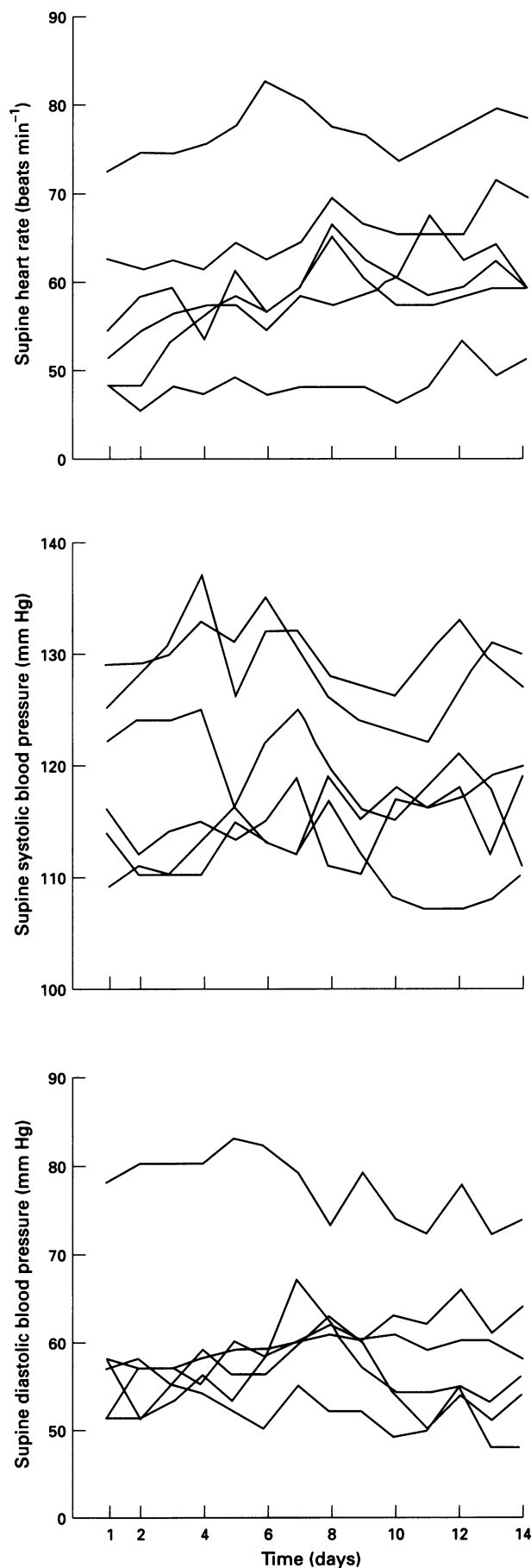


Figure 2 Individual plots of supine haemodynamic parameters in six hospitalised healthy subjects (study A).

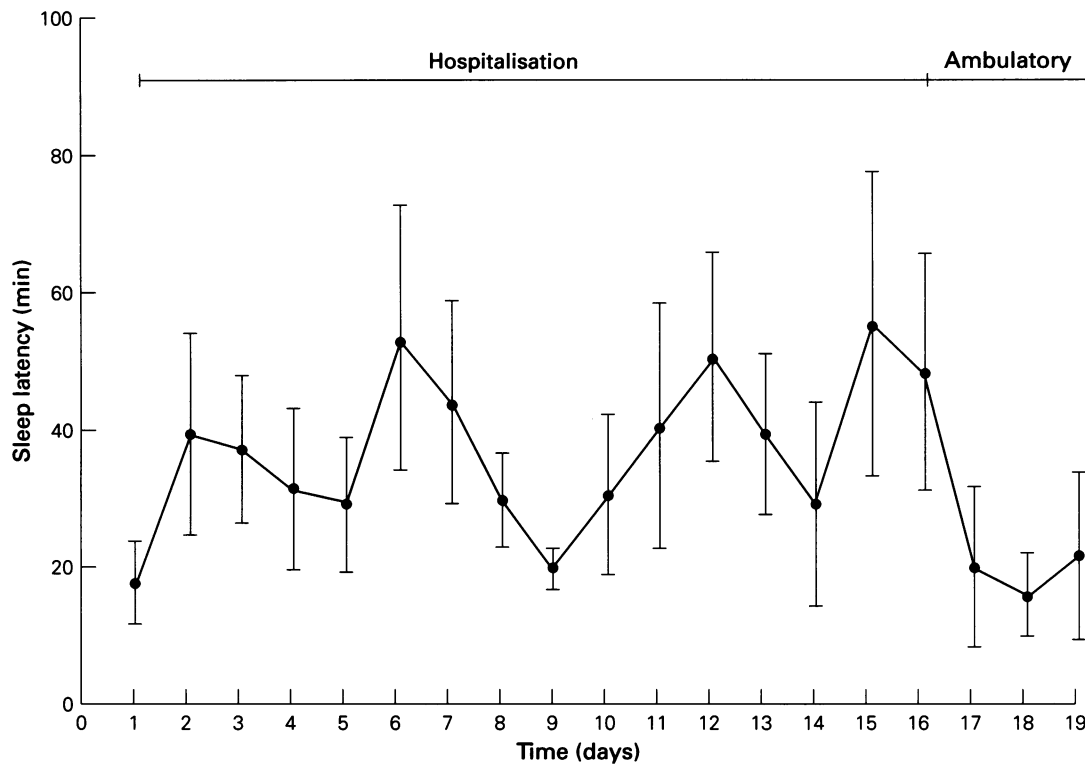


Figure 3 Mean (\pm s.e. mean) self-rated sleep latency (min) in nine healthy subjects given a placebo for 19 days (in the hospital for 16 days, then as outpatient treatment).

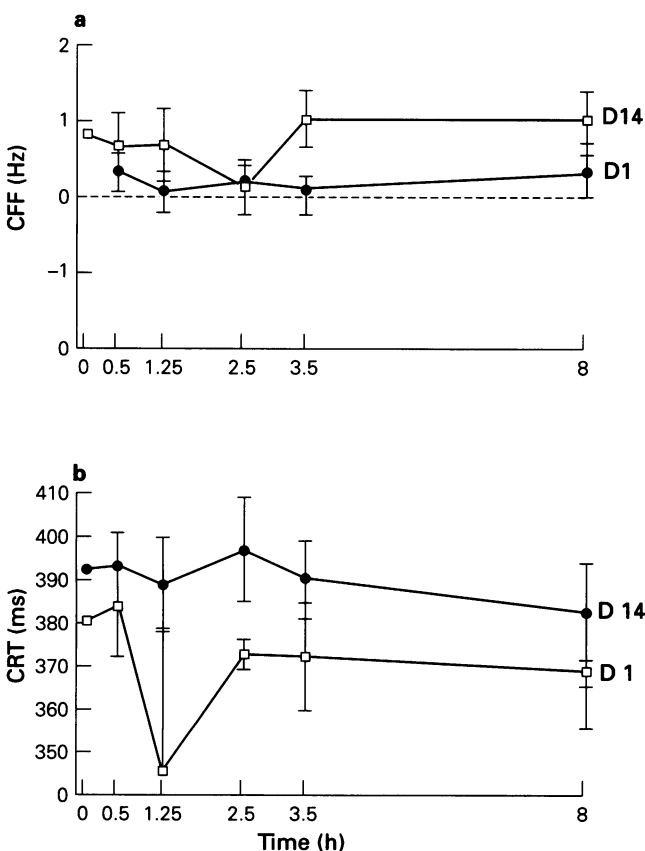


Figure 4 Mean difference from baseline (\pm s.e. mean) a) CFF (Hz) and b) CRT (ms) on day 1 and day 14 of placebo treatment in nine hospitalised healthy subjects.

an example of the influence of bed rest and reduced physical activity of these metabolic parameters.

Discussion

The potential influence of experimental conditions on various physiological parameters was assessed by studying variations in these parameters during placebo periods of several phase 1 studies involving hospitalisation for at least 2 weeks. Observed variations included increases in heart rate, sleep latency, and self-rated tiredness, and enhanced psychometric performance. Many factors may be implicated in these observed effects during the 'placebo period' in healthy volunteers. Young male volunteers for phase 1 trials have a personality structure that is high in extraversion and low in neuroticism and psychoticism [7]. Differences in extraversion score vigilance task performance have also been described [8, 9]. Assertive, independent subjects identified using the Cattell 16PF personality test were less likely to respond to a placebo during a mental stress test done as part of a study of alprazolam [10]. Housing and group effects may also influence the response of volunteers. In two studies intended to assess the reality of the 'Chinese restaurant syndrome' (headache attributed to glutamate additives in Chinese food), volunteers were given beef soup containing either a placebo or monosodium glutamate, according to a double-blind crossover design. The two study

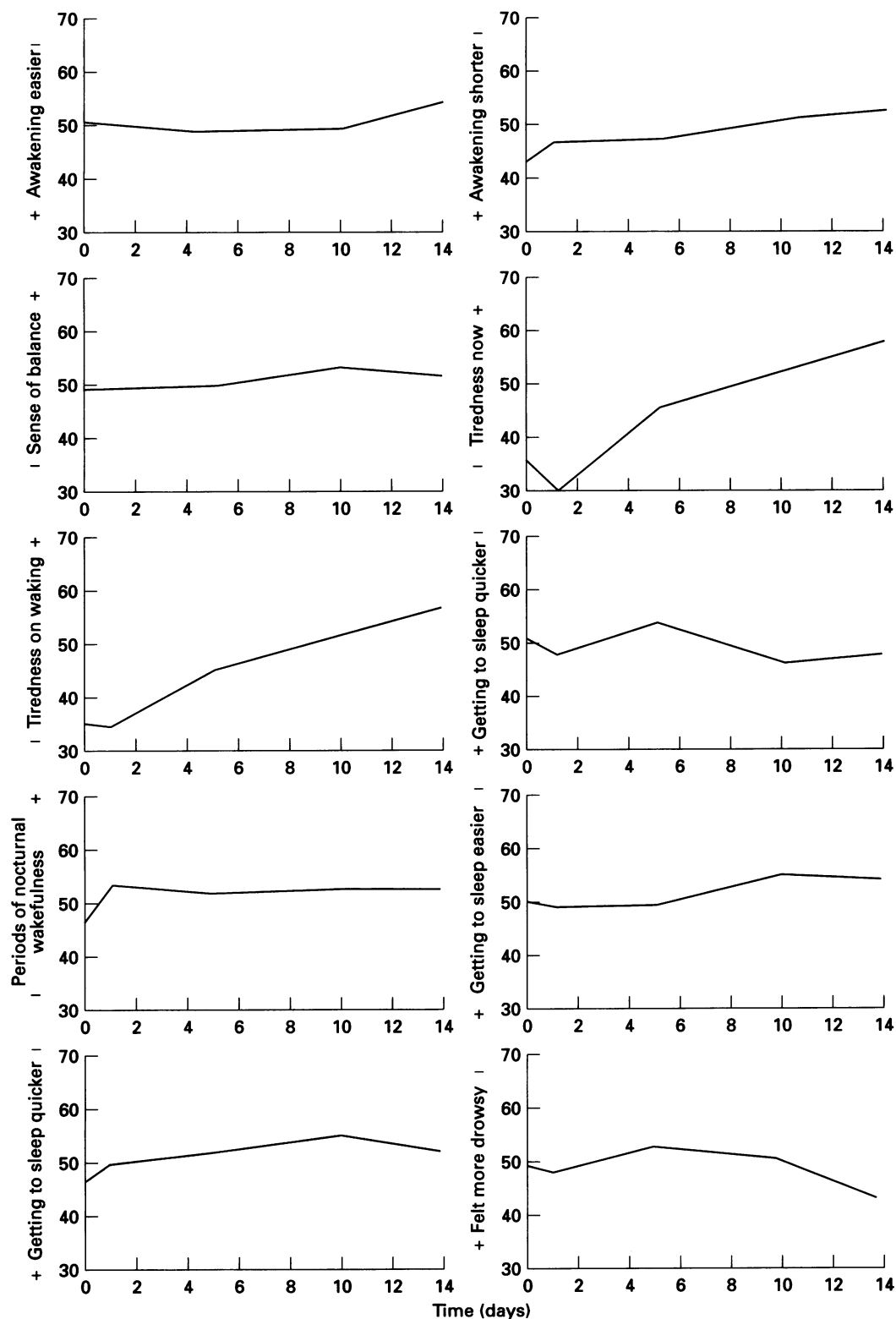


Figure 5 Leeds Sleep Evaluation Questionnaire in six healthy subjects given a placebo for 14 days (self-comparison to usual situation).

protocols were very similar, but differed with respect to housing conditions; 71% of 24 subjects who were required to share the same room for 8 h (first study) reported at least one adverse event, *vs* less than 10% of 73 subjects divided into small groups and allowed freedom of movement in the centre (second study) [11, 12].

Decreased physical activity may also affect homeostasis. During a 7 day study, severe restriction of physical activity resulted in a significant decrease in body weight (due to a loss of muscle mass) and in a decrease in maximum oxygen uptake [13]. Furthermore, several authors have reported increases in glucose-induced insulin level after bed rest [13, 14].

Table 2 TYR 30 criteria (amount of tyramine which induced a greater than 30 mm Hg increase in SBP) under fasting, placebo, and befloxtone conditions, and time to recovery according to the treatment condition (placebo or no drug) in 10 healthy hospitalised male volunteers

Volunteer	Fasting TYR 30 (mg)	Placebo run-in TYR 30 (mg)	Befloxtone 20 mg once daily TYR 30 (mg)	Recovery Condition	Recovery Duration (days)
1	400	1000	350	Placebo	3
2	600	1600	150	Placebo	4
3	400	1600	250	Placebo	3
4	600	1200	300	Placebo	3
5	600	1800	500	No drug	1
6	400	600	200	No drug	1
507	400	1400	400	No drug	1
8	600	1800	200	No drug	1
9	400	800	250	No drug	1
510	600	1600	300	No drug	1
Mean	488.9	1340.0	290.0		
s.d.	105.4	422.2	104.9		—

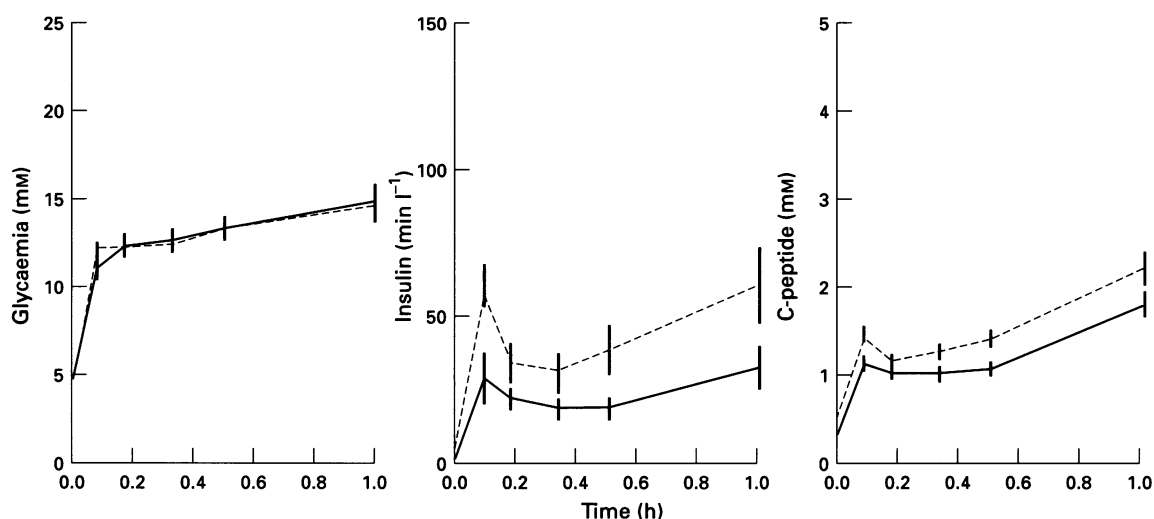


Figure 6 Glycaemia, insulinaemia, and C peptide levels after 1 day (— first GIT) and 12 or 13 days (--- second GIT) of placebo treatment.

Experimental conditions of phase 1 trials have also been described [15–17] as potentially responsible for changes in serum transaminase levels (perhaps ascribable to dietary factors), which may be confusing at this early stage of drug development. Frequent blood sampling during repeated dose studies do have an impact on haemodilution as assessed by decrease in haematocrit and may therefore have also an impact on haemodynamics. The influence of biological rhythms may also affect findings during placebo treatment. For example, sleep latency (Figure 3) seems to oscillate according to an ultradian rhythm with a period of about 5 to 7 days.

Chronopharmacology factors should be taken into account when designing pain studies or endocrine studies [18] but, for practical reasons, this is not always feasible during safety studies primarily aimed at identifying major tolerability problems.

Our tyramine test results during recovery after befloxtone treatment (Table 2) provide a new

example of a conditioning model of placebo effects. This conditioning model has been elegantly demonstrated in a crossover study in hypertensive patients [19]. After discontinuation of atenolol, the antihypertensive response was significantly greater when a placebo was given than in the absence of any treatment. It is not clear to what extent the recorded changes are due to the experimental situation, the placebo medication or both. Accurate evaluation of placebo effects in healthy volunteers would need a no-drug-at-all period, but this would not allow binding, unless the study drug was mixed with food, an approach which would not always be feasible (bad taste, modified release form, ...).

This retrospective study, designed to identify potential changes in several physiological parameters during repeated-dose studies in healthy volunteers, emphasised that use of a placebo-controlled design is essential and that within-group comparisons should not be included in statistical analyses. In particular,

the identification of so-called 'first-day effects' or 'tolerance' during these trials should be viewed with scepticism, since changes ascribable solely to the experimental conditions may confound the results, in particular with drugs that affect the sympathetic system. If one assumes that the level of stress is greater on the first treatment day, then the higher sympathetic tone might affect, for example, the activity of a pre-synaptic α_2 -adrenoceptor blocking agent but not that of the placebo, resulting in a

spurious first-day effect. In such cases, the 'first day effect' should be assessed by blindly and randomly incorporating the first day of active treatment in a run-in placebo period of variable length.

In conclusion, this retrospective study was not aimed at reaching definite conclusions but rather at providing evidence supporting the need to conduct and assess clinical pharmacology studies vs a placebo and to exercise caution when interpreting conclusions based on within-group comparisons.

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