UNIVERSITY OF AMSTERDAM

UvA-DARE (Digital Academic Repository)

Fighting against fear

Novel approaches to understanding, modifying, and manipulating maladaptive memories

Elsey, J.W.B.

Publication date
2020

Document Version
Other version

License
Other

Link to publication

Citation for published version (APA):
Elsey, J. W. B. (2020). *Fighting against fear: Novel approaches to understanding, modifying, and manipulating maladaptive memories*. [Thesis, fully internal, Universiteit van Amsterdam].

General rights
It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations
If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: https://uba.uva.nl/en/contact, or a letter to: Library of the University of Amsterdam, Secretariat, P.O. Box 19185, 1000 GD Amsterdam, The Netherlands. You will be contacted as soon as possible.

UvA-DARE is a service provided by the library of the University of Amsterdam (https://dare.uva.nl)

Download date:05 Mar 2026

Chapter 2.

Encoding or consolidation? The effects of pre- and post-learning propranolol on the impact of an emotional scene

This chapter is *in press* as:

Elsey, J.W.B, Bekker, T.A., De Bree, A.M., & Kindt, M. (2019). Encoding or consolidation? The effects of pre-and post-learning propranolol on the impact of an emotional scene. *Journal of Behavior Therapy and Experimental Psychiatry*.

Abstract

Background and Objectives. Researchers have conceived of post-traumatic stress disorder (PTSD) as a disorder of memory, and proposed that blocking the impact of stress-related noradrenaline release in the aftermath of trauma may be a way of preventing the 'over-consolidation' of trauma-related memories. Experimental research in humans has been limited by typically focusing on declarative memory for emotional stories, and has mainly given propranolol before learning. In contrast, the clinical studies that we comprehensively review are hampered by practical challenges, such as reliably administering propranolol in a time window sufficiently close to the traumatic event. In this study, we aimed to assess the impact of both pre- and post-learning propranolol on emotional and declarative memory for an emotional scene, using the 'trauma film paradigm'.

Methods. To control for drug and timing effects, participants received a pill (40mg propranolol or placebo) both 60 minutes before and within 5 minutes after viewing a 12 minute, emotionally arousing trauma film, and were assigned to one of the three conditions: propranolol-placebo (n = 25), placebo-propranolol (n = 25), or placebo-placebo (n = 25). We assessed participants' immediate emotional responses to the scene, as well as delayed impact (intrusions, Impact of Events Scale) and declarative memory.

Results. Using Bayesian informative hypothesis testing, we found that pre-learning propranolol reduced the initial emotional impact of the 'trauma film'. However, we did not find strong evidence for an impact of pre- or post-learning propranolol on later consequences of having watched the emotional film (intrusions, Impact of Events, or tests of declarative memory). Exploratorily restricting analyses to women, we did find evidence suggesting that *pre*-encoding propranolol could reduce the rate of intrusions and self-reported negative impact of the emotional scene one week later.

Limitations. Floor effects in the delayed impact of the emotional scene could preclude observing differences as a function of propranolol, and propranolol dosage may need to be increased.

Conclusions. An impact of propranolol on encoding could raise difficulties in interpretation when only pre-encoding propranolol is used to make inferences about consolidation. We discuss the challenges of elucidating the mechanistic underpinnings of propranolol's reported effects on memory.

Reducing the individual and societal costs of posttraumatic stress disorder (PTSD) is a major aim of translational research (Hoge et al., 2007; Kessler, 2000). Various means of treating PTSD have been developed in pursuit of this goal. However, an alternative approach is secondary prevention, in which one gears interventions toward preventing the development of illness rather than treatment (Kearns, Ressler, Zatzick, & Rothbaum, 2012). In PTSD, this would involve administering interventions in the aftermath of trauma, but before negative sequelae have arisen. While psychological prevention approaches have been considered (e.g., Hagenaars & Arntz; 2012; Holmes et al., 2009), pharmacological approaches may be another means of achieving this.

Stressful events elicit the release of several adrenal stress hormones/neurotransmitters, such as cortisol, adrenaline, and noradrenaline (de Quervain, Aerni, Schelling, & Roozendaal, 2009; McGaugh & Roozendaal, 2002). (Nor)adrenergic signaling can strongly affect memory in both humans and animals (Cahill & Alkire, 2003; Soeter & Kindt, 2011; 2012b). In light of the influence of (nor)adrenergic signaling on memory, PTSD has been conceptualized as a disorder of memory, in which the heightened release of hormones/neurotransmitters associated with trauma induces an 'over-consolidated' memory for the traumatic event (Pitman, 1989; Pitman & Delahanty, 2015). This strong memory can then manifest in symptoms of PTSD such as intrusions about the event and heightened emotional responsivity (Pitman, 1989, p.222; Pitman & Delahanty, 2015). Other researchers have expressed similar ideas, framing mental health problems such as PTSD and specific phobias as disorders of emotional memory (Kindt, 2014), in which heightened stress hormone signaling produces strong and recalcitrant maladaptive memories. While phenomenologically, such varied responses as intrusive imagery, excessive fear reactions, and defensive reflexes, may seem only tangentially related, they share common features such as being elicited automatically and even in spite of attempted suppression, being easily provoked by small reminders, and being resistant to extinction. These commonalities could point to a common etiology in stress hormone/neurotransmitter-induced 'emotional' or 'over-consolidated' memory. Researchers and clinicians have therefore investigated whether drugs that counter the impact of such neurotransmitters may prevent the development of PTSD (e.g., Pitman et al., 2002).

Several experimental studies highlight the prospect of such an approach, with propranolol typically administered 60 minutes before an emotionally arousing slideshow story. A meta-analysis of such studies indicated that propranolol could reduce the typical emotional enhancement of declarative memory (Lonergan, Olivera-Figueroa, Pitman, & Brunet, 2013). However, as propranolol was given 60-90 minutes before the learning experience in 11 of 12 studies analyzed, it would likely have been at functional levels during encoding. The one study giving propranolol immediately before encoding (van Stegeren, Everaerd, & Gooren, 2002), and a subsequent study assessing post-encoding propranolol administration (Thomas, Saumier, Pitman, Tremblay, & Brunet, 2017), failed to find any impact of propranolol on memory. If propranolol cannot interfere with memory consolidation when given after an emotional event, then preventing the development of PTSD through post-trauma propranolol administration may be unfeasible. In addition, in some cases pre-encoding propranolol could affect the subjective experience of the emotional event itself, as opposed to impacting upon consolidation. Though propranolol has shown mixed effects in treating anxiety disorders (Laverdure & Boulenger, 1991), it can have anxiolytic effects at doses of just 20-40mg (Elman et al., 1998; Khadke, Khadke, & Klare, 2012; Mealy et al., 1996), and is even used by musicians and other public performers to reduce performance anxiety (Slomka, 1996). Propranolol has further been found to reduce amygdala reactivity to emotional experimental stimuli (Hurlemann et al., 2010).

In clinical settings, studies assessing the secondary prevention of PTSD with betablockers range from assessing pediatric emergency department visitors (Nugent et al., 2010) to injured US military personnel (McGhee et al., 2009). Despite some promising results, such as reduced rates of PTSD symptomatology or lower physiological reactivity following post-trauma propranolol (e.g., Ahl et al., 2017; Brunet et al., 2002; Vaiva et al., 2003), other studies found little or no impact of betablockers on trauma sequelae (e.g., Meli et al., 2017; Rosenberg et al., 2018; Sharp et al., 2010). Heterogeneity of study designs, patient group, trauma type, failure to distinguish betablockers that do or do not cross the blood-brain-barrier, plus means and timing of administration, makes it difficult to draw strong conclusions regarding the potential efficacy of this approach.

Crucially, most studies have not achieved beta-adrenergic blockade very shortly after trauma, which is essential for blocking the initial process of

consolidation (Bourtchouladze et al., 1998; Schafe & LeDoux, 2000). It has been found that *de novo* protein synthesis is required in the hours shortly following learning in order for memory to be maintained over the long-term. This protein synthesis is thought to result in structural changes at synapses that support long-term memory (Bourtchouladze et al., 1998; Alberini & Kandel, 2015). Betablockers are argued to interfere with components of the molecular cascade that leads to synthesis of such plasticity-related proteins (Moncada, Ballerini, & Viola, 2015; O'Dell et al., 2015), with one study finding that adrenergic receptor activity during, but not after, emotional learning activates two plasticity-related targets, involved in acquisition/short-term memory (α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic [AMPA] receptors) and consolidation/long-term memory (extracellular regulated kinase) (Schiff et al., 2017). Studies in animal models of different types of emotional memory suggest the involvement of beta-adrenergic receptors in memory consolidation ranging from during encoding (e.g., Schiff et al., 2017) to around 2 hours after learning (e.g., Sara, Roullet, & Przybyslawski, 1999). Hence, it may be difficult or impossible for post-encoding betablocker ingestion to take effect within the window for disrupting at least the initial stages of memory consolidation, and clinical studies with delays of many hours between trauma and propranolol administration would almost certainly miss this window.

In summary, experimental and clinical intervention studies are inconclusive and sometimes uninformative as to the possibility of interfering with the negative effects of trauma using beta-adrenergic blockade. Emotional slideshow studies suggest that only *pre*-encoding propranolol affects emotional enhancement of declarative memory. However, those wishing to interfere with PTSD development are most interested in symptoms beyond declarative memory, such as distress and intrusions. Clinical studies suffer from a number of practical disadvantages, causing delays in drug delivery. Such practical issues mean that many clinical studies cannot determine the potential efficacy of a pharmacological approach.

The trauma film paradigm may more closely approximate trauma than emotional slideshows, while allowing for close control over drug administration. Contemporary trauma film studies typically involve exposing participants to emotionally arousing video clips, such as depictions of traffic accidents and surgeries, and assessing the impact of this 'experimental trauma' through post-event questionnaires, diaries of intrusions, self-reports of aversive effects, and tests of

declarative memory (James et al., 2016). In this study, we used the trauma film paradigm to provide participants with a negative emotional experience, assessing the effects of both pre- and post-trauma film propranolol on the immediate and longer-term impact of the experience. We assessed negative outcomes (intrusive memories and negative impact assessed by the Impact of Events Scale) as well as declarative memory. Comparing effects of pre- and post-learning propranolol in humans can help to understand how noradrenergic functioning affects memory encoding and consolidation, with implications for secondary prevention.

Three primary hypotheses regarding the effects of pre- and post-trauma propranolol can be derived from current findings/theory. H_0 reflects the *null hypothesis*, that neither means of administering propranolol will affect outcomes in the trauma film paradigm. Not all human experimental studies found an effect of pre- or post-learning propranolol on memory, several clinical studies failed to find any effect of betablockers, and some animal studies suggest that propranolol is ineffective in blocking the consolidation of a range of emotional memories (Dębiec & LeDoux, 2004; Villain et al., 2016). $H_{encoding/rapid\_con}$ reflects the *encoding* or *rapid consolidation* hypotheses, which predict that only propranolol administered before encoding should affect the impact of the trauma film. Propranolol may affect the initial encoding of the event, reducing how stressful or aversive the experience is, and diminishing its later impact. Alternatively or simultaneously, pre-learning propranolol may interfere with a rapid post-encoding consolidation process, which post-learning oral propranolol could not affect due to taking 60-90 minutes to reach peak bioavailability (Goodman, 1996). One way of determining whether propranolol likely exerts an influence through affecting encoding or a rapid consolidation process is by assessing the initial impact of the trauma film: if propranolol affects immediate responses to the trauma film, and especially if these initial responses also predict later outcomes, an encoding account may be more parsimonious than a rapid consolidation one.

Table 1. Clinical studies assessing the impact of propranolol and other betablockers on trauma outcomes

Authors	Trauma type/sample	Participants	Time of drug administration	Betablocker type and dose	Outcome
Pitman et al., 2002	Emergency department visitors with a DSM-IV PTSD criterion event, mostly motor vehicle accidents	18 young adult patients (44% men) receiving propranolol and 23 (51% men) receiving placebo	<6 hours post-trauma	Propranolol, 4x 40mg each day for 10 days	Categorical PTSD diagnoses were not significantly different between groups at 1- or 3-month follow-ups, and CAPS scores only suggested a possible trend in favor of propranolol at 1 month. Psychophysiological assessment suggested lower reactivity to trauma scripts in the propranolol group.
Vaiva et al., 2003	Emergency department visitors with a DSM-IV PTSD criterion event, mostly motor vehicle accidents	11 young adult patients receiving propranolol (64% men) and 8 completing study measures but not receiving any intervention (50% men)	2-20 hours post-trauma, mean 9.5 hours	Propranolol, 3x 40mg for 7 days + an 8-12 day taper	PTSD diagnosis and symptoms were lower among those receiving post-trauma propranolol than those who refused treatment, assessed 2 months post-trauma.
Stein et al., 2007	Emergency department visitors, mostly having experienced motor vehicle accidents	17 young adult patients receiving propranolol, 17 receiving placebo, + 14 receiving gabapentin. 54% participants were men.	Up to 48 hours post-trauma	Propranolol, beginning with 20mg 3x daily, uptitrating the dose for 2 days up to 40mg, full dosage for 8 days, then 4 days tapering down.	Propranolol, placebo, and gabapentin groups were not significantly different in PTSD symptoms, depressive symptoms, acute stress symptoms, and overall PTSD diagnosis up to 8 months follow-up.
McGhee et al., 2009	Burn injuries in US military personnel	31 patients who had been administered propranolol as part of treatment vs. 34 who had not.	Timing was not assessed	Propranolol. Dose was not assessed	Rates of PTSD in the propranolol and placebo groups were not significantly different, nor were levels of PTSD symptoms. Propranolol patients did have higher injury severity.

Krause-neck et al., 2010	Coronary artery bypass grafting or cardiac valve replacement in adult cardiac patients	84 patients receiving metropolol (18 women, 66 men) vs. 44 who did not (15 women, 29 men).	Any time post-operatively	Metropolol, starting with 23.75mg and titrated upwards to up to 100mg per day as tolerated. Adrenaline may also have been administered during operations.	Women receiving metropolol showed fewer traumatic memories and less PTSD symptoms at 6 months than those who did not, whereas in men there was no such effect. In men, adrenaline administration was positively correlated with traumatic memories.
Nugent et al., 2010	Children/teens treated at a pediatric emergency department, mostly for motor vehicle accidents	14 patients who received propranolol (57% men) and 15 who received placebo (47% men)	<12 hours from admission	Propranolol, 20mg twice daily for 10 days with a 5-day taper	There were no significant differences in full or partial PTSD between groups, with some exploratory analyses suggesting a possible reduction in symptoms in boys and a possible increase in symptoms in girls.
Sharp et al., 2010	Children receiving acute care for severe burns.	126 patients who received propranolol (71% men) and 237 who did not (60% men)	On average, 2 days post-burn	Propranolol, 1mg/kg every 6 hours, increasing to average 4mg/kg/day within the first 10 days, continued for 4 weeks post-injury.	There were no significant differences in rates of acute stress disorder in propranolol vs. control groups.
Hoge et al., 2012	Emergency department visitors with a DSM-IV PTSD criterion event, mostly motor vehicle accidents	20 patients who received propranolol (35% men) and 21 patients who received placebo (52% men).	On average, 4 hours post-trauma	Propranolol, 40mg short-acting, followed 1 hour later by 60mg long-acting. Long-acting propranolol continued at home with 2x 120mg/day for 10 days, followed by a 9-day taper	There were no significant differences between propranolol and placebo participants in either PTSD symptoms or psychophysiological reactivity at follow-up assessments 4-5 weeks later. In high-adherence participants, there was some lower physiological reactivity in the propranolol group.
Tarsitani et al., 2012	Adult cardiac patients who underwent coronary artery bypass grafting or valve surgery	71 (65% men) patients who received cardiac surgery and completed follow-up questionnaires.	Preoperatively, perioperatively, postoperatively, and at hospital discharge. Other drugs/hormones, including adrenaline,	Carvedilol, atenolol (hydrophilic), and metropolol at 'standard' dosages	Women who were treated with perioperative betablockers had lower PTSD symptoms and fewer traumatic memories at 6-month follow-up. Betablockers were not related to PTSD symptoms or memories in men.

			may have been used in treatment.		
Lindgren et al., 2013	Adult women diagnosed with breast or colorectal cancer	210 women who were (n=39) or were not (n=171) using beta-blockers.	Continual betablocker use for hypertension	Predominantly water-soluble/hydrophilic betablockers characterised the betablocker group (32 of 39)	Use of betablockers was associated with fewer intrusive memories, accounting for 42.5% of the variance.
Bhuvanes-war et al., 2014	Implantable intracardiac defibrillator (ICD) discharges in male cardiac outpatients	18 men using lipophilic (n=12) and hydrophilic (n=6) betablockers	Continual betablocker use for hypertension	Propranolol, metoprolol, and carvedilol (all lipophilic), and atenolol and sotalol (hydrophilic).	There was a trend towards lower PTSD symptoms in the patients taking lipophilic betablockers at the time of the ICD discharge
Ahl et al., 2017	Trauma center visitors with severe traumatic brain injury	80 patients using betablockers (71% men) and 80 patients not using betablockers (71% men)	Betablocker use had begun prior to the traumatic event and was continued within a median 48 hours after admission.	Majority metropolol (66%), but unspecified.	Patients who had been taking betablockers had a lower incidence of post-traumatic depression in the year following discharge.
Meli et al., 2017	Acute coronary syndrome patients	50 patients who were administered betablockers (62% men) or not (52.3% men)	Timing was not assessed	Betablocker type was not recorded	Controlling for confounding factors, receipt of betablockers during the emergency department visit was associated with lower PTSD symptom severity, but accounted for very little variance.
Rosenberg et al., 2018	Children receiving acute care for severe burns.	89 patients receiving propranolol (70% men) and 113 not receiving propranolol (65% men)	Within 48-72 hours of admission, but admission averaged 5 days post-burn	Propranolol, averaging 3.64mg/kg/day for 7 or more days during acute care. A range of other drugs and psychotherapeutic care may also have been provided.	Rates of PTSD or PTSD symptomatology in children treated with propranolol were not significantly different to those not given propranolol

If both pre- and post-encoding propranolol reduce the later impact of the film, this would be strong evidence for the *consolidation hypothesis* (H_{consol}), and suggest that post-encoding consolidation processes occur within a time frame that renders a consolidating memory vulnerable to disruption by propranolol given before or immediately after learning. See the 'Statistical Approach' section for further information on how these hypotheses were tested using the R Package (R Core Team, 2013) '*Bayesian informative hypothesis testing'* (*BAIN:* Gu, Hoijtink, Mulder, & van Lissa, 2019: https://CRAN.R-project.org/package=bain; Gu, Mulder, & Hoijtink, 2018; Hoijtink, Mulder, van Lissa, & Gu, 2019 *in press*).

Methods

Participants

The project was approved by the institutional review board under the code 2017-CP-7578. All participants gave informed consent and were recruited through the University's online lab system. Participants received 4 credits or €40 for participation. Inclusion criteria were: age 18-35, not taking psychoactive medications, not suffering from physical or psychological illnesses, and absence of contraindications for taking propranolol (exhaustive list in Appendix 1). We reminded participants that they could decline to participate after informing them of the nature of the trauma film. The total sample of participants (N = 75) was aged between 18-29 (M = 21.64, SD = 2.99) and mostly women (n = 53). Two further participants took part in session 1 in the placebo-placebo group, but did not return for session two. These were excluded from all analyses, and we do not have a record of their computer responses to determine if, for example, they were particularly upset by the trauma film. Participants at the university are not required to give a reason for dropping out, and may simply have terminated their participation after earning the bulk of their compensation for session one, which is longer than session two.

Materials and Measures

Propranolol. To control for both the effect of drug and timing of administration, all participants received two pills, one 60 minutes before viewing the trauma film, and another within 5 minutes of viewing it. Participants were randomly split into 3 groups (25 per group), to receive either placebo both times (placebo-placebo), 40mg propranolol and then placebo (prop-placebo), or placebo then propranolol (placebo-prop). Pills were placed in sealed envelopes by the first author and given to the two experimenters, who were blind to their contents. Propranolol

pills were made by Accord Healthcare Ltd. (UK), and provided along with placebo pills by Huygens Apothecary (NL).

Trauma Film. The trauma film was 12m7s long, comprising scenes depicting traumatic and graphic events, such as fictional footage of road traffic accidents and real footage of surgeries. These scenes functioned as an experimental analogue of trauma, and have been used to induce negative affect and provoke intrusions in multiple studies (e.g., Holmes et al., 2009; James et al., 2015). The film was presented in Presentation on a 51x29cm computer screen, with participants positioned in a chin rest 60cm from the screen. Participants were instructed to pay close attention to the film, and to watch it as if they were a bystander observing the events really happening.

Intrusion Diary. Participants received a Dutch version of Holmes and colleagues' (2009) intrusion diary, which includes space for participants to record intrusions each day, starting from the waiting period after having watched the trauma film up to handing in the diary at the 1-week follow-up session. When given the diary, participants were provided with a definition of intrusions as spontaneously arising images from the film, and that they should not include memories that they had purposely recalled. They were also told that intrusions should involve some kind of image-based content, and not be purely verbal thoughts. Participants were requested to keep the diary with them in order to record intrusions as soon as they occurred, and to set aside time daily to ensure the diary was completed.

Visual Analogue (VAS) and Likert Scales. VASs were used to measure negative mood induced by the film. Shortly before and after watching the trauma film, participants reported how much they were experiencing 6 negative mood states (from 0 = *not at all* to 100 = *extremely*). We operationalized 'emotional change' as the average change across mood states. After watching the film, participants also reported how distressing they found it (from 0 = *not at all* to 100 = *extremely*) and how much attention they had paid (from 0 = *none at all* to 100 = *total attention*).

At the second session, participants reported on a Likert scale how accurately they had kept their diaries (from 0 = *very inaccurately* to 10 = *very accurately*). To determine whether participants might need further debriefing/care, they were also asked how unpleasant they had found recalling items from the previous week and how difficult they found it to answer the questions. No participants required more than a standard debriefing.

Declarative memory tests. Tests of verbal and visual declarative memory were based upon James et al. (2015). Some modifications/additions to the tests were made to give an equal number of items per scene. Participants were given a brief description of the scene referred to, and then presented with 4 true/false statements about that scene. After each set of 4 questions for a scene, participants reported their confidence using a VAS (from 0, *not at all confident* to 100 = *completely confident*). These scores were averaged to provide an overall confidence score for verbal memory. For visual declarative memory, we included 11 correct images from the 11 movie scenes, and 11 foils depicting similar events. Participants were presented with a real or foil image and asked to indicate whether it was an image from the film. After the 22 items, participants indicated their confidence on a VAS as in the verbal test. Raw scores could range from 0-44 and 0-22 for verbal and visual memory, respectively.

Modified Impact of Events Scale – Revised (IES-R, Weiss & Marmar, 1996). The IES-R is a 22-item questionnaire that assesses the experience of PTSD-related symptoms – hyperarousal, intrusions, and avoidance – over the previous week as a result of a traumatic experience. Participants indicate the degree to which they have experienced various symptoms (from 0 = *not at all* to 4 = *extremely*), yielding a total score from 0-88. Following Holmes et al. (2009), we used a modified version of the scale in which scale items reference the film, rather than a real traumatic event. The IES-R has shown excellent internal consistency and good convergent validity in clinical and community samples (Creamer, Bell, & Failla, 2003).

Individual Differences Questionnaires. Several official translations of well-validated psychological tests were used to determine any baseline group differences in depression (Beck Depression Inventory II, BDI: Beck, Steer, & Brown, 1996; van der Does, 2002), anxiety (Spielberger State-Trait Anxiety Inventory, STAI: Spielberger, Gorsuch, Lushene, Vagg, & Jacobs, 1983; van der Ploeg, 1980), anxiety sensitivity (Anxiety Sensitivty Index, ASI: Peterson & Reiss, 1992; Peterson & Heilbronner, 1987; Vujanovic, Arrindell, Bernsetin, Norton, & Zvolensky, 2007), and exposure to traumatic events (Life Events Checklist, LEC: developed by the National Center for PTSD, see Gray, Litz, Hsu, Lombardo, 2004).

Procedure

Session 1. Participants were screened to assess any contraindications in their medical history for taking propranolol, and underwent heart rate and blood pressure

checks using a sphygmomanometer (Omron). Participants then received their first pill (double blind). A 60-minute filler period was then started to allow pre-film propranolol to reach bioactive levels (Goodman, 1996), during which participants completed baseline questionnaires (BDI-II, STAI-T, STAI-S, LEC, ASI). For the remaining time, participants listened to *BBC Inside Science* (https://www.bbc.co.uk/programmes/b036f7w2). This was chosen as a relatively engaging but neutral means of passing the time. *Inside Science* episodes last 30 minutes, and involve discussions about interesting scientific topics to engage a non-specialist audience. Other studies have used tasks such as music judgment. However, some research suggests that music may interfere with the emotional enhancement of memory (Rickard, Wong, & Velik, 2012), and music can induce subjective and physiological states (Etzel, Johnsen, Dickerson, Tranel, & Adolphs, 2006), which may affect memory encoding.

Sixty minutes after their first pill, participants were taken to a computer cubicle, informed how to watch the movie, and to place their head in the headrest. The experimenter started the experimental program, turned off the lights, and left the cubicle. Participants completed the pre-film VASs, watched the film, and filled in the post-film VASs, before leaving the cubicle. The experimenter then gave the participant their second pill within 5 minutes of the participants finishing the film, and another 60-minute waiting period began. At the start of the waiting period, the experimenter fully explained the intrusion diary to participants and answered any questions before giving them the diary. Participants then listened to more *Inside Science* in the remaining time. After the 60-minute wait period, participants had a final blood pressure check and finished the session. Though exact timing between blood pressure/HR assessments was not measured, general timing of the protocol means that assessments were made 140-155 minutes after pill 1, and 60-75 minutes after intake of pill 2. To avoid unblinding experimenters to possible pill allocation, but to better track HR change over time, a continual and non-invasive ambulatory HR measure may be preferable in future studies.

Session 2. One week after their first session, participants returned to the lab, handed in their diaries and rated their accuracy, then filled in the STAI-S and IES-R. They were then led to a computer cubicle and underwent the verbal and visual declarative memory tests. Finally, participants indicated how much trouble they had

with the day's tasks, reported what pills they thought they had received, and were thanked and debriefed.

Statistical Approach. We used the R Package (R Core Team, 2013) '*Bayesian informative hypothesis testing'* (*BAIN:* Gu, Hoijtink, Mulder, & van Lissa, 2019: https://CRAN.R-project.org/package=bain; Gu, Mulder, & Hoijtink, 2018; Hoijtink, Mulder, van Lissa, & Gu, 2019 *in press*) to assess the relative evidence for each of our three hypotheses in the main outcome data.[1] With typical frequentist null hypothesis significance testing, one would perform an ANOVA on the outcome variables. If the ANOVA indicated a difference among groups, post-hoc tests would be conducted to try to infer the pattern of these differences. Using BAIN, one instead specifies what patterns of group means would be most consistent with each competing hypothesis (e.g., for H_{consol}, intrusions in the pre- and post-learning propranolol conditions should be approximately equal to one another, but less than in the placebo-placebo condition). The primary metric of interest that BAIN provides is an 'approximate adjusted fractional Bayes factor' (henceforth simply Bayes factor), quantifying the relative support in the data for competing hypotheses. Combining information about the prior probability of each hypothesis and the evidence provided by the data collected, BAIN computes the posterior model probabilities (PMPs) of the specified hypotheses. The Bayes factor favoring one hypothesis over another reflects the ratio of their respective PMPs. If the PMP for H_{consol} is .80, and the PMP for H_0 is .20, the Bayes factor for H_{consol} vs. H_0 is 4, meaning that the support in the data is 4 times greater for H_{consol} than for H_0. When alternative hypotheses are not favored, BAIN allows for the quantification of evidence for H_0, rather than only indicating that there is insufficient reason to reject it as in null hypothesis significance testing. This Bayesian approach is therefore particularly informative when aiming to assess evidence for more than one alternative hypothesis as well as the possibility of null effects.

[1] Full mathematical explication of how BAIN determines prior and posterior distributions is beyond the scope of this chapter, but is presented in Gu et al. (2018) and Hoijtink, Gu, and Mulder (2018). Priors are not user specified but are constructed using a fraction of information in the data (Gu et al., 2018; Hoijtink et al., 2018 provide a non-technical elaboration). As we demonstrate below, it is possible to determine how robust the results are to prior specification by conducting a sensitivity analysis, in which varying fractions of information are used to specify the prior. Parameters used in computation of the Bayes factor are the estimates of the group means, the sample sizes per group, and covariance matrix of the estimates of the means in each group (presented in Appendix 1). Finally, the algorithm and sampling method used to compute the Bayes factor is described in section 5 of Gu, Hoijtink, Mulder, and Rosseel (2018)

Based on previous research and theory, we identified three plausible hypotheses to include in our analyses: H_0, $H_{encoding/rapid\_con}$, and H_{consol}, which are described at the end of the introduction, and again briefly in the results. These hypotheses do not exhaust all the possible observable differences between groups that could occur. For example, post-encoding propranolol could be the most effective at reducing later intrusions. However, such proposals do not receive support from existing literature, and parsimony in hypothesis selection is ideal when using informative hypotheses. Should an unexpected pattern of results arise in the data, however, inclusion of a 'failsafe/unconstrained' hypothesis – included by default in BAIN – allows the investigator to determine if none of their pre-specified hypotheses are really supported by the data.

Sample size estimation was based upon power calculations reported by James et al. (2015), and derived from a cognitive intervention for intrusive memory consolidation in Holmes et al. (2009). As James and colleagues were investigating *re*consolidation, they reduced the presumed effect size of the Holmes et al. (2009) consolidation study, which had 20 participants per group, to arrive at a more conservative sample size of 26 per group in order to achieve 80% power at a .05 alpha level. We maintained a relatively conservative sample size of 25 per group. As these are frequentist calculations and relate to a non-pharmacological intervention, they are an imperfect estimate for our experiment. However, if an effect of propranolol cannot be observed under similar conditions to those of Holmes et al. (2009), then this may be an argument in favour of a non-pharmacological approach, which would have less contraindications and logistical issues than the use of propranolol.

To constrain the likelihood of false positives through conducting multiple simple Bayesian ANOVAs on our outcome variables, where possible we performed MANOVAs on outcome variables. When different variables are expected to be influenced similarly by an experimental manipulation, conducting a MANOVA on those variables can increase the likelihood of detecting a legitimate effect while reducing the number of statistical tests that are conducted. Hence, for the effect of propranolol on the initial impact of the emotional scenes, 'Emotional Change' and 'Distress' were entered into a MANOVA. Though 'Intrusive Memories' and 'IES' scores each measure in some sense the longer-term emotional impact of the scenes, intrusions are a count variable and were thus assessed in a separate ANOVA specifying them as Poisson distributed, separately from IES scores. Finally, both

visual and verbal declarative memory have been found to correlate with one another in previous studies, but not with measures indicative of 'emotional memory', and we expect that they ought to be influenced similarly to one another by propranolol, if at all. Verbal and visual declarative memory were thus assessed my means of a MANOVA.

Results

Baseline group comparisons. Table 2 depicts baseline measures of age, gender, and individual difference questionnaires in each group. One participant in the placebo-prop group who scored on the border of moderate-to-severe depression on the BDI and was a statistical outlier (>3 *SD* above the mean) was excluded from all analyses. There was no evidence for group differences in the baseline variables assessed (all $p > .2$). Bayesian ANOVAs conducted in *JASP* (*JASP* Team, 2018) using the default analysis settings (i.e., priors based on a Cauchy distribution: Wagenmakers et al., 2018) also favored the null, relative to an alternative hypothesis of any group differences, in age, STAI, BDI, and ASI, indicating a lack of confounding group differences (all $BF_{01} > 3.5$).

Overall, significant changes in heart rate and blood pressure from the start to end of the first session were observed across groups (Main effect of time HR, $F(1,71) = 251.39$, $p = <.001$, $\eta^2 = .767$; BP_{Sys}, $F(1,71) = 120.65$, $p = <.001$, $\eta^2 = .622$; BP_{Dia}, $F(1,70) = 6.091$, $p = .016$, $\eta^2 = .080$). Changes were descriptively larger for the two propranolol groups than the placebo-placebo group, but only heart rate showed a marginally significant group*time interaction ($F(2,71) = 2.74$, $p = .071$, $\eta^2 = .017$). The long rest periods could have caused decreases across all groups, obscuring a propranolol effect.

Forty-nine participants reported being unable to guess what pills they received. Those who guessed did not perform significantly above chance level at guessing their condition ($\chi^2(4) = 2.68$, $p = .613$), or whether they had received propranolol ($\chi^2(1) = 2.72$, $p = .099$).

Emotional induction by the trauma film. Two participants had extreme outlying values in emotional change in the prop-placebo group (>3 SD above the mean, Appendix 1, Figure S1), and were removed from further analyses. The included participants reported excellent attention to the film (M = 94.29, SD = 6.91, Med =

98), with no evidence for differences between groups (Kruskal-Wallis $\chi^2 = 1.311$, $p = .519$).

We used BAIN to assess evidence for $H_{encoding}$ (for emotional change and distress, prop-placebo < placebo-prop = placebo-placebo) versus H_0 (prop-placebo = placebo-prop = placebo-placebo) in a MANOVA with emotional change and distress ratings after the movie as outcome variables.

Table 2. Baseline comparisons between groups.

	Prop-Placebo	Placebo-Prop	Placebo-Placebo			
	M (SD)	*M (SD)*	*M (SD)*	*F*	*p*	BF_{01}
Age	21.52 (2.69)	21.08 (2.26)	22.20 (3.81)	.86	.426	4.48
STAI-S[a]	31.79 (6.29)	31.71 (6.79)	33.52 (7.20)	.56	.573	5.57
STAI-T[b]	34.20 (7.75)	36.50 (9.28)	35.90 (8.53)	.46	.635	5.98
BDI[c]	6.00 (5.34)	4.87 (5.15)	5.68 (5.79)	.27	.763	6.96
ASI[d]	10.17 (4.87)	7.95 (4.85)	9.64 (6.22)	1.05	.356	3.81
HR Δ	-21.68 (11.42)	-16.58 (10.43)	-15.68 (6.84)	2.74	.071	1.07
BP_{Sys} Δ	-14.12 (9.13)	-13.21 (10.62)	-10.12 (9.55)	1.15	.32	3.59
BP_{Dia} Δ[f]	-4.96 (13.31)	-2.21 (7.21)	-2.16 (8.45)	.12	.88	5.33
	Med (range)	*Med (range)*	*Med (range)*	*Kruskal-Wallis*	*p*	
LEC - direct[e]	0 (0-4)	0 (0-2)	0 (0-5)	1.51	.470	
LEC - total[e]	2 (0-8)	2 (0-13)	3 (0-13)	.68	.711	
	M:F	M:F	M:F	Chi^2	*p*	
Gender	5:20	10:14	7:18	2.81	.246	

BF_{01} = Bayes factor for the null hypothesis; M (SD) = mean and (standard deviation); Med = Median; M:W = men:women distribution; STAI-S & T = State Trait Anxiety Inventory -State and -Trait subscales; BDI = Beck Depression Inventory 2nd Edition; ASI - Anxiety Sensitivity Index; LEC = Life Events Checklist; HR Δ = Change in heart rate over session 1; BP_{Sys} Δ = Change is systolic blood pressure over session 1; BP_{Dia} Δ = Change in diastolic blood pressure over session 1; a = 1 STAI-S score was incomplete in the prop-placebo group; b = placebo-prop and placebo-placebo groups each had 1 incomplete STAI-T score; c = placebo-prop group had 1 incomplete BDI score; d = prop-placebo group had 1 and placebo-prop group had 2 incomplete ASI scores; e = placebo-prop group had 1 incomplete LEC score, f = one participant with an extreme outlying BP value was removed from the Prop-Placebo group

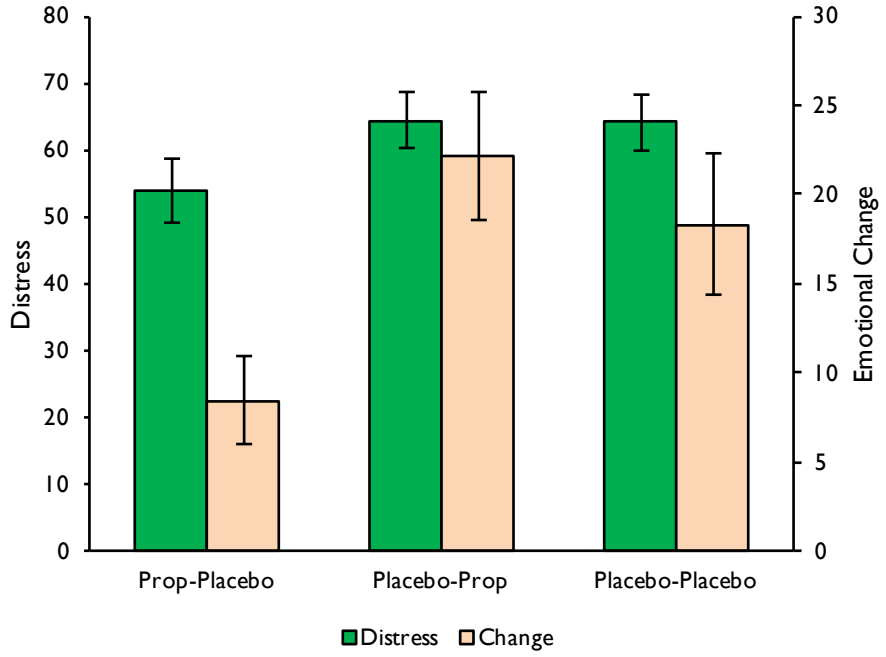

Figure 1. Mean distress and emotional change in each group. Error bars = +/- 1 standard error.

Mean emotional change and distress scores were lower in the Prop-Placebo group (n = 23) relative to the Placebo-Prop (n = 24) and Placebo-Placebo (n = 25) groups, which scored similarly to each other, consistent with $H_{encoding}$ (Figure 1). The Bayes factor for $H_{encoding}$ vs. H_0 was 10.465, indicating that the data provided over 10 times more evidence for $H_{encoding}$ than H_0 (Table 3a, Figure 2a). Appendix 1 provides sample sizes and prior and posterior covariance matrices per group used by BAIN for each analysis. As change scores were used for the measurement of emotional change, Appendix 1 also reports the results of a frequentist repeated measures ANOVA, indicating that baseline differences in emotion between groups do not explain the reduced emotional change in propranolol participants relative to those initially receiving placebo.

Bayes factors involving a null hypothesis can be sensitive to the selected prior variance used in their computation. Hoijtink and colleagues (2019) suggest additionally assessing Bayes factors at one half and one third of the default prior variance to confirm whether analyses are sensitive to reasonable changes to prior variance. We conducted a sensitivity analysis by multiplying the default prior variance by fractions ranging from 10/10 (or 1: the default prior variance) to 3/10, and

re-computing the Bayes factors and posterior probabilities (see Hoijtink et al., 2019). Figure 2a shows the posterior probabilities associated with $H_{encoding}$ and H_0 across these fractions, corresponding to increasingly small prior variance. $H_{encoding}$ was consistently favored across reasonable variations in the prior variance.

Table 3. Bayes factors for each of the hypotheses compared in each Bayesian analysis.

3a. MANOVA for Emotional Change and Distress

Hypotheses compared	BF
$H_{encoding}$ vs. H_0	10.465

3b. ANOVA for Emotional Change

Hypotheses compared	BF
$H_{encoding}$ vs. H_0	14.991

3c. Robust ANOVA for Emotional Change

Hypotheses compared	BF
$H_{encoding}$ vs. H_0	3.044

3d. ANOVA for Distress

Hypotheses compared	BF
$H_{encoding}$ vs. H_0	1.901

3e. ANOVA for Intrusions

Hypotheses compared	BF
H_0 vs. $H_{encoding/rapid\_con}$	1.100
H_0 vs. H_{consol}	5.868
$H_{encoding/rapid\_con}$ vs. H_{consol}	5.345

3f. ANOVA for IES

Hypotheses compared	BF
H_0 vs. $H_{encoding/rapid\_con}$	1.280
H_0 vs. H_{consol}	5.442
$H_{encoding/rapid\_con}$ vs. H_{consol}	4.250

3g. MANOVA for Verbal and Visual Declarative Memory

Hypotheses compared	BF
H_0 vs. $H_{encoding/rapid\_con}$	6.846
H_0 vs. H_{consol}	2.469
H_{consol} vs. $H_{encoding/rapid\_con}$	2.773

BF = Relative Bayes factor favoring the first hypothesis vs. the second; H_0 = null hypothesis; H_{consol} = the consolidation hypothesis; $H_{encoding}$ = the encoding hypothesis, used when assessing initial emotional responsivity; $H_{encoding/rapid\_con}$ = the encoding and rapid consolidation hypotheses, which specify the same pattern of means as $H_{encoding}$, but used for long-term assessments where consolidation can have played a role; IES = Modified Impact of Events Scale

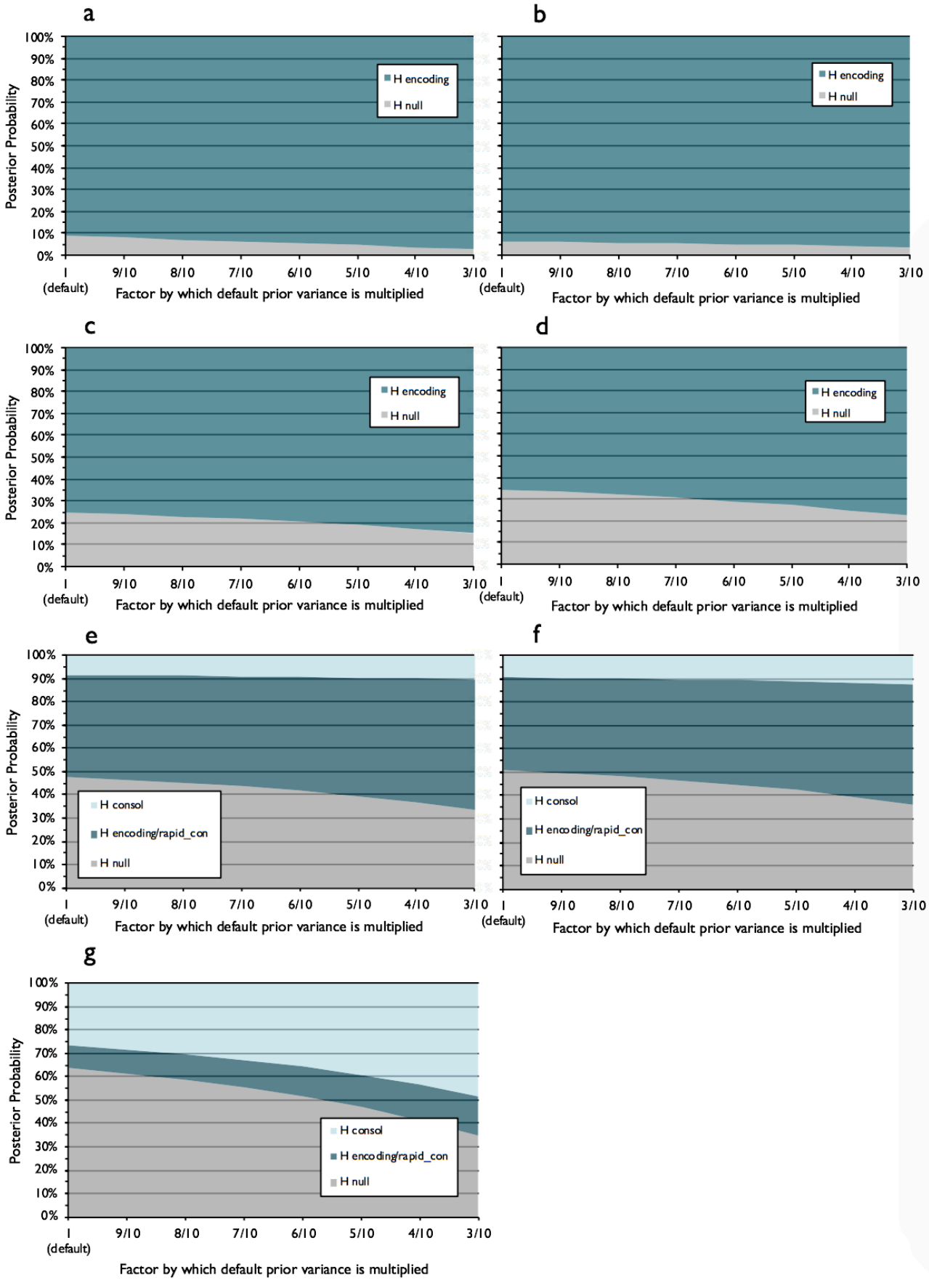
a
b
c
d
e
f
g
Posterior Probability
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
H encoding
H null
H consol
H encoding/rapid_con
1
(default)
9/10
8/10
7/10
6/10
5/10
4/10
3/10
Factor by which default prior variance is multiplied

Figure 2. Posterior probabilities associated with each hypothesis across the sensitivity analyses. a) MANOVA for emotional change and distress. b) ANOVA for emotional change. c) Robust ANOVA for emotional change. d) ANOVA for distress. e) ANOVA for intrusions. f) ANOVA for Impact of Events Scale. g) MANOVA for verbal and visual declarative memory. Note: Bayes factors for competing hypotheses can be approximated by dividing the posterior probability associated with one hypothesis by the other in these graphs.

Subsequent Bayesian ANOVAs provided further insight into primary driver of the MANOVA results (Table 3b-3d). For emotional change, the Bayes factor for $H_{encoding}$ vs. H_0 suggests that the data provides 14.991 times more evidence in favor of $H_{encoding}$ than for H_0, indicating strong evidence for $H_{encoding}$ over H_0. These findings were also robust to a sensitivity analysis (Figure 2c). As noted, two extreme outliers in change scores were removed from the prop-placebo group. We also ran a 'robust Bayesian ANOVA' with these outliers included, using 20% trimmed means and variances (Meijerink-Bosman & Hoijtink, 2018). With this conservative assessment, $H_{encoding}$ vs. H_0 (BF = 3.044) was also favored and insensitive to variations in the prior (Table 3c, Figure 2c). In the ANOVA for distress (Table 3c), $H_{encoding}$ achieved a less convincing Bayes factor of 1.901 vs. H_0 (Table 3d, Figure 2d).

In summary, the MANOVA and ANOVAs provide evidence in favor of $H_{encoding}$, suggesting that propranolol reduced the initial emotional impact of the trauma film.

Effect of propranolol on subsequent intrusions and IES. Compliance with filling in the diary was very good (M = 8.42, SD = 1.04), with no evidence for differences between groups ($F(2,71)$ = .193, p = .825, BF_{01} = 7.31, Error = .027). As in previous studies measuring intrusions and IES, there was positive skew in our two outcome variables (Cristea, Naudet, Shanks, & Hardwicke, 2017; James et al., 2015). A log transformation produced more normal distributions of the IES scores, on which the following Bayesian analyses were conducted. For Intrusions, our analysis stipulated that the variable was Poisson-distributed. Appendix 1 describes non-parametric analyses of the raw IES data.

We assessed three informative hypotheses in relation to intrusions and IES using two ANOVAs in BAIN (Table 3e & 3f, Figure 2e & 2f). H_0 was the same as in previous analyses. $H_{encoding/rapid\_con}$ jointly reflects the *encoding* and *rapid*

consolidation hypotheses ($H_{encoding/rapid\_con}$: Prop-Placebo < Placebo-Prop = Placebo-Placebo), and we added the *consolidation hypothesis* (H_{consol}: Prop-Placebo = Placebo-Prop < Placebo-Placebo). Intrusion rates and log-transformed IES scores were quite similar across groups (see Table 4, Figure 3 depicts intrusions each day). Evidence for H_0 *vs.* $H_{encoding/rapid\_con}$ was quite equivocal for both intrusions (1.100) and IES scores (1.280). Bayes factors for $H_{encoding/rapid\_con}$ vs. H_0 nevertheless remained unconvincing at the furthest end of our sensitivity analyses, at 1.662 for intrusions and 1.426 for IES scores. For both intrusions and IES scores, H_0 and $H_{encoding/rapid\_con}$ had between 4-6 times more support from the data than H_{consol}. The pattern of results is very similar when only intrusions after day 1 are considered, which may be important if consolidation effects are only observed after a night of sleep (see Appendix 1, Table S4, Figure S2). Hence, H_{consol} finds little support in the data, and the evidence is generally equivocal between $H_{encoding/rapid\_con}$ vs. H_0. Supplementary non-parametric analyses of IES scores also failed to detect significant differences between groups.

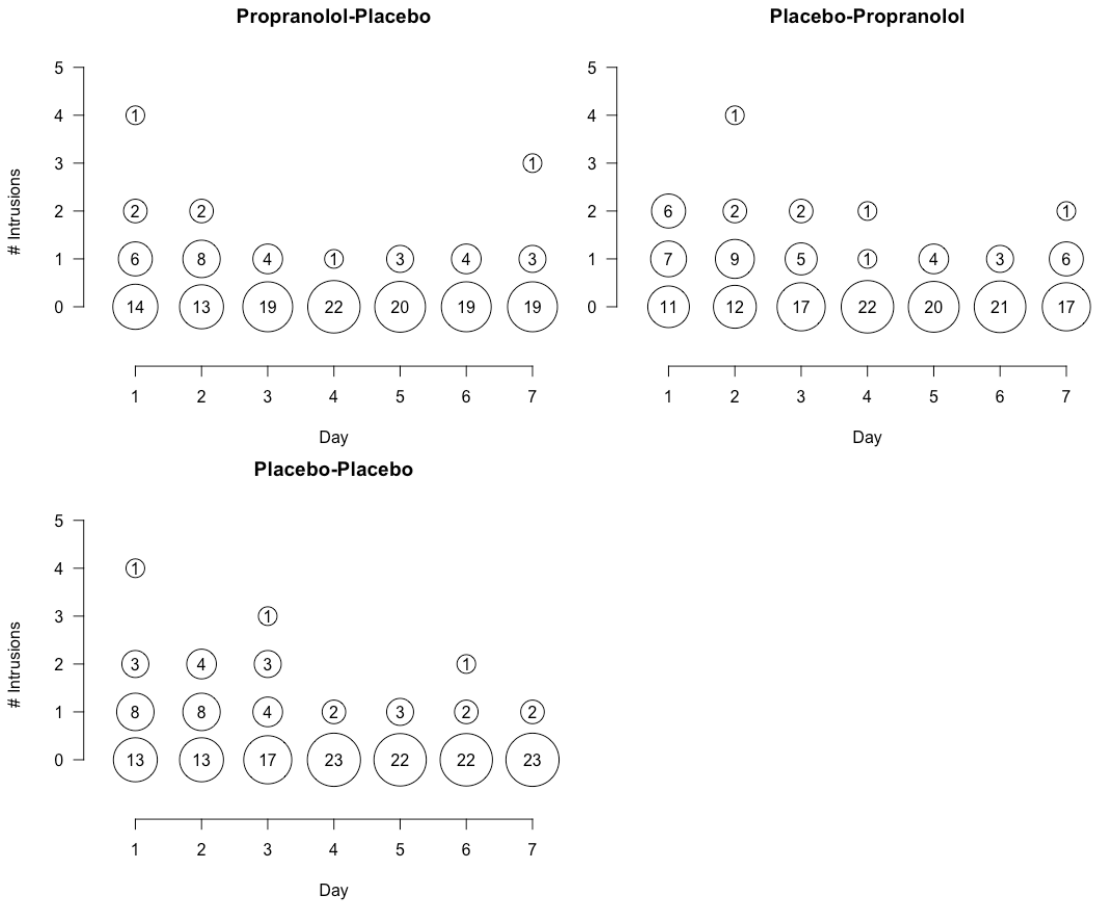

Figure 3. Daily intrusions across experimental conditions. Numbers within the circles indicate the number of participants with the respective number of intrusions.

Table 4. Descriptive statistics for intrusions and IES scores across groups.

	Intrusions			IES			
	Estimated rate over week	Median	Range	Mean (SD)*	Median	Range	Mean Rank
Prop-Placebo	0.65	2	0-8	1.83 (1.00)	9	0-18	34.07
Placebo-Prop	0.97	2	0-9	2.21 (.87)	9	0-41	40.98
Placebo-Placebo	0.84	1	0-9	2.05 (.74)	7	0-31	34.44

*mean and standard deviations are for log-transformed data, median, range, and mean rank for raw data, IES = Impact of Events Scale

Effect of propranolol on subsequent declarative memory. *D-prime* scores for each participant were used for declarative memory assessments. Data for one Placebo-Placebo participant was lost through a technical malfunction. Participants performed above chance on both tests (Visual memory: $t(70) = 13.380$, $p < .001$; Verbal memory: $t(70) = 5.035$, $p < .001$), although performance was significantly better for visual ($M = 1.23$, $SD = .78$) than verbal memory ($M = .33$, $SD = .56$) ($t(70) = 10.820$, $p < .001$). This was reflected in greater confidence in responses to visual (mean rank = 38.93) versus verbal memory (mean rank = 12.94) ($Z = 6.730$, $p < .001$).

Table 5. Means and standard deviations of visual and verbal memory *D-prime* scores.

	Declarative memory	
	Visual Mean (SD)	Verbal Mean (SD)
Prop-Placebo	1.11 (.93)	.30 (.65)
Placebo-Prop	1.16 (.81)	.42 (.50)
Placebo-Placebo	1.41 (.53)	.33 (.56)

Group means and standard deviations for *D-prime* scores are presented in Table 5, and indicate similar scores across groups. We used a Bayesian MANOVA to assess evidence for H_0, $H_{encoding/rapid\_con}$, and H_{consol} (Table 3g, Figure 2g). The data furnished 6.846 times more evidence for H_0 than for $H_{encoding/rapid\_con}$, and 2.469 times more evidence for H_0 than H_{consol}. Hence, H_0 was generally favored, but not overwhelmingly more than H_{consol}. Sensitivity analyses suggested that H_{consol} was only moderately favored relative to H_0 (1.350) even at the far end of prior variances

considered in the sensitivity analysis. Hence, of our three specified hypotheses, H_0 was most supported by the data, but H_{consol} cannot be completely rejected.

To check that floor effects in verbal memory did not obscure an influence of propranolol on visual declarative memory, we assessed visual memory in a separate ANOVA, and found that the Bayes factor for H_0 vs H_{consol} was similarly equivocal (1.190), and that evidence favoring H_{consol} was not strong even at the far end of prior variances considered (BF H_{consol} vs H_0 = 1.535).

Consideration of alternative hypotheses. Bayes factors for competing hypotheses can give a false impression of the evidence if none describe the data well: a high Bayes factor could reflect the best of several bad hypotheses. An unconstrained or 'failsafe' hypothesis can assess whether specified hypotheses reflect the data well. Posterior model probabilities associated with a failsafe hypothesis never reached 10% at the default prior variance in the above analyses, indicating that our hypotheses provided a good approximation to patterns in the data.

Relationships between emotional induction and emotional and declarative memory. Relatively few intrusions were reported, and one concern could be that such longer-term measures of the movie's impact were arbitrary. To the contrary, intrusions and IES were positively correlated with one another (Table 6). In addition, emotional change and distress were positively correlated with one another and with longer-term measures of emotional impact. Hence, the initial emotional impact at encoding predicted later emotional impact. In contrast, while visual and verbal declarative memory were correlated with one another, they were not correlated with either the initial or longer-term emotional impact of the trauma film.

Table 6. Correlations between emotional change, distress, and emotional and declarative memory.

	Emotional Change		Distress		Intrusions		IES		Log Intrusions		Log IES		Visual Memory		Verbal Memory	
	r℗p [95% CI]	*p*	*r℗p* [95% CI]	*p*	*r℗p [95% CI]*	*p*	*r℗p [95% CI]*	*p*	*r℗p [95% CI]*	*p*	*r℗p [95% CI]*	*p*	*r℗p [95% CI]*	*p*	*r℗p [95% CI]*	*p*
Emo Change	-	-	.46 [.26-.63]	<.001	.34 [.11-.52]	.004	.30 [.06-.52]	.010	.34 [.11-.52]	.004	.30 [.06-.52]	.010	.06 [-.18-.29]	.597	.05 [-.18-.29]	.669
Distress	.46 [.20-.65]	<.001	-	-	.24 [.00-.43]	.047	.37 [.15-.56]	.001	.24 [.00-.43]	.047	.37 [.15-.56]	.001	-.10 [-.34-.14]	.432	.05 [-.17-.29]	.702
Intrusions	.22 [.00-.48]	.061	.22 [.00-.43]	.060	-	-	.70 [.52-.81]	<.001	N/A	N/A	N/A	N/A	-.06 [-.29-.17]	.605	.00 [-.22-.22]	.990
IES	.34 [.12-.54]	.004	.35 [.19-.49]	.003	.67 [.52-.79]	<.001	-	-	N/A	N/A	N/A	N/A	-.01 [-.27-.26]	.929	.07 [-.18-.32]	.589
Log Int	.27 [.03-.50]	.024	.21 [-.02-.42]	.079	N/A	N/A	N/A	N/A	-	-	.70 [.52-.81]	<.001	-.06 [-.29-.17]	.605	.00 [-.22-.22]	.990
Log IES	.33 [.13-.51]	.004	.29 [.10-.47]	.014	N/A	N/A	N/A	N/A	.61 [.51-.71]	<.001	-	-	-.01 [-.27-.26]	.929	.07 [-.18-.32]	.589
Visual Mem	.13 [-.12-.36]	.276	-.06 [-.30-.19]	.606	-.10 [-.32-.13]	.406	-.03 [-.27-.21]	.801	-.04 [-.28-.20]	.767	.11 [-.19-.38]	.344	-	-	.48 [.26-.67]	<.001
Verbal Mem	.02 [-.23-.29]	.855	-.01 [-.27-.27]	.917	-.03 [-.23-.18]	.793	.00 [-.23-.25]	.971	-.01 [-.21-.20]	.913	.05 [-.20-.31]	.680	.49 [.22-.70]	<.001	-	-

N = 72, except 71 for visual and verbal memory; *r℗p* = Pearson's *r* (lower left matrix) or Spearman's *rho* (upper right matrix); all *p*-values assessed 2-tailed, 1000 sample bootstrap with bias corrected accelerated confidence intervals.

Discussion

We aimed to assess whether propranolol could reduce the negative impact of an emotional event and, if so, what mechanism best accounted for the results: an effect of propranolol on initial encoding or very early consolidation process, or a clear effect on post-encoding consolidation. Our data provided most evidence for no effect of propranolol administered either before or after the movie on the later negative consequences of having watched the movie, though Bayes factors indicated that an impact of pre-encoding propranolol on reducing the later negative impact of the analogue trauma could not be completely ruled out.

Previous laboratory studies found that propranolol administered 60+ minutes before emotional learning affected the emotional enhancement of memory (Lonergan et al. 2013). Thomas and colleagues (2017) suggested that a consolidation-blocking effect of pre-learning propranolol was a better explanation than an impact on encoding/acquisition, as physiological reactivity during encoding was not significantly affected by propranolol in their studies. As pre-learning propranolol could also affect a rapid post-learning consolidation process, this remains possible. However, we found that the negative emotional state induced by the movie was less intense in participants who received propranolol beforehand, indicating an effect of propranolol on the film's initial encoding and emotional impact. Moreover, greater mood induction was positively correlated with intrusions and self-reported impact of the experience one week later. Hence, if there were an effect of pre-learning propranolol on the later negative impact of the trauma film (which could not be ruled out), it might be partially explained as an encoding or acquisition effect, rather than interference with post-learning consolidation.

Of course, effects on encoding can be expected to lead to different levels of memory consolidation, and certain processes important for consolidation may be initiated during learning and only prevented by pre-learning propranolol. For example, some animal studies indicate that the effect of beta-adrenergic signaling on long-term memory was already initiated during learning, and was related to a delayed action of ERK, stimulated by *β*-adrenergic activation *during* encoding (Schiff et al., 2017). While pre-acquisition propranolol blocked long-term memory formation, even rapidly delivered intracerebral post-encoding blockade of *β*-adrenergic receptors was found to have no effect (Bush, Caparosa, Gekker, & LeDoux, 2010; Dębiec & LeDoux, 2004). Hence, *β*-adrenergic receptors may be important for both

encoding/acquisition and consolidation of emotional memory, but in some cases even delayed effects on consolidation are initiated during learning. It may thus be useful to differentiate between delayed effects on consolidation that begin during acquisition, and those that can be produced by post-acquisition manipulations. This distinction is crucial for the feasibility of using post-trauma propranolol to block consolidation. To more specifically differentiate between encoding and rapid consolidation effects in humans, it may be necessary to assess more rapid means of drug delivery, such as intravenous injection. One could also assess effects of the manipulation on short-term memory expression, which should be largely unaffected if only consolidation is being disrupted (Schafe & LeDoux, 2000).

While a mechanistic understanding of treatment effects is useful in developing evidence-based interventions, it is sometimes less consequential for the patient whether potential benefits from taking propranolol result from blocking consolidation or another mechanism. This study and those using the emotional slideshow paradigm suggest that post-learning oral propranolol is unlikely to have an effect on later memory formation. However, pre-'trauma' propranolol administration is conceivably useful in some cases. When not contraindicated, propranolol might be useful for reducing stress for predictable events such as certain kinds of operations, and may also reduce later negative effects. In such cases, any effects of propranolol on declarative memory would not pose severe ethical concerns (cf. Elsey & Kindt, 2016; Kolber, 2006), and the hospital setting would provide practical advantages for drug administration. Effects of propranolol on emotional experiences could potentially also explain some observed effects of post-trauma propranolol in clinical studies. High or repeated doses of propranolol could reduce the negative mood and distress caused by thinking about the trauma afterwards and may also more rapidly reduce any lingering arousal. This may reduce the desire to suppress such thoughts and images and consequently reduce intrusions or other symptoms (c.f., Ehlers, Mayou, & Bryant, 1998). After very distressing experiences, it is also possible that the memory of the episode incorporates not just the specific event itself, but the subjective impact of it and one's subsequent feelings, which may be less negative under the influence of propranolol. However, we would caution against strong conclusions based on current experimental findings. Even the trauma-film paradigm, which may more closely approximate a trauma than an emotional slideshow does, remains far-removed from a truly traumatic experience. Pitman (1989) has further suggested that effects of

propranolol administered after trauma might also affect stress hormones released upon remembrance of the traumatic event, which would likely only occur when extremely distressing events are recalled.

Beyond assessing propranolol's impact on the negative sequelae of the trauma film, we assessed declarative memory. Our results provided most support for no effect of propranolol on verbal or visual declarative memory, although we could not rule out a consolidation effect. Unlike story slideshow studies, 'emotional' and 'neutral' elements are difficult to differentiate in the trauma film paradigm: each scene is closely preceded by a distressing event and, if it starts out neutral, rapidly becomes negative. However, particularly the items used in the verbal declarative memory test may be regarded as quite neutral, involving statements of fact about clothing in a scene, for example. In the current study, verbal and visual declarative memory were strongly correlated with one another, and both our indices of emotional memory/impact were correlated with one another. There was no relationship between declarative and emotional memory measures, nor was declarative memory correlated with the initial emotional impact of the movie. Hence, these declarative memories may have been of a more neutral kind than the 'emotional' declarative memories that were affected by propranolol in previous studies (Lonergan et al., 2013), and thus largely unaffected by propranolol.

Some limitations of our study must be considered in interpreting the results. Firstly, our participants experienced few intrusions. A floor effect may prevent a possible impact of propranolol from being observed – an issue found in other trauma film studies (Siegelsleitner et al., 2019). More graphic or personally disturbing scenes can be expected to produce more intrusions, but also raise ethical concerns about negatively affecting participants. It is also possible that having English audio for the film clips could have prevented an immediate emotional connection with the film clips, despite Dutch participants typically having high competency in English (Siegelsleitner and colleagues' participants were not English either). Though low intrusions may have precluded observation of group differences, the numbers of intrusions reported were not meaningless or arbitrary, as they correlated with the initial emotional induction. An effect of propranolol on the initial emotional impact of the trauma film is also informative as to an impact of propranolol on encoding even in the absence of a clear effect on intrusions. While we cannot be sure that effects of

propranolol on intrusions would not emerge if participants had more intrusions overall, propranolol nevertheless appears to have affected encoding.

We did not observe a clear effect of propranolol on blood pressure and heart rate. This may owe to when we conducted measurements (at the very beginning and end of the experiment, to avoid unblinding the experimenters) and to overall decreases over time in all groups. This collection approach was selected to avoid possibly unblinding the experimenters to the pill allocations if they saw drops in heart rate during the experiment. We expected that heart rate changes from pill 2 would be apparent by 60-75 minutes after administration, and that changes would still be observable after a longer duration for participants who received propranolol in their first pill, as the decline in propranolol's bioavailability is not sharp. In future studies, using an unobtrusive and continuous measure of heart rate could firstly help keep experimenters blind to any changes (if the collection device is kept out of sight), while also detecting possible differences in the *rate* of heart rate change, should all groups experience a decline due to prolonged rest.

The possibility of insufficient dosage should also be considered, though 40mg has been sufficient in a host of *re*consolidation studies (Soeter & Kindt, 2013) and is commonly used in consolidation experiments (Lonergan et al., 2013). In an exploratory restriction of analyses to women (Appendix 1, Table S7 and Figure S3), who likely had lower body weight than men, we observed a slightly clearer effect of propranolol on heart rate. More notably, among women, we found Bayes Factors for $H_{encoding/rapid\_con}$ vs. H_{null} of approximately 3-4 at default prior variances, and from 13 to 18 for $H_{encoding/rapid\_con}$ vs. H_{consol}, in determining the impact of propranolol on intrusions and IES scores. Hence, using higher dosages or a larger sample of just women could yield more evidence for propranolol affecting later outcomes. Nevertheless, the effect of propranolol on emotional induction suggests that propranolol was likely operating effectively.

Despite our instructions, several participants reported consuming alcohol on the night of the first study session. We did not find evidence that alcohol consumption differed between groups, or that alcohol consumption was related to emotional or declarative memory, and therefore do not expect that alcohol consumption significantly influenced our findings (see Appendix 1, Table S8). Finally, a general consideration using the trauma film paradigm with paper diary entries is that we cannot be certain participants are truly recording intrusions each day or as they

happen, and may forget to complete the diary or even invent intrusions. Self-report measures of diary adherence are of course subject to social desirability effects. At present, this limitation cannot be avoided, though digital input of intrusions could at least clearly indicate exactly when intrusions were recorded. Relationships between intrusions and other meaningful measures such as the emotional response to the movie in this and many other studies (James et al., 2016), however, indicate the general validity of this admittedly imperfect data collection approach.

Conclusion

Propranolol has been proposed as a means of preventing the development of PTSD symptoms after trauma, but much experimental work has focused on administering propranolol *before* emotional events. Clinical studies are hampered by practical difficulties of administering propranolol shortly after trauma. Using a 'trauma film', we were unable to find a convincing effect of propranolol on emotional or declarative memory, although an effect of pre-learning propranolol on emotional memory, and of pre- and post-trauma propranolol on visual declarative memory, could not be completely ruled out. Notably, an absence of effects of propranolol on later emotional memory could be due to the generally low later impact of the emotional scene. However, we did find an effect of propranolol on the initial emotional impact of the scene, indicating that propranolol can affect encoding, and raising possible difficulties in interpretation when only pre-encoding propranolol is used to investigate consolidation. Study designs and analytic approaches designed to assess competing hypotheses can help to elucidate the mechanisms of propranolol's effects on memory.