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The effect of sodium nitroprusside on psychotic symptoms and spatial working memory in patients with schizophrenia – a randomized, double-blind, placebo-controlled trial

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23 **Abstract**

24 *Background:* Sodium nitroprusside (SNP) has been reported to rapidly reduce psychotic symptoms in
25 patients with schizophrenia. This has the potential to revolutionize treatment for schizophrenia. In
26 this study, we tested the hypothesis that SNP leads to a reduction in psychotic symptoms and an
27 improvement in spatial working memory performance in patients with schizophrenia. *Methods:* This
28 was a single-center, randomized, double-blind, placebo-controlled trial performed from 27 August
29 2014 to February 10 2016 (clinicaltrials.gov Identifier: NCT02176044). Twenty patients with
30 schizophrenia aged 18-60 with a diagnosis of schizophrenia or schizoaffective disorder were
31 recruited from psychiatric out-patient clinics in the South London and Maudsley NHS Trust, London,
32 UK. Baseline symptoms were measured using the Positive and Negative Syndrome Scale (PANSS) and
33 the 18 item Brief Psychiatric Rating Scale (BPRS), and spatial working memory was assessed using
34 the CANTAB computerized test. Participants received either an infusion of sodium nitroprusside (0.5
35 micrograms per kilogram per hour for 4 hours) or placebo and were re-assessed for symptoms and
36 spatial working memory performance immediately after the infusion, and 4 weeks later. *Results:*
37 Sodium nitroprusside did not lead to any reduction in psychotic symptoms or improvement in spatial
38 working memory performance compared to placebo. *Conclusions:* Although this study was negative,
39 it is possible that the beneficial effects of sodium nitroprusside may occur in patients with a shorter
40 history of illness, or with more acute exacerbation of symptoms.

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Background

Sodium nitroprusside (SNP) is a medication used in intensive care medicine as an antihypertensive. Its main mechanism of action is through the release of nitric oxide (NO) leading to vasodilation. A recent study by Hallak and colleagues reported that SNP may exert an antipsychotic effect in patients with schizophrenia (Hallak *et al* 2013). They reported that a 4 hour infusion of 0.5 mcg/Kg/min SNP led to a significant reduction in psychotic symptoms, as measured by the 18 item Brief Psychiatric Rating Scale (BPRS-18), and the negative subscale of the Positive and Negative Syndromes Scale (PANSS), which persisted for 4 weeks following the infusion.

The interest in SNP as a potential antipsychotic agent arose from studies in animals that showed that SNP could reverse the effect of the N-methyl-D-aspartate (NMDA) receptor antagonist phencyclidine, suggested to be a glutamatergic model of schizophrenia, and also that mice with reduced nitric oxide synthase did not show the normal response to phencyclidine administration (Bird *et al* 2001, Bujas-Bobanovic *et al* 2000). These findings suggested that the potential effect of SNP might occur via the NMDA receptor or downstream pathways. Interestingly, SNP has been reported to act directly on NMDA receptor function – although it is not clear whether this is related to its function as an NO donor, or through the action of Fe(II) ions (Hoyt *et al* 1992, Manzoni *et al* 1992, Oh and McCaslin 1995, Shuto *et al* 1997).

Cognitive impairment is a significant problem in schizophrenia, and, given the suggested therapeutic mechanism of SNP in patients with psychosis via NMDA dependent glutamatergic neurotransmission, we hypothesised that SNP may improve functioning in tasks dependent on this mechanism. We chose spatial working memory as a cognitive function that has been shown to be impaired in patients with schizophrenia (Piskulic *et al* 2007), and that is hypothesised to be related to NMDA receptor function via a NO-dependent mechanism (Wass *et al* 2006). We hypothesised that treatment

with a single dose of SNP would lead to a significant improvement in spatial working memory in patients with schizophrenia, and that this improvement would be accompanied by a similar improvement in psychotic symptoms, as previously reported (Hallak *et al* 2013).

In this study, we aimed to attempt to replicate the findings of Hallak and colleagues (Hallak *et al* 2013), and to investigate the effect of sodium nitroprusside on spatial working memory in patients with schizophrenia.

Methods

This was a double-blind placebo-controlled clinical trial. Ethical approval was obtained from the National Research Ethics Service (Dulwich Research Ethics committee). This study was carried out according to the principles of the Declaration of Helsinki (Amendment 2008), and all applicable regulatory requirements. As there are no reported studies on the effect of NMDA receptor modulation on spatial working memory in schizophrenia, the study was designed to provide information about likely effect size, in order to facilitate power calculations for future studies. The sample size was chosen based on a recent study of SNP in patients with schizophrenia (Hallak *et al* 2013). We planned to recruit up to 30 participants, with an interim analysis after the recruitment of 20. Criteria for discontinuing the study at the interim analysis stage were non-significant ($p > 0.2$) superiority of SNP over placebo for the 18 item Brief Psychiatric Rating Scale (BPRS-18) (Overall and Goram 1962), the Positive and Negative Syndrome Scale (PANSS) negative subscale (Kay *et al* 1987), and spatial working memory performance using the Cambridge Neuropsychological Test Automated Battery (CANTAB) (Sahakian *et al* 1988).

Patients with a diagnosis of schizophrenia or schizoaffective disorder were recruited from the South London and Maudsley NHS Trust. Potential participants were approached via their treating consultants based in first episode psychosis teams, continuing care community mental health teams.

Following both verbal and written explanation of the study, potential participants were invited to give written consent to the study. Following this, evaluation of their suitability according to the inclusion and exclusion criteria was made (Table 1).

The study took place at the Clinical Research Facility based at King's College Hospital. This is a clinical trials unit based in a general hospital setting, with full medical and nursing cover, as well as in-patient beds available. Participants attended on 3 occasions.

At the screening visit written informed consent was obtained by one of the study doctors. The participants then underwent psychiatric evaluation including assessment with PANSS and BPRS-18. Current antipsychotic medication, drug, alcohol and smoking history were obtained, and a physical examination was performed to establish the patients' eligibility for the trial. Routine bloods (FBC, U&Es, LFTs, TFTs and B12), urine testing (blood, protein, glucose as well as drug and, in females, pregnancy), height/weight, vital signs and ECG were completed. Participants then underwent spatial working memory assessment using the CANTAB spatial working memory task.

On their second visit to the Clinical Research Facility ("randomisation visit"), dipstick testing for illicit substances and pregnancy was done prior to randomisation and an intravenous cannula sited in the right arm. Baseline PANSS and BPRS-18 assessment was completed.

Randomisation of participants was via an independent web based service hosted at the King's Clinical Trials Unit (www.ctu.co.uk). Participants were randomised 1:1 to SNP or Placebo using the method of block randomisation, with randomly varying block sizes of 2 and 4. Researchers entered the participants detail to the randomisation and received an automatic blinded confirmation email that randomisation had occurred. Unblinded emails were automatically sent to the dispensing pharmacists, with details of treatment allocation. The pharmacy issued either SNP solution and

isotonic 5% glucose solution or isotonic 5% glucose solution only (i.e. placebo) in accordance with the randomisation email. In the case of a SNP treatment, an unblinded clinical research nurse diluted the SNP with isotonic glucose solution to achieve the required dose of SNP to be delivered over 4 hours. The prepared solution or placebo was concealed in an opaque encasing to protect the SNP from ultraviolet light and to ensure blinding of the study team. The infusion was run at a rate to achieve 0.5 mcg/kg/min (SNP) or a similar rate for placebo over the course of 4 hours. The unblinded clinical research nurse did not take part in any of the ratings.

During the infusion of SNP or placebo, blood pressure and heart rate measurements were taken every 5 minutes for the first 20 minutes of the infusion and then every 10 minutes until the end of the infusion. The Spatial Working Memory task, PANSS and BPRS-18 assessments were repeated after completing the infusion. Full medical and nursing support, including emergency crash team support were available on site at all times.

Participants attended for a third appointment 4 weeks after the randomisation visit at which time they repeated the CANTAB Spatial Working Memory task, and PANSS and BPRS-18 assessment were repeated.

The change in the CANTAB spatial working memory task, PANSS negative subscale and BPRS-18 ratings over time between participants in the SNP and placebo group was analysed using repeated measures ANOVA.

Results

40 participants were screened for suitability. 21 were randomised (19 were excluded as they did not fulfil the inclusion/exclusion criteria at the time of screening). 1 subject was excluded from analysis after randomisation because they did not meet inclusion criteria at the time of starting the infusion

(Figure 1). Participants were well matched for demographic details (Table 2). All patients were treated with a wide variety of different antipsychotic medications, with 1 patient on aripiprazole (SNP), 1 patient on aripiprazole and olanzapine (SNP), 3 patients on clozapine (2 placebo, 1 SNP), 1 patient on clozapine and aripiprazole (placebo), 1 patient on clozapine and amisulpride (placebo), 1 patient on clozapine and haloperidol (placebo). 1 patient on flupentixol and olanzapine (placebo), 1 patient on haloperidol (SNP), 3 patients on olanzapine (1 placebo and 2 SNP), 1 patients on paliperidone depot (SNP), 1 patient on paliperidone depot, olanzapine and sulpride (SNP), 1 patient on pipotiazine (placebo), 2 patients on quetiapine (1 placebo and 1 SNP), and 2 patients on risperidone depot (1 placebo and 1 SNP).

Sodium nitroprusside led to a significant reduction in systolic ($p < 0.05$) and diastolic blood pressure during the infusion ($p < 0.005$) and an increase in heart rate ($p < 0.05$; Table 3; Figure 2). During the study, There were 6 recorded adverse events – 2 in patients on SNP (muscular spasm in left upper arm in 1; mild suprapubic tenderness at time of infusion, and psychiatric admission approximately 4 weeks after the infusion judged unrelated to the study in 1), and 3 on placebo (asymptomatic but clinically relevant hypotension (86/36) in 1 requiring interruption of infusion between 121-227 minutes, with mild dizziness between 50-60 minutes after stopping infusion; drowsiness in 1; and psychiatric admission approximately 3 weeks after the infusion judged unrelated to the study in 1).

With repeated measures ANOVA for the 3 time points (baseline, post-infusion, follow-up), there was no significant group x time effect for BPRS-18 or for PANSS negative subscale ($p > 0.2$; Figure 3, Figure 4). There was also no significant group x time effect for any spatial working memory outcome measure (time, errors or strategy) ($p > 0.2$, Figure 5). Post-hoc analyses of other PANSS subscales did not reveal any significant group x time effect for either PANSS positive or general subscales ($p > 0.2$). Excluding the participant in whom we had to interrupt the placebo infusion from the analysis did not significantly affect the results (all outcome measures $p > 0.2$).

Discussion

In this study, we sought to investigate the effect of SNP infusion on psychotic symptoms measured with PANSS-N and BPRS-18. In addition, we investigated the effect of SNP on the CANTAB SWM task in patients with schizophrenia. In contrast with an earlier study (Hallak *et al* 2013), we did not find any effect of SNP over placebo on reducing symptoms measured using the BPRS-18, PANSS-N (or other PANSS subscales). Furthermore, we did not find any effect of SNP on improving performance on spatial working memory.

The most striking difference between the current study compared to the earlier clinical trial is that, although we found a reduction in symptoms on SNP, there was a similar improvement in those on placebo, whereas the earlier study did not find any placebo effect (Hallak *et al* 2013). Another difference between this study and the first clinical trial of SNP in schizophrenia (Hallak *et al* 2013), is that the patients in the present study were significantly older, with a longer history of illness. If SNP does act via glutamatergic mechanisms, as hypothesised, it may be predicted that it would work most effectively in patients earlier on in the illness (Stone 2011) where glutamatergic abnormalities are more prominent (Marsman *et al* 2013), and possibly by actively selecting those with a demonstrable glutamatergic abnormality (Egerton *et al* 2012). Against this hypothesis, one case study has suggested that SNP may be effective in more chronic patients who have not responded to clozapine (Maia-de-Oliveira *et al* 2014), but it should be noted that in that report, there was no placebo arm.

Another difference between this and the earlier clinical trial of SNP in schizophrenia is that our sample had less severe negative symptoms (Hallak *et al* 2013). The reason for this is not clear, as neither study had negative symptoms as a specific inclusion criteria, but it is possible that SNP may be more effective in populations with higher levels of negative symptoms.

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Although no other groups have studied spatial working memory performance, a recent study examined the effect of SNP on stroop colour word test, verbal fluency and n-back. Although not corrected for multiple comparisons, the SNP group had fewer n-back errors after treatment compared to placebo, and also performed better than the placebo group on one of the stroop colour word test cards (Maia-de-Oliveira *et al* 2015). Again, this may be due to differences in the length of illness of the patients studied, or in the sensitivities of different cognitive domains to the effects of SNP.

Regarding adverse events, SNP was relatively well tolerated, but we did see a significant effect of SNP on blood pressure and heart rate despite using the lowest infusion rate used clinically, a finding that had not previously been reported in this group (Hallak *et al* 2013). The reason why we found a more marked effect of SNP on blood pressure compared to the earlier study is not clear, but may be due to the fact that our sample comprised an older group of patients, who were on a wider variety of antipsychotic medication than had previously been studied. Despite this, the effect on blood pressure was mild and well tolerated, and the only patient in whom we stopped the infusion because of concerns about low blood pressure was in the placebo arm.

In summary, we did not find any evidence that SNP improved spatial working memory function or psychotic symptoms in patients with relatively chronic schizophrenia. Further work is required to clarify whether an effect is present in a younger cohort of patients, including those with demonstrable brain glutamate abnormalities.

224 Disclosures

225 SK has received grant support from Lundbeck and Roche and has served as a one-off consultant
226 and/or speaker for Otsuka, Sunovion and Takeda and served on the Scientific Advisory Boards for
227 Forum, Acadia, Lundbeck and Roche. JS has received honoraria for speaking appointments from
228 Janssen and Roche.

229

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235 Table 1: Inclusion/exclusion criteria

Inclusion Criteria	Exclusion Criteria
Capacity to give informed consent	Prior history of intolerance to sodium nitroprusside
18 to 60 years of age	Presence of a seizure disorder,
Patients with a diagnosis of schizophrenia or schizoaffective disorder according to DSMIV	Any change in psychotropic medication in previous 6 weeks
Currently experiencing an exacerbation of symptoms (a score > 20 for PANSS-Positive subscale)	Diagnosis of substance abuse
Currently taking antipsychotics	Pregnancy (as determined by urine test) or breastfeeding
	Relevant medical history including untreated hypothyroidism, hyponatraemia, ischaemic heart disease, impaired cerebral circulation, renal impairment, vitamin B12 deficiency or Leber's optic atrophy

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238 Table 2: Demographic details

	Placebo (n=10)	SNP (n=10)	P
Gender (M/F)	8/2	7/3	0.5
Age – Mean Years (SD)	40(10)	34(9)	0.20
Years of Education (SD)	12(3)	12(2)	0.76
Ethnicity (Black British, Black Other, White British, Asian, Mixed)	5,2,1,1,1	6,2,0,1,1	0.83
Diagnosis (Schizophrenia/ Schizoaffective)	9/1	9/1	1
Current Smoker (Y/N)	7/3	5/5	0.325
Cigarettes/day (SD)	10.8(12.7)	9(9.6)	0.73
Illness Duration – Mean Years (SD)	17(8)	12(7)	0.18
Current cannabis use (Y/N)	6/4	1/9	0.06

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241 Table 3: Change in blood pressure and heart rate during SNP and placebo infusions

	Placebo (n=10)	SNP (n=10)	P
Mean reduction in systolic BP during infusion – mean(SD) mmHg	2.3(7.6)	10.3(8.5)	0.04
Maximum reduction in systolic BP during infusion – mean(SD) mmHg	15.8(9.4)	34.7(29.25)	0.11
Mean reduction in diastolic BP during infusion – mean(SD) mmHg	1.2(6.5)	15.6(5.1)	<0.001
Maximum reduction in diastolic BP during infusion – mean(SD) mmHg	14.1(12.3)	29.5(8.0)	0.004
Mean increase in HR during infusion – mean(SD) beats per minute	2.0(7.6)	9.2(6.6)	0.04

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- 245 Figure 1 – Recruitment flow diagram.
- 246 Figure 2 – Change in systolic and diastolic blood pressure with time (minutes) following
247 commencement of placebo or sodium nitroprusside (SNP) infusion.
- 248 Figure 3 – PANSS positive, negative and general subscales, and BPRS-18 scores before and 4 hours
249 after placebo or sodium nitroprusside (SNP) infusion, and at 4 weeks follow-up.
- 250 Figure 4 – Individual patient data for BPRS-18 before and 4 hours after placebo or sodium
251 nitroprusside (SNP) infusion, and at 4 weeks follow-up.
- 252 Figure 5 – Mean errors and strategy score for CANTAB spatial working memory task at screening, 4
253 hours after placebo or sodium nitroprusside (SNP) infusion, and at 4 weeks follow-up.
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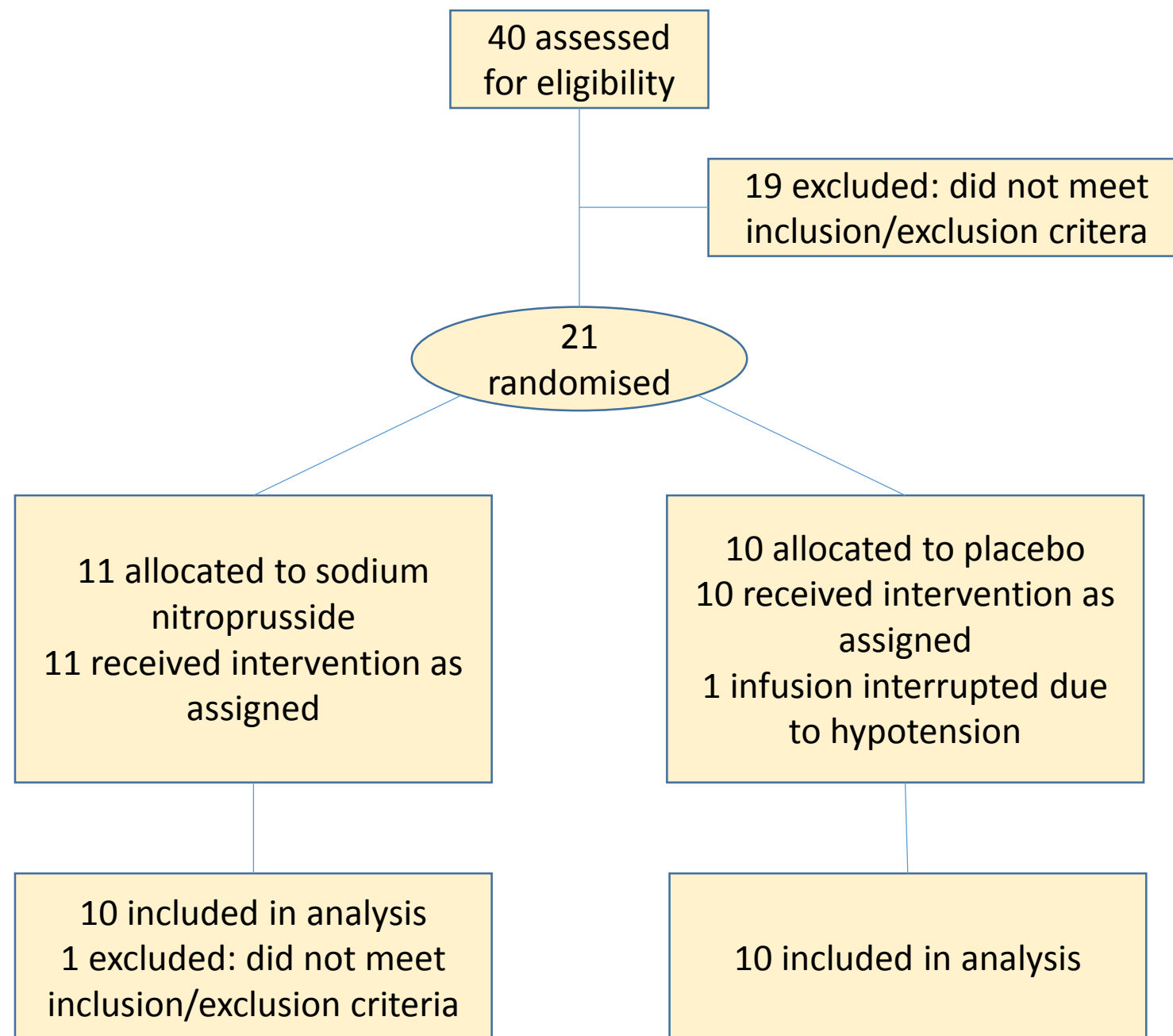


Figure 2

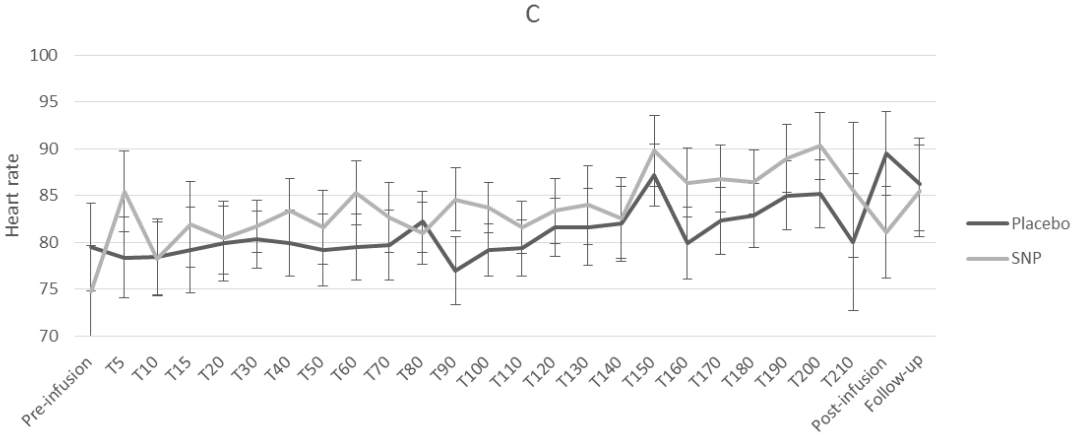
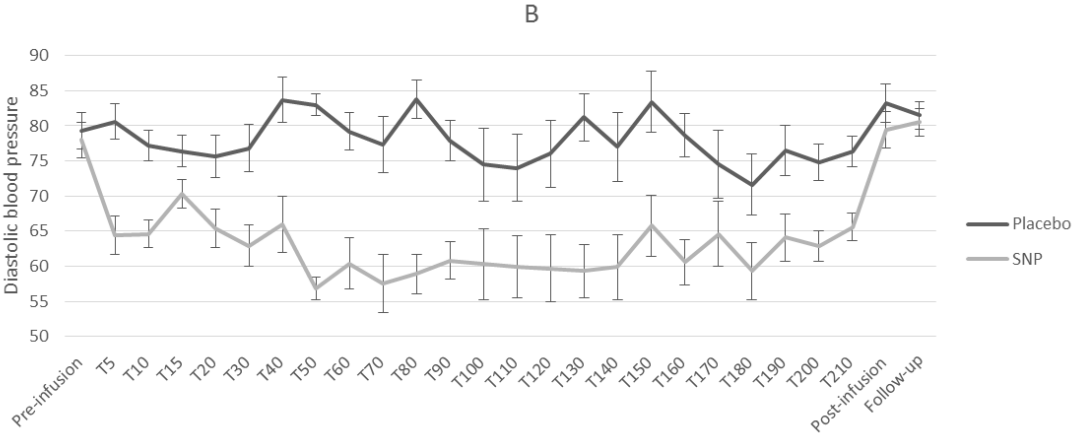
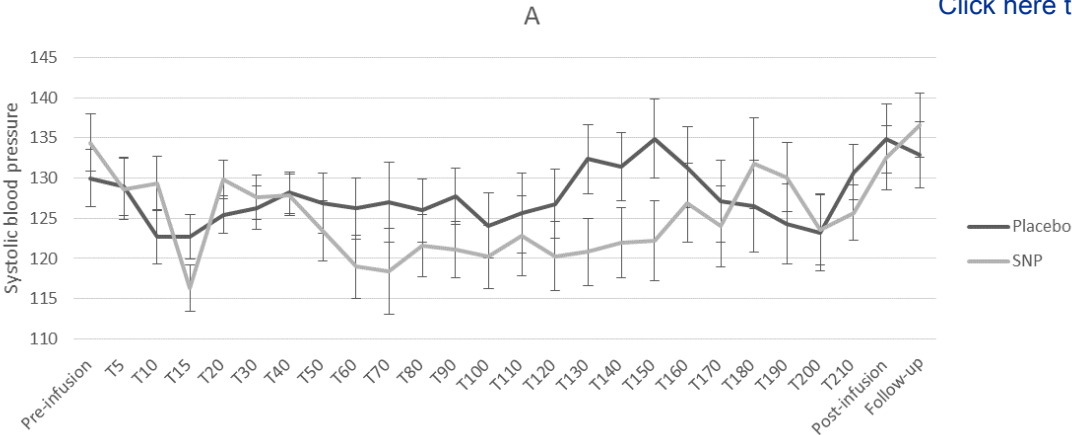
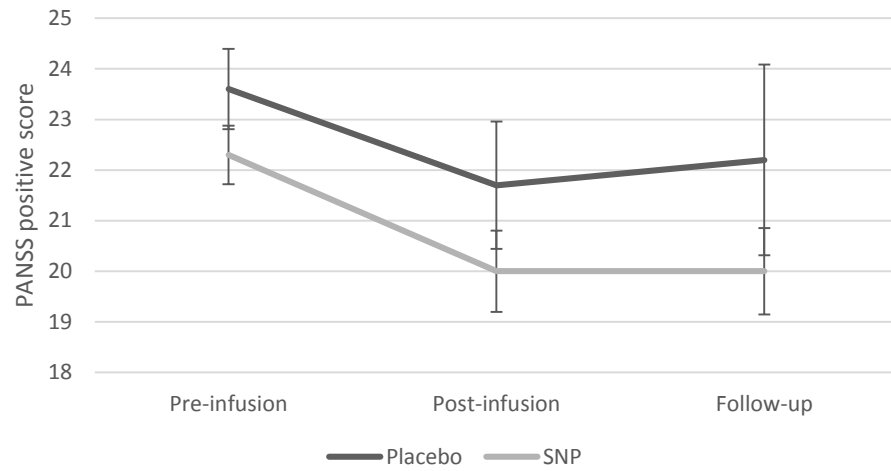


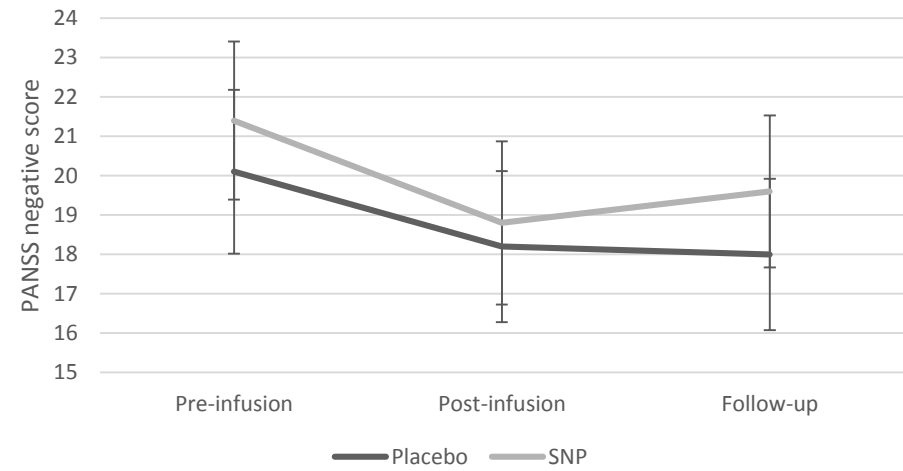
Figure 3

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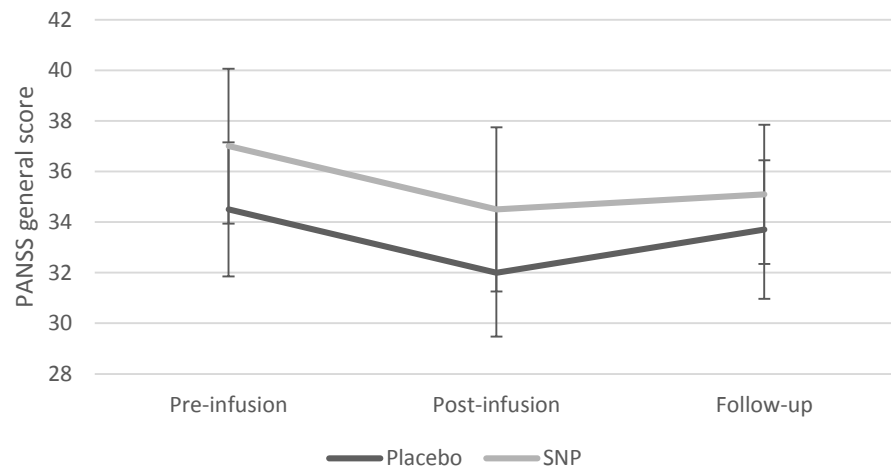
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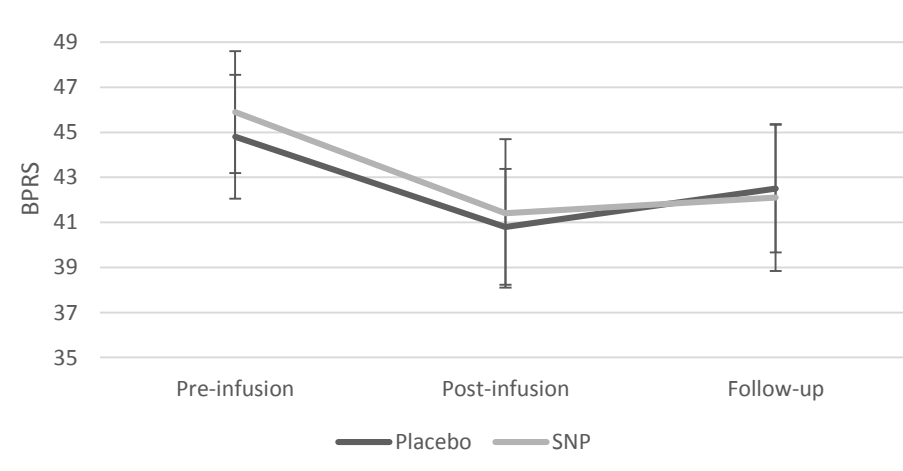


Figure 4

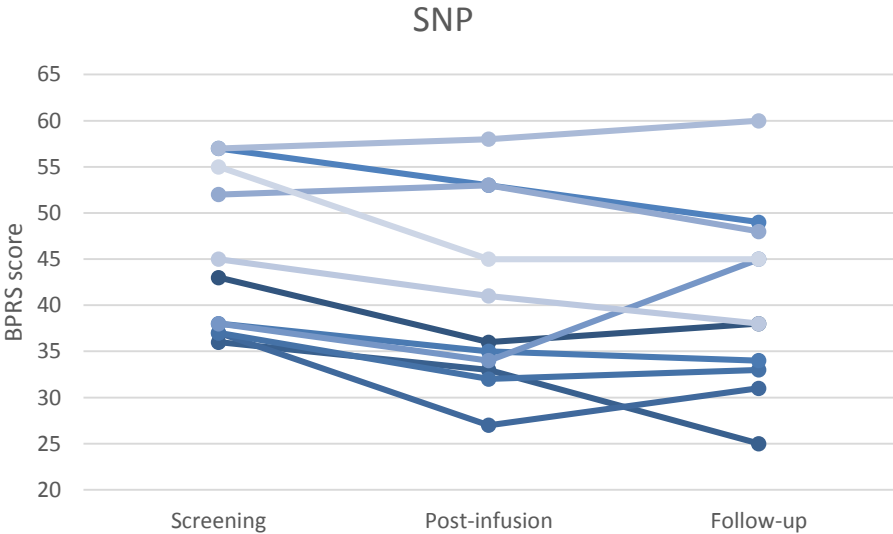
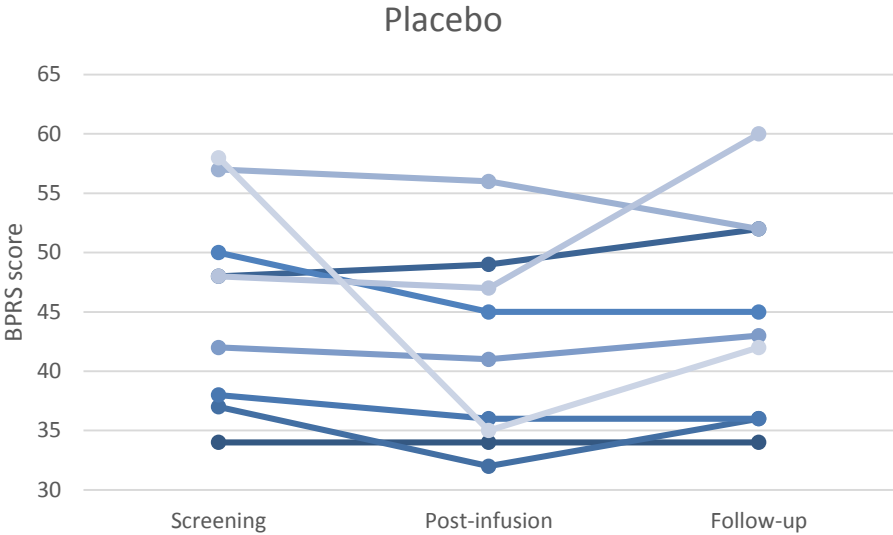


Figure 5

