Effect of the NK$_3$ receptor antagonist, talnetant, on rectal sensory function and compliance in healthy humans

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Abstract *Visceral hypersensitivity is important in the pathophysiology of irritable bowel syndrome and thus a target for modulation in drug development. Neurokinin (NK) receptors, including NK$_3$ receptors, are expressed in the motor and sensory systems of the digestive tract. The aim of this study was to compare the effects of two different doses (25 and 100 mg) of the NK$_3$ receptor antagonist, talnetant (SB223412) with placebo on rectal sensory function and compliance in healthy volunteers studied at two centres. Rectal barostat tests were performed on 102 healthy volunteers, randomized to receive either oral talnetant 25 or 100 mg or placebo over 14–17 days. Studies were performed on three occasions: day 1 immediately prior to 1st dose, day 14 h postdose, and after 14- to17-day therapy. Compliance, and pressure thresholds for first sensation, urgency, discomfort and pain were measured using ascending method of limits, and sensory intensity ratings for gas, urgency, discomfort and pain determined during four random phasic distensions (12, 24, 36 and 48 mmHg). Talnetant had no effect on rectal compliance, sensory thresholds or intensity ratings compared with placebo. In general, the results obtained at the two centres differed minimally, with intensity scores at one centre consistently somewhat lower. At the doses tested, talnetant has no effect on rectal compliance or distension-induced rectal sensation in healthy participants.*

Keywords *antagonist, ascending method of limits, neurokinin, perception, sensory intensity ratings, visceral.*

INTRODUCTION

Visceral hypersensitivity, the excessive perception of visceral stimuli that are not usually painful, is considered an important determinant of functional bowel disorders and visceral pain.[1–3] Novel therapies for these disorders are being sought amongst drug classes that are proven to modulate afferent neurotransmission in animals or to reduce the perception of gut stimuli in humans.[4]

Neurokinin (NK) receptors are involved in both motor and sensory functions of the gastrointestinal tract.[5] Tachykinins are neurotransmitters that interact with NK receptors in the enteric nervous system and in the visceral sensory system. They include substance P (SP) and neurokinin A (NKA) and neurokinin B (NKB), which exert their activity by preferentially binding to the NK$_1$, NK$_2$ and NK$_3$ receptors respectively. However, the differences in affinities are small and considerable potential exists for overlap of action between each neuropeptide and each receptor type.[5–7] To date, most of the research into the effects of tachykinins on gastrointestinal function has focused on the roles mediated via NK$_1$ and NK$_2$ receptors. In animal models, the NK$_1$ receptor appears to play a significant role in pronociception, while the NK$_2$ receptor may have a greater influence on smooth muscle contractility.[6,8,9] Studies

in humans, however, are limited, with one small study in irritable bowel syndrome (IBS) patients showing that the NK_1 receptor antagonist, ezlozipant, decreases the anger response and tends to reduce visceral sensitivity to rectosigmoid distension.[10] In healthy volunteers, the NK_2 receptor antagonist, nepadutant reduces contractile frequency and amplitude of migrating motor complexes in the small intestine and effectively antagonizes the motility-stimulating effects of infused NKA.[11]

More recently, the advent of new and selective antagonists at the NK_3 receptor has allowed the gastrointestinal activities of tachykinins be more fully explored. NK_3 receptor antagonists can reduce rat viscero-motor response to noxious levels of colo-rectal distension.[12–16] Furthermore, the use of an antagonist with no demonstrable ability to cross the blood–brain barrier, suggested that the mechanism of this antinociceptive activity does not necessarily depend on activity within the brain and spinal cord.[15] Consequently, a hypothesis was developed that NK_3 receptor antagonists, which have little or no activity on normal gastrointestinal motility, could reduce the effects of tachykinins released from enteric intrinsic primary afferent neurons (IPANs), following activation by a 'supra-maximal' or noxious stimulus.[17] In addition, to modulating C-fibre nerve terminal sensitivity, these effects included an ability to influence the sensitivity of connecting IPANs, influencing enteric nerve sensitivity and prolonging peristalsis during periods of intense intestinal distension.[18]

Thus, the general aim of this study was to evaluate the significance of these findings in humans in two separate centres using talnetant (SB223412), an orally active, competitive, non-peptide, selective NK_3 receptor antagonist which was previously demonstrated to be effective as an intestinal antinociceptive agent in animal models.[15,17]

The specific aim of the present study was to compare the effects of 25 and 100 mg doses of talnetant *vs* placebo on rectal sensation and compliance during standardized distensions with the barostat in healthy volunteers. The studies were performed in two separate centres and the comparison across the two centres and overall reproducibility of rectal sensation and compliance under control conditions have been previously published.[19]

METHODS

Participants, study design and questionnaires

One hundred and two healthy volunteers [60 females; age, 27 ± 1 years (mean ± SEM); body mass index (BMI), 24 ± 0.5 kg m^{-2}] were enrolled in this study with

similar numbers enrolled in each centre. All participants signed informed consent, and this study was approved by the Mayo Clinic Institutional Review Board and South Manchester Research Ethics Committee. Current co-morbidities, abdominal symptoms, previous abdominal surgery and concomitant somatization or psychological disorders were excluded by means of a validated bowel symptom questionnaire, a somatic checklist and a physical exam.[20] In order to test for a potential association between anxiety or fear of pain and the sensitivity thresholds or sensory ratings, subjects also completed the 30-item Fear of Pain Questionnaire [FPQ-III][21] before each barostat procedure and a revised version of the 36-item Anxiety Sensitivity Index [ASI-R][22] at screening. The rectal barostat procedures were performed using an identical sequence on three separate occasions at each centre: immediately prior to initial dosing (Day 1 baseline), 4 h postinitial dose (Day 1 postdose), and once after 14–17 days of drug exposure. The subjects were given a 150 mL glucose drink (Lemon Lucozade Sport, 118 kJ (28 kcal) per 100 mL; SmithKline Beecham, Brentford, UK) 1 h prior to setting baseline operating pressure (BOP) on each of the three test occasions. This was performed to provide standard hydration and minimal calories in participants who would have fasted from midnight and were embarking on 9 h of studies on the first study day.

Talnetant

Talnetant is a selective NK_3 receptor antagonist, which has high affinity for the human form of the receptor (Ki value of 1.0 nM). The solubility of talnetant results in less than proportional bioavailability with increasing dose. In humans, talnetant is rapidly absorbed with a median time to reach maximum concentration typically occurring at 4.0 h postdose. The human pharmacokinetics is moderately variable. Thus, the between-individuals coefficient of variation is typically 15–30% after repeat dosing. Moreover, pharmacokinetics are non-linear after repeat dosing so that the increase in plasma exposure is less than proportional to dose administered. It has a mean estimated elimination half-life of 35–40 h, which is dose-independent. The 25 and 100 mg doses were chosen based on the expected plasma exposures extrapolated from those seen in young adult healthy volunteer phase I pharmacokinetic studies, with the addition of pharmacokinetic modelling to identify a top dose that was felt to be adequate to match the animal data. These exposures were associated with no clinically significant effects on blood count, electrolytes, liver enzymes, reproductive hormones.[23] Talnetant is generally well-tolerated.

Headache is the most common reported adverse event; although, in controlled studies, often the incidence of headache was similar in the placebo and talnetant dose groups. No clinically relevant findings related to talnetant have been observed in laboratory parameters, including liver function tests or electrocardiograms.

Drug administration and assessment of compliance

Participants were randomly allocated, using a computer-generated sequence, into equal groups, to receive either a 14- to 17-day course of oral talnetant, 25 or 100 mg, or matching placebo. The window of 14- to 17-day treatment was allowed to provide some flexibility in scheduling the last barostat testing. The investigators and the study statistician were blinded of the allocation sequence, which was kept by the study pharmacists. Blood samples to measure talnetant plasma concentrations were taken at Day 1 predose and as a trough sample on the last day of dosing in the 42 last subjects (14 on placebo, and 13 on 25 mg and 15 on 100 mg talnetant) of the study across both research centres. Compliance with the study drug administration scheme was measured by counting the returned pills at the end of the study period.

Rectal barostat equipment and procedure

All subjects presented to the research centres of the Mayo Clinic and the University Hospital of South Manchester after bowel preparation [Fleet® phosphate enema (C.B. Fleet Co., Lynchburg, VA, USA and Laboratories Casen-Fleet SA, Zaragosa, Spain), self-administered at least 1 h before reporting to the centres] and an overnight fast. A catheter (customized rectal barostat catheter, part no. C7-2CB-R-22F; MUI Scienti-

fic, Mississauga, ON, Canada), to which was attached a polyethylene bag (Pillow Type Rectal Barostat Balloon, part no. CT-BP600R; length, 22 cm; diameter, 15 cm; capacity 600 mL; MUI Scientific), was inserted into the rectum so that the middle of the balloon was located approximately 10 cm from the anal verge. To decrease the effects of abdominal viscera on the balloon volume, the subjects were placed in a semi-prone position and the foot end of the bed elevated 15°. The bag was then unfolded by transiently inflating it with 75 mL of air and then deflating it completely. After a 20- to 30-min recovery period, the catheter was connected to a barostat (G&J Electronics Inc., Toronto, ON, Canada) and the pressure in the bag increased from 4 mmHg in steps of 1 mmHg for 1 min per step until respiratory excursions were observed. The baseline operating pressure was defined as 2 mmHg above the minimal distension pressure at which respiratory excursions were clearly recorded from the barostat tracing. If respiratory variations were not seen by 18 mmHg, BOP was set at 12 mmHg. An initial 'conditioning' distension of the rectum was then performed in which the pressure was increased from 0 mmHg in steps of 4 mmHg for 15 s per step until 20 mmHg was reached. Previous studies have shown that an initial 'conditioning' distension to 20 mmHg renders subsequent assessments of compliance and perception more reproducible.[24] The bag was then deflated to 0 mmHg and the subjects were allowed to rest for 10 min before proceeding to the ascending method of limits.

The experimental design is shown in Fig. 1.

Ascending method of limits: compliance and sensory thresholds

Rectal compliance and sensory thresholds were measured by ramp inflation, starting at 0 mmHg and
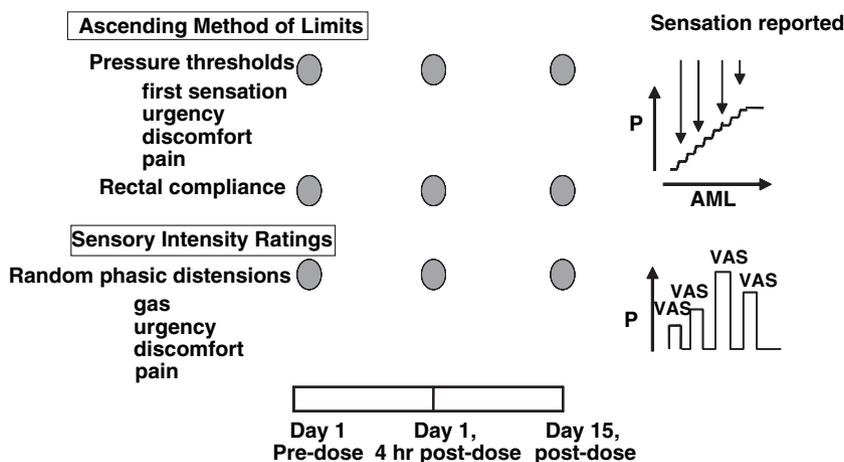

Figure 1 Experimental design of the study.

increasing in steps of 4 mmHg for 1 min per step to a maximum of 60 mmHg. Thresholds for first sensation, urgency, discomfort and pain were indicated by the subjects by pressing a button at the distension pressure at which sensations were perceived. Ramp inflation was terminated as soon as the subjects reported the first sensation of pain. Following this procedure, the bag was deflated to BOP and the subjects allowed to rest for 10 min.

Random order phasic distensions: sensory intensity ratings

After the ascending method of limits' protocol, phasic distensions of 12, 24, 36 and 48 mmHg above BOP were each applied once in random order. Each distension was maintained for 60 s with an inter-stimulus interval of 2 min during which the balloon was deflated to BOP. This approach has been shown to be reliable in multiple previous studies, as the distension pressure is associated with sensory intensity ratings that are generally proportional to the magnitude of the distension pressures.[25–27] Study participants were blinded to the distension order, which was provided by the study statistician (A.R.Z.) and was also randomized between study days. Subjects were asked to mark four separate 100-mm visual analogue scales (VAS) 30 s after the onset of the distension for the sensations of gas, urgency, discomfort and pain. These scales were anchored at each end by the descriptions 'unnoticeable' and 'unbearable'. Pressure was immediately released if the subject reported greater than 80 mm of discomfort or pain on the VAS scale and higher distensions were not subsequently administered. During the assessment of sensation, the interaction between the subject and the study investigator was kept to a minimum.

Data analysis

The following measurements were derived: (i) the sensory thresholds for first sensation, urgency, discomfort and pain during ascending method of limits, (ii) the aggregate sensation score (gas, urgency, discomfort and pain) in response to the four random phasic distensions (12, 24, 36 and 48 mmHg above BOP) and (iii) rectal compliance.

Since rectal pressure-volume relationships are sigmoidal, compliance was summarized, as in previous studies,[26–28] using a power exponential model for the proportional volume (observed volume at each pressure divided by the maximum observed volume) as a function of 1/pressure : $Vol/Vol_{max} = r + \exp[-(\kappa \times 1/Pr)^{\beta}]$,

where r represents the minimum observed volume divided by the maximum observed volume (Vol_{max}), Pr is pressure, and κ and β are the model parameters that are estimated for each compliance curve. A summary value for each compliance curve was then calculated from these parameter estimates for each subject, specifically the pressure observed at one half of the maximum observed volume ($Pr_{1/2}$), where a smaller $Pr_{1/2}$ corresponds to higher compliance.

Scores from the 'Fear of Pain' questionnaires (administered prior to each of the three barostat tests) and from the 'Anxiety Sensitivity' questionnaire (administered at baseline) were computed for each subject by first computing the mean value over all non-missing items on each subjects questionnaire (overall and for the three subscales for fear of pain: severe, minor and medical). The mean values were then multiplied by the corresponding total number of questions on each questionnaire (subscale). This yielded 'sum' scores that assumed any missing values for individual questions would have the mean value for that subject over all their non-missing responses.

Statistical analysis

Sensation pressure thresholds A 'time to event' approach (i.e. pressure required to evoke specific sensations) was used to assess four sensation thresholds (first sensation, urgency, discomfort and pain), separately for the two study sessions. As some threshold levels are 'censored' (e.g. pain not evoked by the max distension used), a Kaplan–Meier summary of the pressure thresholds was used to univariately depict the cumulative probability of evoking the sensation, and proportional hazards regression models were used to assess treatment group effects separately for each of the sensation types at each of the two study sessions. The covariates included in these models were the subject-specific corresponding sensation pressure threshold for the predrug study, the maximum volume reached in the predrug study, the predrug 'Fear of Pain' subscale scores (Severe, Minor, Medical), the 'Anxiety' score, and gender. Main effect terms for drug dose and study centre were used in these models.

Sensory intensity ratings The assessment of differences in observed sensory intensity ratings (rectal sensation VAS scores) at the two study centres were based on analysis of covariance (ANCOVA) for repeated measures (the four phasic distension levels) separately for the two distinct study sessions (day 1 postdrug and day 14 postdrug). Each type of VAS score was analyzed individually (gas, urgency, discomfort and pain). The

covariates in these models were gender, the per subject mean (over pressure distension levels) VAS score pre drug, the 'Fear of Pain' subscale scores, the 'Anxiety' score, and the corresponding pressure (12, 24, 36, 48 mmHg) at each phasic distension level. Each of the ANCOVA models included treatment group (drug dose), and study centre main effect terms, as well as two-way interaction terms for treatment group by centre, treatment group by pressure level, centre by pressure level and the three-way interaction term for treatment by centre by pressure.

Rectal compliance The κ and β parameters for the fitted compliance curves were estimated using the NLIN procedure in the SAS software package,[29] and used to calculate the $Pr_{1/2}$ value for each compliance curve for each subject.

An analysis of covariance was then used to compare the effects of drug and placebo on the calculated compliance ($Pr_{1/2}$) values separately for each of the two study sessions. Treatment group, centre and the treatment group by centre interaction term were examined in these models incorporating age, gender, BMI and the corresponding $Pr_{1/2}$ value from the predrug study in each subject as covariates.

Sample size calculation Based on previous data, we estimated that a sample size of 16 per treatment group would be capable of detecting a 38% treatment difference on the primary outcome variable (rectal pain sensation threshold) with 80% power. Thus, we planned that each centre would recruit 16 subjects per treatment group. If the coefficient of variation for sensory measurements between centres was similar, the combined group size of 32 subjects per treatment group would have 80% power to detect a treatment difference of approximately 26% and 40% for pain sensation and first sensation respectively. These are based on a two-sample *t*-test (with a two-sided $\alpha = 0.05$), which does not account for potential censored values, but the proportional hazards approach incorporated relevant covariates which should have provided similar power for somewhat smaller differences.

RESULTS

Participants, trial flow and conduct of study

The demographics and characteristics of the participants appear in Table 1. A total of 102 subjects were

Table 1 Baseline characteristics (Day 1 immediately prior to dosing)

	Rochester			Manchester		
	25 mg	100 mg	Placebo	25 mg	100 mg	Placebo
Age (years)	31.2 ± 1.9	30.2 ± 1.7	29.4 ± 2.2	26.4 ± 2.0	24.7 ± 1.8	26.2 ± 1.7
No. of females	10	9	13	10	7	11
No. of males	7	7	4	7	11	6
BMI (kg m^{-2})	25.6 ± 1.3	25.2 ± 1.1	25.5 ± 1.4	23.1 ± 0.7	23.4 ± 0.8	24.5 ± 1.3
Anxiety score	116.7 ± 20.7	98.1 ± 18.5	96.9 ± 14.2	218.4 ± 35.7	208.9 ± 32.6	168.5 ± 31.7
FOP (overall)	53.5 ± 3.8	58.0 ± 4.1	56.7 ± 4.7	77.3 ± 4.7	75.3 ± 4.8	79.3 ± 4.6
FOP (severe)	23.9 ± 2.1	27.2 ± 2.6	24.6 ± 2.7	35.2 ± 2.0	33.3 ± 2.1	35.4 ± 2.1
FOP (minor)	12.6 ± 0.7	13.1 ± 0.8	13.1 ± 0.9	19.1 ± 1.7	17.6 ± 1.7	19.3 ± 1.4
FOP (medical)	16.9 ± 1.5	17.7 ± 1.3	19.1 ± 1.9	22.9 ± 1.7	24.0 ± 1.7	24.6 ± 1.8
BOP (mmHg)	9.0 ± 0.6	8.2 ± 0.4	9.1 ± 0.6	7.6 ± 0.4	8.3 ± 0.4	8.4 ± 0.7
Compliance ($Pr_{1/2}$)	12.8 ± 0.9	12.4 ± 0.8	12.1 ± 0.9	15.7 ± 1.2	17.7 ± 1.8	17.9 ± 1.7
Threshold (mmHg)						
First sensation	9.6 ± 0.9	10.0 ± 2.1	9.4 ± 1.2	11.5 ± 1.6	7.4 ± 1.5	10.4 ± 1.5
Urgency	23.3 ± 2.2	22.0 ± 2.1	20.2 ± 2.1	19.8 ± 2.0	18.4 ± 1.4	20.7 ± 1.2
Discomfort	25.2 ± 1.7	23.8 ± 1.9	23.1 ± 3.1	28.5 ± 3.2	23.8 ± 1.5	27.1 ± 1.4
Pain	39.8 ± 2.2	42.3 ± 2.9	37.9 ± 3.4	38.1 ± 3.6	36.7 ± 2.9	36.2 ± 2.6
Intensity rating						
Gas/bloating	56.9 ± 4.2	50.4 ± 4.3	52.7 ± 4.6	55.8 ± 4.4	53.8 ± 4.9	51.9 ± 3.3
Urgency	61.4 ± 3.2	56.0 ± 2.7	57.2 ± 4.0	57.0 ± 4.5	53.7 ± 4.6	51.0 ± 3.0
Discomfort	59.4 ± 3.0	52.0 ± 3.7	55.5 ± 3.9	47.8 ± 4.9	47.0 ± 5.0	47.0 ± 2.3
Pain	46.2 ± 5.0	29.9 ± 4.8	41.9 ± 5.5	33.1 ± 4.7	26.4 ± 4.4	38.1 ± 3.5

Sensory ratings shown are mean values per subject over the different distension levels by centre and treatment group. These data provided the covariate values used in the ANCOVA models in the post-treatment comparisons. Data presented as mean ± SEM. BMI, body mass index; FOP, fear of pain.

enrolled in the study; 48 subjects completed the study in Rochester and 48 in Manchester, with no reported significant side effects. A total of six subjects (two, Rochester, four, Manchester; 2, 3 and 1 subjects in the 25, 100 mg, and placebo dosing groups respectively) did not complete the study. The reasons for not completing the study were: withdrawal due to non-compliance to study protocol ($n = 1$); adverse event assigned by the principal investigator as unrelated to study medication ($n = 3$) or as suspected (reasonable possibility) to be related to study medication ($n = 1$: this subject was identified at study analysis as in the placebo treatment group); and one serious adverse event assigned by the principal investigator as unrelated to study medication ($n = 1$). There were no significant differences in age, gender distribution, BMI, anxiety score and fear of pain across centres and between treatment groups (Table 1). As the protocol required that distension sequences be interrupted if the subjects reported pain, data for sensation ratings at the highest pressures (during phasic distensions) were available for at least 22 participants per treatment group at 48 mmHg and for at least 27 participants per treatment group at 36 mmHg pressures. All participants completed the distensions during ascending method of limits. Seven subjects did not have compliance curve values for the day 15 study, and one of these also did not have a value for the initial postdrug study. Overall reproducibility data across the two research centres of rectal sensation and compliance under control conditions have been previously published.[19]

There was a modest association of centre with BOP ($P = 0.047$); the Manchester centre having slightly lower values across all treatment groups. However, as the actual (average per person) rectal tone level during predrug studies was used as a covariate in the analyses of tone, the analysis accounted for these differences in BOP. There were also associations of centre with age, anxiety scores, fear of pain (FOP) score and predrug compliance ($Pr_{1/2}$).

In summary, sensation and compliance prior to treatment were similar amongst the three groups of subjects receiving talnetant (25, 100 mg) or placebo (Table 1). There was no difference in comparisons of drug *vs* placebo for sensation and compliance in either of the two centres.

Rectal compliance using ascending methods of limits

There were no significance effects of talnetant, 25 or 100 mg, on rectal compliance compared with placebo control (Fig. 2). There was also no significant interac-

Figure 2 Effect of talnetant on rectal compliance on Day 1, 4 h postdose, and on Day 15. $Pr_{1/2}$ is pressure at half maximum value. Note there is no effect of talnetant on rectal compliance.

tion of treatment effects with age, gender, BMI or study centre on rectal compliance, although a significant overall centre effect was detected ($P = 0.015$) on the first postdose assessment period on day 1, but not at the second assessment on daylight hours 14 ($P = 0.20$). Note that the $Pr_{1/2}$ value on study day 1, 4-h postdose, for one centre was 14.1 ± 0.5 mmHg and for the other centre, it was 12.1 ± 0.5 mmHg.

Sensation pressure thresholds using ascending method of limits

The effects of test medication on thresholds for first sensation, urgency, discomfort and pain to distensions on Day 1 (4-h postinitial dose) and Day 15 (Days 14–17 post-treatment) are shown in Fig. 3. The figures plot the proportion of participants that reach specified thresholds with increasing pressure distensions. Note that there is a progression from left to right as volunteers experienced first sensation, urgency to have a bowel movement or discomfort (which overlapped) and pain. However, it also worth noting that the plots show that there were no differences amongst treatments for the proportion of patients reaching their threshold pressures for the different sensations. Thus, there were no differences noted in threshold responses to talnetant *vs* placebo. No associations with 'Fear of Pain' scores or 'Anxiety' scores were detected, and no significant centre effects were detected for either study session.

Sensory intensity ratings using random order phasic distensions

The effects of medication on sensory intensity ratings to distensions on Day 1 and Day 15 post-treatment are
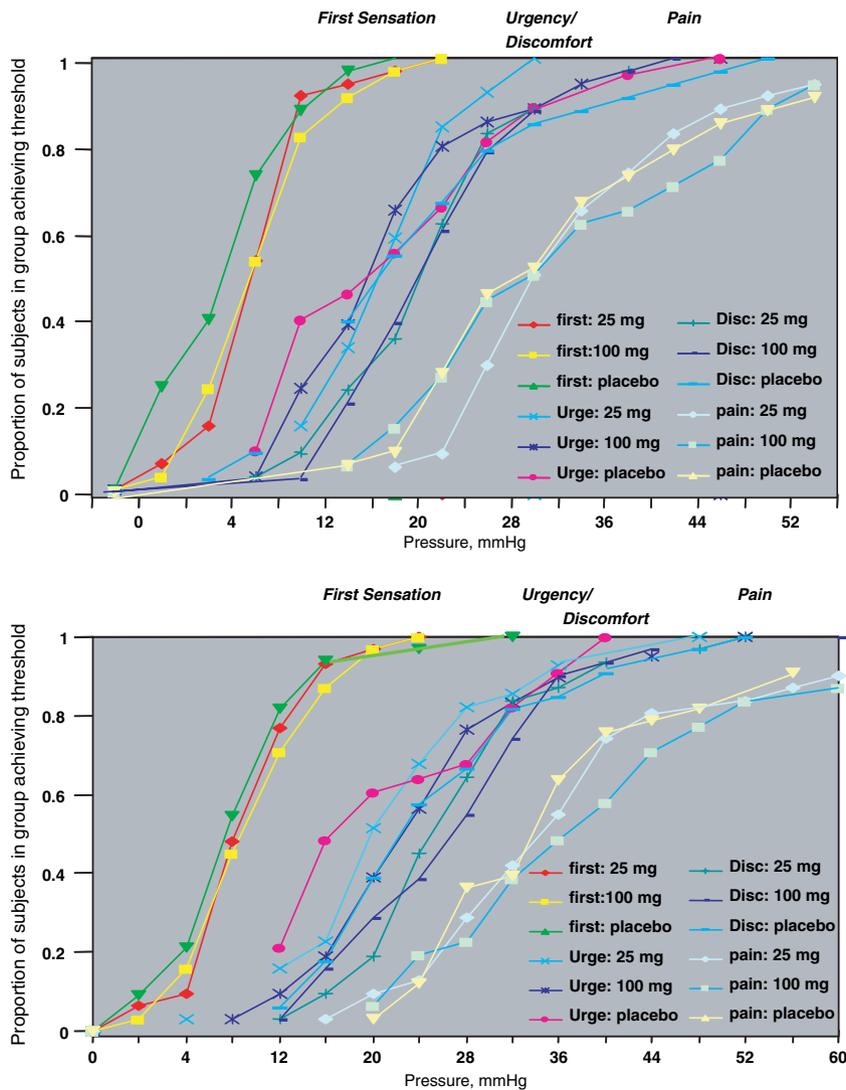
Figure 3 Sensory thresholds in the two-centre study. Note the lack of effect of talnetant on different sensory thresholds Day 1 (4 h postdose), upper panel; and Day 15, lower panel. First, first sensation threshold; urge, urgency threshold; disc, discomfort threshold; pain, pain threshold.

shown in Figs 4 and 5. There are no differences noted in response to medications. On study Day 1, a borderline significant centre by treatment interaction was observed ($P = 0.076$) for the discomfort scores, and on study Day 15, a three-way interaction pressure by centre by treatment was detected ($P = 0.005$) for gas/bloating scores. The interaction effects detected on day 15 for gas/bloating scores implied that treatment effects depended in the distending pressure, but this dependence varied between centres. In general, the magnitude of the overall differences between centres (roughly 5–10 on the 100-mm VAS scale) is likely not clinically relevant.

Plasma concentrations

Though talnetant plasma concentrations were 1635 ng mL^{-1} (median, range 127–2472 ng mL^{-1}) and

3470 ng mL^{-1} (242–10080 ng mL^{-1}) for the 25 and 100 mg doses, respectively, they were consistent with the pharmacokinetics observed in other studies in humans[23] for the formulation, doses and duration of dosing used in the present study.

DISCUSSION

This randomized, two-centre, placebo-controlled study is the first to investigate the effect of a NK$_3$ receptor antagonist on visceral sensation and on rectal compliance in healthy volunteers. The study shows that, at the doses tested, talnetant, a selective NK$_3$ receptor antagonist, did not significantly influence visceral sensation or compliance of the rectum in healthy subjects. However, these findings may not be generalizable to IBS patients, in whom anxiety or fear of pain levels may be higher and in whom increased
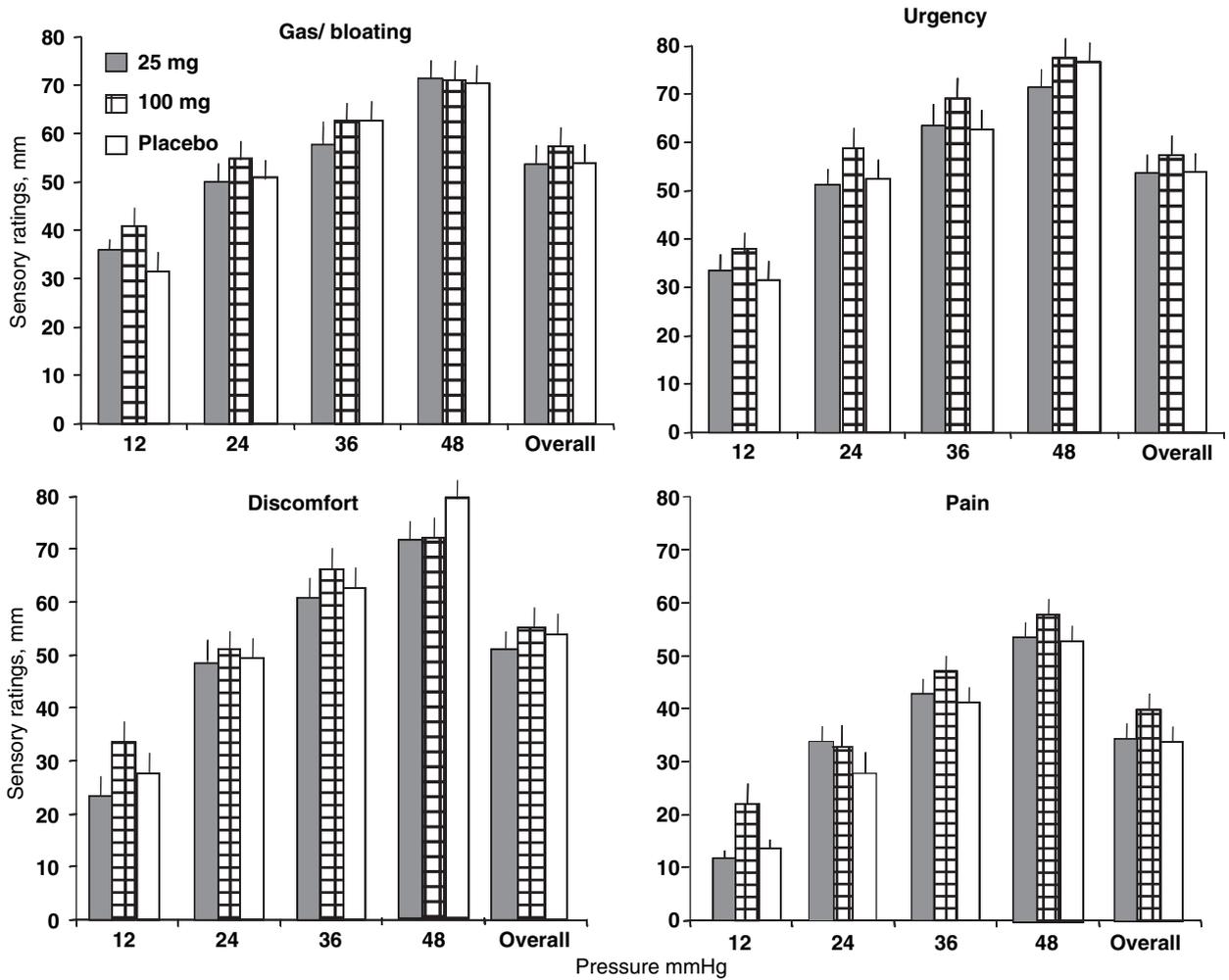
Figure 4 Effect of placebo and talnetant on sensory ratings, Day 1, 4 h postdose, obtained at 12, 24, 36 and 48 mmHg above BOP. The overall rating is the (least squares adjusted) mean values (over distension levels) for each treatment from the ANCOVA model. Note the lack of efficacy of talnetant, 25 or 100 mg, relative to placebo.

perception of non-noxious sensations has been reported.[30]

Visceral sensitivity can be quantified by measuring either the sensory threshold or intensity rating in response to a stimulus, such as distension or electrical stimulation.[31] Intensity is rated while applying a known stimulus, and measuring the perception, typically using VAS[31] while a threshold is determined by applying an increasing stimulus, pressure, volume or electrical stimulus until a particular sensation, such as pain, is reported.[32] During measurement of thresholds with the ascending method of limits, the viscus compliance can be concomitantly assessed and its relationship to sensitivity determined. Thus, predefined and standardized distensions of the bowel wall using a barostat device constitute the current standard for the assessment of sensorimotor function in experi-

mental trials in health and disease.[31] We have previously reported that these rectal sensation studies can be standardized across two specialized centres and that the results for compliance, sensory thresholds and ratings in response to mechanical distension differed minimally across the two centres.[19] In the present study, these two well-validated techniques were performed at two different time points postdose. In both studies, talnetant had no effect on visceral sensation, strengthening our conclusion and suggesting that the lack of demonstrable effects of talnetant was not due to methodological issues.

The NK$_3$ receptor is one of three NK receptors that mediate the biological effects of tachykinins; they belong to the super family of seven transmembrane-spanning, G-protein-coupled receptors. The tachykinins are a family of small peptides distributed in both
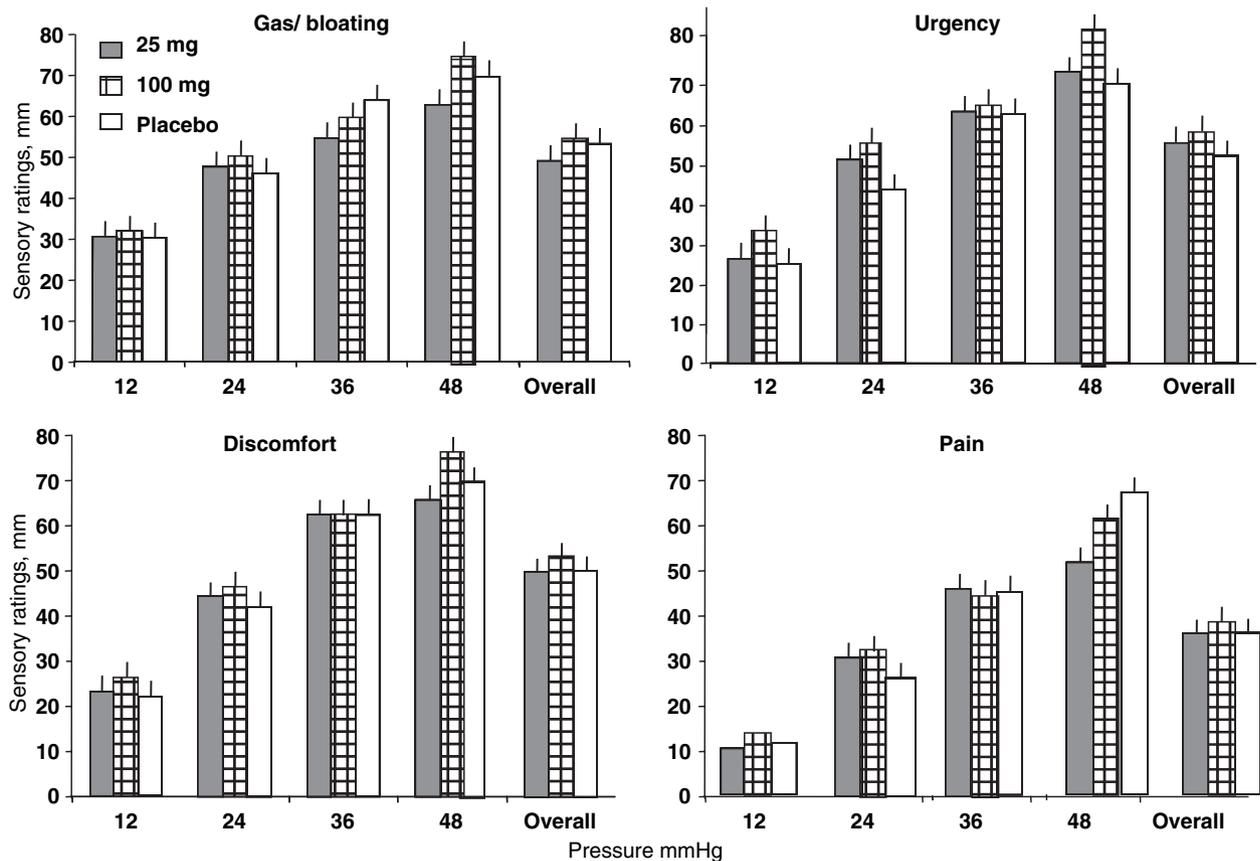
Figure 5 Effect of placebo and talnetant on sensory ratings, Day 15, obtained at 12, 24, 36 and 48 mmHg above BOP. The overall rating is the (least squares adjusted) mean values (over distension levels) for each treatment from the ANCOVA model. Note the lack of efficacy of talnetant, 25 or 100 mg, relative to placebo.

the central and peripheral nervous systems. In the latter, the predominant locations are the peripheral endings of capsaicin-sensitive primary afferent neurons (un-myelinated, sensory C fibres) which innervate many sites, notably the urinary and gastrointestinal tract, airways and the skin.[12,33,34]

In preclinical models, talnetant inhibits visceral pain and disturbed patterns of intestinal motility[15,17] in the absence of any significant changes in patterns of normal gastrointestinal motility.[15] In addition, NK_3 receptor antagonists have been shown to have potential anxiolytic and antidepressive effects in small animal models.[35,36] Given these anti-anxiety effects, we included validated questionnaires to measure anxiety and fear of pain[21,22] to assess their contribution to the outcomes of the sensitivity testing. At the doses tested however, talnetant does not appear to alter anxiety or fear of pain in healthy volunteers.

A fundamental difference in the animal studies is that they typically assess sensation with pseudo-

affective endpoints in the sensitized colon, whereas our human studies show that talnetant had no effect on normal visceral sensation. It is conceivable that the drug may have shown efficacy in the context of a sensitized rectum, such as in IBS patients with baseline hypersensitivity. While a theoretical possibility, it has not actually been demonstrated using barostat studies that any effective antinociceptive agent is effective in IBS but not in healthy volunteers.

Thus, for example, the somatostatin analogue, octreotide, is effective in both healthy controls and in IBS patients[37–40], and the μ-opiate agonist, fentanyl, reduces pain sensation (increased rectal sensory thresholds for pain) in response to distension in both healthy subjects and patients with IBS[41]. In animal models, the alpha$_2$ delta ligand, pregabalin, has been shown to reduce the sensitized, but not the normally sensitive, rectum.[42,43] This medication reduced sensation in IBS patients with rectal hypersensitivity,[44] although no study to date has been carried out in normally sensitive IBS patients or healthy controls to prove unequivocally

that the drug's efficacy is restricted to those with hypersensitivity.

The animal models using pseudo-affective endpoints involve brain stem reflexes and do not necessarily reflect the central circuits involved in the perception of visceral stimuli in humans. The literature documents several other examples of the discrepant conclusions between the pseudo-affective endpoints in animals and antinociceptive activities recorded in human studies and the challenge that this presents in the development of medications targeting visceral pain in humans.[45] Another explanation is also possible to explain the discrepancy between animal and human data. The peripheral effects of the barostat (mediated by spinal or brain stem reflexes) might have been altered by talnetant, but talnetant did not affect the ascending or descending pain pathways involving higher brain centres that may represent the major component of pain sensation in humans.

The lack of effect of talnetant in this study does not reflect a type two error, as a large sample size was used, or difference across the two centres, as there was generally a lack of study centre-by-treatment interaction. However, it is possible that the doses tested, 25 and 100 mg, were not high enough, as penetration into tissues is limited by high protein binding and low aqueous solubility.[23] However, in two double-blind, randomized, controlled studies in patients with IBS, exposures were tested up to 400 mg bid, and there was dose-dependent exposure without any evidence of efficacy on the primary endpoint of adequate response of IBS symptoms.[46]

Thus, it seems likely that the mechanism may not work in humans, and the present negative barostat study is consistent with the negative clinical effect; although the dose tested in the present study was only up to 100 mg. Moreover, since most drugs have failed clinical efficacy in IBS, the correlation that has been lacking for predictability of the barostat test is with positive studies predicting positive clinical effects. A further possibility for the lack of any demonstrable effect is that talnetant selectively blocks only the NK_3 receptor, whereas the endogenous ligands can bind to all three receptor subtypes. Thus, it may require concomitant blockade of more than one NK receptor subtype to obtain an effect on sensation due to rectal distension.

There are similar examples where lack of effect on visceral sensation of piboserod in healthy volunteers (using the same methods described here[47]) conflicted with information in animal studies[48] and even with the trend to improved rectal sensation studies (using a compliant latex balloon) in a small study of 15 patients with IBS.[49] Yet, the clinical trials performed with piboserod have not led to full reports of efficacy to date, despite the passage of more than 5 years as it was publicly acknowledged that the drug was in phase II trials.

Several studies suggest that modulating NKs through more or less specific NK antagonists has the potential to reduce pain sensation in animal models of IBS.[15,17,50–52] Future pharmacodynamic studies appear to be worth pursuing, given the discrepant results between animal models and clinical trials.[45]

The study included an assessment of the treatment by centre interaction. Essentially, there were some differences in the measurements of sensory ratings at different distension pressure levels; however, in general, there were statistically significant overall centre differences detected for most scores at both study sessions, with scores at the Mayo Clinic being consistently lower, but the magnitude of these differences (roughly 5–10 mm on the 100-mm VAS scale) is likely not clinically relevant.

In summary, the results showed no effect of the 25 and 100 mg doses of talnetant on rectal sensitivity. However, the present study demonstrated that a pharmacodynamic study can be carried out to provide results with adequate reproducibility and appropriate power to evaluate sensitivity, if it is large enough and even if it is performed in two different expert centres. This therefore supports the strategy of carrying out human pharmacodynamic studies before embarking on phase IIB clinical trials on the basis of results obtained using animal models of visceral pain.

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