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Unraveling the universality of chemical fear communication: evidence from behavioral, genetic, and chemical analyses

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Abundant evidence indicates that humans can communicate threat-related information to conspecifics through their body odors. However, prior research has been primarily conducted on Western (WEIRD) samples. In this study, we aimed to investigate whether threat-related information can be transmitted by individuals of East Asian descent who carry a single-nucleotide polymorphism (SNP) 538G → A in the ABCC11 gene, which significantly reduces (noticeable) body odor. To examine this, we recruited 18 self-identified male East Asian AA-homozygotes and 18 self-identified male Western individuals who were carriers of the functional G-allele. We collected samples of their fear-related and neutral body odors. Subsequently, we conducted a double-blind behavioral experiment in which we presented these samples to 69 self-identified female participants of Western Caucasian and East Asian backgrounds. The participants were asked to rate faces that were morphed between expressions of fear and disgust. Notably, despite the "odorless" phenotypic expression of the ABCC11-mutation in East Asians, their fear odor caused a perceptual fear bias in both East Asian and Caucasian receivers. This finding leaves open the possibility of universal fear chemosignaling. Additionally, we conducted exploratory chemical analysis to gain initial insights into the chemical composition of the body odors presented. In a subsequent pre-registered behavioral study (*N* = 33), we found that exposure to hexadecanoic acid, an abundant compound in the fear and neutral body odor samples, was sufficient to reproduce the observed behavioral effects. While exploratory, these findings provide insight into how specific chemical components can drive chemical fear communication.

Key words: olfaction, emotion, chemical communication, ABCC11 gene, analytical chemistry, pre-registered.

Millennia-old beliefs that humans are poor smellers have hindered scientific progress in debunking this myth (Le Guérer 2002). Over time, pseudoscientific notions that our expanded frontal lobes and unique rationality resist "animalistic" olfactory urges have been abandoned. Empirical studies have highlighted humans' excellent sense of smell, including its role in social communication, thus dispelling these ideas (Stevenson 2010; de Groot et al. 2017; McGann 2017). The quest for human pheromones, considered one of the most significant knowledge gaps across scientific disciplines (Kennedy and Norman 2005), has gained attention. Several studies have demonstrated that human body odors can convey biologically significant states such as sickness (e.g. Olsson et al. 2014) and emotions like disgust (e.g. de Groot et al. 2012), anger (e.g. Pause et al. 2020), happiness (e.g. de Groot et al. 2015), and fear (e.g. Chen and Haviland-Jones 2000).

Here, our focus is on the emotion fear, which has received robust empirical support from psychological and neuroscience research (de Groot and Smeets 2017) and carries survival-related information (Darwin 1872/1998; Susskind et al. 2008). We examined the cross-genetic universality of fear communication through body odor. Previous research has shown fear communication in Western Caucasian populations, but the generalizability is uncertain. In Study 1, we investigated behavioral and physiological responses to fear odor in Western Caucasian and East Asian individuals, considering the influence of a different genotype directly impacting body odor (Study 1A, 1B). Additionally, we analyzed the chemical compositions of fear and neutral body odor samples across different populations (Study 1C). In Study 2, we evaluated the effectiveness of individual substances to elicit behavioral and physiological responses comparable to body odor samples. The findings will contribute to understanding chemical fear communication and its genetic variations.

The primary objective of Study 1 was to examine if the human ability to chemically communicate fear from a sender to receiver is broadly shared across different genotypes. Previous studies have provided robust support for this phenomenon, employing well-controlled double-blind experiments that used various indicators of successful fear

communication, such as stronger startle reflexes (e.g. Prehn et al. 2006), increased amygdala activity (e.g. Mujica-Parodi et al. 2009; de Groot et al. 2021), and fearful facial expressions (e.g. de Groot et al. 2014; Kamiloglu et al. 2018). However, this evidence is based on Western, Educated, Industrialized, Rich, Democratic (WEIRD) populations. It is important to consider that assuming universal processes without accounting for underlying genotypes can lead to erroneous conclusions (WRONG: Gaertner et al. 2010). While all humans benefit from successful threat detection, there may be a specific gene variant hindering the chemical signaling of fear. Previous research unknowingly overlooked a non-WEIRD population of over a billion individuals who carry a single-nucleotide polymorphism (SNP) 538 G → A in the ABCC11 gene. This SNP is responsible for dry earwax (Yoshiura et al. 2006) and weaker armpit odor (Martin et al. 2010; Harker et al. 2014). The mutation occurred around 30–40,000 years ago in a Northern Mongoloid tribe and rapidly spread to the Far East, reaching over 95% prevalence in countries like China, South Korea, and Japan (Yoshiura et al. 2006), arguably due to positive selection pressure for the phenotype (Natsch and Emter 2020).

The ABCC11 gene plays a crucial role in the secretion of (mal)odorants and their precursors from the apocrine sweat glands in the armpit. The armpit has been the primary location for sampling fear odor (de Groot and Smeets 2017) because axillary apocrine sweat glands contain β-adrenoceptors that are activated during fear experiences (Harker 2013). However, individuals need to carry at least one copy of the G-allele on the ABCC11 gene, which is present in 97–100% of Western Caucasians (Yoshiura et al. 2006), to secrete characteristic components from the apocrine glands that contribute to axillary (mal)odor. Additionally, G-allele carriers bear significantly higher amounts of odoriferous steroids, such as 5α-androst-16-en-3-one (Martin et al. 2010). On the other hand, both G-allele carriers and "non-odorous" AA haplotypes emit comparable levels of long-chain fatty acids. The AA genotype produces only a faint acidic odor (Martin et al. 2010), suggesting that chemical communication may have lost its adaptive advantage in the Far East (Natsch and Emter 2020).

We aimed to test this hypothesis in a behavioral experiment, evaluating whether axillary odors from East Asians transfer fear information. In this pre-registered research (https://osf.io/c5wb2/), we employed an established behavioral paradigm to investigate the potential species-wide human capacity to produce and perceive the smell of fear. We tested groups with and without the crucial genetic variant (A-allele vs. G-allele) to provide falsifiable evidence for universality. Knowing that exposure to Western Caucasian fear odor induces fear-specific processes in receivers (e.g. fearful facial expressions, enhanced startle reflexes, and seeing more fear in ambiguous facial expressions; meta-analysis: de Groot and Smeets 2017), presumably through emotional contagion (Hatfield et al. 1993), we hypothesized: (H1) Exposure to fear odor (vs. neutral) from G-allele Western Caucasians induces *fear contagion* in both Western Caucasian and East Asian receivers. Fear contagion was operationalized by having receivers rate facial expressions morphed between fear and disgust as more fearful (Zhou and Chen 2009; de Groot et al. 2021). By using this fear-disgust dichotomy, we aimed to distinguish between the veritable communication of emotional content (fear) and any potential bias introduced by the superficial perception of sweat as a merely unpleasant odor (disgust) (Alaoui-Ismaïli et al. 1997), creating a falsifiable case. We also explored fear-specific physiological changes. (H2) Assuming universality as the default, exposure to fear odor (vs. neutral) from A-allele East Asians would induce fear contagion in both Western Caucasian and East Asian receivers. (H3) We employed a Bayesian analysis approach to examine whether there is evidence for no difference in the way fear odor from individuals with different genotypes affects fear responses in Western Caucasian and East Asian receivers.

The second objective of Study 1 was to explore the similarities and differences in the chemical compounds present in the body odor samples collected from the participants. To identify potential pheromones, several steps are needed (Wyatt 2020). This involves conducting repeatable bio-assays to demonstrate that putative pheromones elicit pre-defined behavior (Step 1), followed by isolating, identifying, and synthesizing the chemical substances using chemical analysis (Step 2), and, finally, confirm that key molecules in their natural concentrations can recreate the bioassay-observed behavior (Step 3) (Wyatt 2020). To our knowledge, only one published study on chemical communication of fear via body odors has gone beyond the initial behavioral experiments (Smeets et al. 2020). Initial evidence was gained from Caucasian samples: the composition of armpit odor of fearful individuals was significantly different from those individuals' odor when feeling neutral or happy (Smeets et al. 2020). Here, we aimed to gain insights into the composition of fear and neutral body odor samples across Western and non-Western samples. Due to the limited samples, formal statistics were not performed. The aim of the chemical analysis was to identify any qualitative or quantitative differences between the sample pools. We hypothesized that compositions will differ between senders experiencing fear and those in a neutral state.

Finally, based on the outcomes of the behavioral and chemical analysis, Study 2 was conducted to determine whether individual constituents of the body odor samples are sufficient to reproduce the expected behavioral effects observed in natural body odors.

Methods

This research consisted of 2 studies, namely: Study 1, which comprised 3 parts: (i) a body odor collection experiment involving senders (Methods and Results reported in the Supplementary Materials); (ii) a body odor exposure experiment involving receivers; and (iii) chemical analysis; and Study 2, an experiment where a new group of receivers was exposed to individual constituents of the body odor samples. All studies complied with the Declaration of Helsinki for Medical Research Involving Human Subjects, with approval from the Ethics Review Board of the Faculty of Social and Behavioral Sciences of Utrecht University (20-0184) and blanket approval from the Donders Center for Cognitive Neuroimaging. Informed consent was obtained from all subjects.

Participants

To determine the sample size, we referred to prior research by de Groot et al. (2021) that showed fear sweat induced a fear bias in perception. Based on a medium effect size ($d = 0.51$), an α of .05, and 80% power, G*Power 3.1 determined a minimum sample size of 33 per receiver group (Western

Caucasians, East Asians). To account for potential drop-outs, we slightly oversampled, aiming for a maximum of 36 observations per group. Study 1B involved 69 self-identified female receivers (36 Western Caucasian: M = 23.17 years, SD = 3.35 years; 33 East Asian: M = 26.15 years, SD = 5.62 years). Considering a 3:8 sender–receiver ratio (de Groot et al. 2021), the allocation of body odor samples for chemical analysis (¼) and behavioral experiments (¾), and the finding that fear odors can be presented at least twice without altering responses (Gomes et al. 2020), we recruited 18 senders per genotype group (ABCC11 A-allele, ABCC11 G-allele) to provide sufficient material for the receivers in study 1. For Study 2, 33 healthy self-identified female Caucasian receivers were recruited (M = 22.82 years, SD = 3.94 years).

Self-identified females were chosen as they outperform males on any olfactory task (Sorokowski et al. 2019) and are more sensitive to the smell of fear (de Groot et al. 2014), enhancing the study's power. Screening criteria were applied (Doty 2001) to ensure participants had a functional sense of smell, were non-smokers, aged between 18 and 40 years, not pregnant, had no respiratory diseases or allergies, and did not use drugs or medications that could affect their perception. Participants were also excluded if they had a history of psychiatric disorders or belonged to a high-risk group for COVID-19. Recruitment was done through the University's Facebook channel, word-of-mouth, and the university's participation credit system, and participants were compensated financially (€10/hour; €24) or with course credit.

Materials

Odor presentation

Study 1:

The odor stimuli were body odors collected in Study 1A, namely fear and neutral sweat samples from 18 G-allele Western Caucasian and 18 A-allele East Asian self-identified males (see Supplementary Materials, for Study 1 Methods). The 4 body odor stimuli will from now on be labeled as follows: G-Fear, A-Fear, G-Neut, A-Neut. Senders provided 200 cm² of sweat samples, while receivers smelled 75 cm² of material (3:8 sender:receiver ratio = 600 cm²/8 = 75 cm²). To minimize variations in sweat production between individuals, the 75 cm² of odor stimulus for a receiver was a combination of sweat samples from 6 different senders, each contributing 12.5 cm². The samples were exposed using the olfactometer for approximately 30 min and reused once (i.e. Western Caucasian receiver #1 and East Asian receiver #1 were exposed to the same stimulus). An olfactometer with 7 channels (Dancer Design, St. Helens, UK), controlled by a computer and connected to a medical-grade air tank (AIRAPY Linde Gas, Schiedam, NL), was used to present the odors. Each axillary sample was placed in a glass bottle sealed with a plastic cap. The bottle had 2 polytetrafluoroethylene (PTFE) tubes (4 mm ø) attached, one for inlet, one for outlet, secured by collect-type seals. The glass bottle was filled halfway with fiber wick to maximize the surface area for evaporation. Air flow from the gas tank entered the olfactometer, and one of the 4 different valves connected to the glass bottles would randomly open at preprogrammed moments, allowing the odors to be delivered to a nasal end piece connected to the participant's nose. The timing of odor presentation was controlled using parallel ports within an Inquisit 6 script. In addition to the 4 odor valves, a fifth valve was connected to an empty glass bottle. This "air" valve was opened when no odor was presented to ensure stable air flow and minimize cross contamination. The average air flow across conditions was 2.89 l/min (SD = 0.13 l/min). To prevent contamination, the plastic caps were disinfected with alcohol and dried for 30 min before switching odors.

Study 2:

We evaluated behavioral responsiveness to 3 different acids and an odorless air condition. Hexadecanoic acid (HDA) was selected in view of its high abundance in the sweat samples and apparently increased amounts in fear samples. Propanoic acid (PA) was used as a representative of short-chain acids occurring in the samples, and isovaleric acid (IVA) was selected as a branched acid, which was expected to induce disgust rather than fear based on previous results (de Groot et al. 2021). The odor conditions were as follows: (i) Odor 1: HDA; (ii) Odor 2: PA; (iii) Odor 3: IVA; (iv) Odorless control condition: Air. The concentrations (1.5 ppm HDA, 1 ppm PA, 0.1 ppm IVA) were selected to be of low intensity. All odors were presented in 10 mL mineral oil.

Face morphs

We deliberately created a visually demanding face morph task for participants, where they would need to rely on sources of information beyond visual cues (i.e. olfactory information) to make their intuitive decisions after brief face presentation. This approach finds support in established research paradigms where brief (200 ms) stimulus durations have been intentionally and effectively employed in studies such as de Groot et al. (2021); Zhou and Chen (2009), and Mujica-Parodi et al. (2009). Notably, the strongest odor-biasing effects were typically observed at the most ambiguous visual level for 200 ms presented faces (Zhou and Chen 2009; de Groot et al. 2021), enhancing statistical power for this receiver task. The current pre-validated morph task (de Groot et al. 2021) involved rating grayscale images of fearful and disgusted facial expressions from two different male Caucasians (actors 28 and 33 of the Radboud Face Database) (Langner et al. 2010). These images were modified by removing external features like hair, ears, and background, and then morphed to create ambiguous facial expressions. To tailor the task to each participant, their individual point of subjective equality (PSE), which represents the most ambiguous face (i.e. the morph with a 50–50 "fear"–"disgust" decision), was determined for each actor through a primer morph task prior to the main experiment. If a participant had a PSE for a given actor's face morphed with 40% veridical fear and 60% disgust, they would be presented with that actor's face morphed with 25% (−15), 31% (−9), 37% (−3), 43% (+3), 49% (+9), and 55% (+15) veridical fear during the main experiment.

Procedure

To comply with COVID-19 regulations, the experiments were divided into an online and on-site portion.

The online portion was completed using Qualtrics and Inquisit 5 Web on participants' personal laptop or computers. Initially, participants filled out a screening questionnaire, including a self-administered smell test. This test assessed their smell ability over the past 6 months and required them to rate the intensity of a self-selected food item. Participants with poor or non-existent smell ability or who rated the food

item's intensity as barely perceivable were excluded. If they passed the screening, they received an information letter. After providing informed consent, participants completed the primer morph task within the Inquisit Player to determine their individual PSEs for the on-site experiment. The primer morph task involved categorizing faces morphed around the objective equality point (50% fear) by matching them to the word "fear" or "disgust" on the left or right side of the screen using keyboard buttons. The word locations were counterbalanced between blocks. Participants completed 2 practice blocks of 4 trials each, using prototypical expressions (i.e. actors' faces at 100% fear and 100% disgust), followed by 4 blocks of 32 trials each with morphed faces.

After completing the primer morph task, participants scheduled an appointment for the on-site experiment. Approximately 15–20 min before the scheduled arrival, sweat samples were thawed from the freezer for use in the main experiment. During the on-site testing, participants were situated in a separate room from the experimenter (blind to the odor condition), with communication maintained through an intercom system. To adhere to COVID-19 distancing rules, participants self-applied the electrodes for skin conductance measurement and the nosepiece (two 10 cm-long PTFE tubes, ø 4 mm) connected to the olfactometer for odor presentation. Skin conductance, a physiological measure of fear, was recorded using Acqknowledge software (BIOPAC Systems Inc.), with electrodes applied to the middle phalanges of the index and middle finger of the non-dominant hand. To test whether participants could distinguish between the smells consistently and above chance, we conducted a 2-Alternative Forced-Choice Reminder Task (2-AFCR) following the method outlined by van Hout et al. (2011). Participants completed a total of 12 trials, or 12 rounds of testing. In each trial, participants were first exposed to a reference odor (denoted as "R"). After that, they were presented with 2 test odors, with one of them being R, the other a comparison odor presented in counterbalanced order. The participants' task was to select R from the 2 test odors (50% chance level). Each of the 4 odors used in the main experiment served as the reference odor 3 times, each time compared to one of the 3 non-reference odors. This created 6 unique comparison pairs: A-B, A-C, A-D, B-C, B-D, C-D; adding the inverse of this order (all counterbalanced) makes 12 trials. The main task, the morph task, followed the discrimination task. Participants completed 2 practice blocks and then 5 blocks of 24 trials each. Each trial included a unique combination of 6 different morph levels and 4 different odors. Odors were randomly presented with an inhale cue a few seconds before morph presentation (Fig. 1). Following the morph task, participants rated the intensity and pleasantness of the odors and odorless air to account for potential hedonic differences. During debriefing, participants were informed about the nature of the odors and their expected effects on behavior.

The procedure for Study 2 was almost identical to Study 1B; yet, Study 2 did not have the primer morph task or the skin conductance measure.

Statistical analysis

Statistical analyses were generally conducted using JASP 0.15 (JASP Team 2021), and for specific tasks such as fitting Sigmoid curves in the primer morph task to calculate participants' PSE (Point of Subjective Equality) and for data visualizations, RStudio (RStudio Team 2020) was employed.

Study 1B: Data exclusion.

One participant from the Caucasian receiver group who reported awareness of odors and their effects on behavior was excluded. Consistent with our pre-registration, where we outlined the criteria for data exclusion in the morph task, we excluded the data from one Asian receiver (missingness rate 46.67%). The exclusion was based on the predefined criterion that any participant with more than 20% of non-valid responses on the morph task (defined as responses lasting less than 200 milliseconds, responses exceeding 2,500 milliseconds, or missing responses) would be excluded. Problems with recording skin conductance occurred for 2 Caucasians and 5 East Asians, resulting in a total of 35 Caucasian and 32 Asian subjects for the morph task (minus 2 for skin conductance) data analysis.

Study 1B: Skin conductance.

For skin conductance measurements, tonic mean skin conductance level (SCL) and skin conductance response amplitude (SCR amp) were used as outcome measures. This differed from the pre-registration and analyses in Study 1, where skin conductance responses per minute (SCR/min) were used. Most SCRs only *start* 2 s after a stimulus (Sjouwerman and Lonsdorf 2018; Amin and Faghih 2022) and may not top within the 4.35 s measurement window used in Study 2 (Fig. 1), meaning the SCR would go undetected, which is not the case for amplitudes, simply the highest SCL in the 4.35 s interval. We chose this relatively short measurement window to avoid potential confounds introduced by the timing of visual stimulus presentation; yet, we understand that this choice may have limited our ability to capture the full range of skin conductance responses. Due to non-normality, a Friedman's test was used on skin conductance data.

Study 1C: Chemical analysis.

Chemical analysis was conducted on sweat samples obtained from a subset of senders in Study 1A, which were not previously used in the facial morph experiment (Study 1B). This particular subset consisted of 9 G-allele and 9 A-allele senders. Each participant provided a single armpit sample (body side counterbalanced) for each emotion condition (notably, the sweat samples from the other armpits of the same senders were used in Study 1B, alongside sweat samples from an additional 18 senders). This division resulted in the creation of 4 distinct sample pools: G-Fear, A-Fear, G-Neutral, A-Neutral. Please refer to the Supplementary Materials, where we document
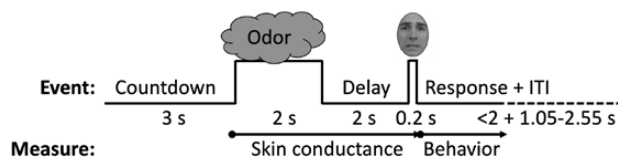
Fig. 1. Depiction of trial sequence in face morph task. Note. Each trial on the face morph task began with a 3-s visual countdown (3, 2, 1), followed by the instruction "INHALE" (2 s). The odor valve opened 150 ms before the inhale cue to ensure odor presentation within the 2-s window. The odor valve closed once the inhale cue disappeared. After a 2-second delay, a face morph was briefly presented (200 ms), and skin conductance was measured throughout the 4.35-s interval. Participants categorized the morphs as "fearful" or "disgusted" by pressing a key. Inter-trial intervals were jittered.

effective emotion induction in this subset of senders based on subjective and physiological indicators, demonstrating that this subset (n = 18) providing material for chemical analysis is representative of the total sample of Senders (N = 36) providing material for the Receiver experiment. The composition of pooled samples was of interest to us for 2 main reasons. First, pooling samples is a common practice in psychological experiments to study chemical communication, aiming to minimize variation in receivers' responses due to individual sample properties. Second, body odor samples exhibit variation between and within individuals, with a higher number of detected peaks than available samples. Analyzing such high-dimensional data ideally requires a large number of samples, which was not feasible in our study. Instead, we aimed to explore whether a general pattern of volatiles related to the emotional state would emerge when pooling the samples, thereby reducing the impact of compounds that occur in only a few samples and inter- and intra-individual variations. However, due to the pooled samples, standard statistical analysis was not possible as we only had two data points per condition. Therefore, the results were descriptive in nature.

The samples were extracted using 300 ml of freshly distilled dichloromethane (DCM) at room temperature for 30 min. After decanting, solvent-assisted flavor evaporation (SAFE) (Engel et al. 1999) was performed at 55 °C. The distillate was then dried over anhydrous sodium sulfate and concentrated to 100 μl at 50 °C using Vigreux and micro distillation methods (Bemelmans 1979). The 4 distillates were analyzed using an Agilent 7890 A GC (Agilent, Santa Clara, CA, USA) equipped with a DB-FFAP column (30 m × 0.25 mm, film thickness of 0.25 μm, J&W Scientific, Agilent Technology, Santa Clara, CA, USA). Helium was used as the carrier gas, at a flow rate of 1.0 ml/min in the constant flow mode. The distillates were injected in cold-on-column mode at 40°C using a multipurpose autosampler MPS2 (Gerstel GmbH & Co.KG, Mülheim an der Ruhr, Germany), with an injection volume of 1 μL. The temperature program involved specific heating and holding steps: 2 min at 40 °C, heating up to 240 °C with a ramp of 8 °C/min, holding 240 °C for 10 min. Mass spectrometric data were recorded in scan mode (40–400 m/z) with an ionization energy of 70 eV using an Agilent 5975 C MS (Agilent Technology, Santa Clara, CA, USA). The chromatograms were screened for the most abundant peaks. The ten signals with the highest peak areas were tentatively identified by comparing their mass spectra with the NIST 20 standard reference database (National Institute of Standards and Technology, Gaithersburg, USA). The compounds selected for quantitative analysis were further identified by comparing their mass spectra and retention indices with a reference compound. The concentrations of the target compounds in the distillates were estimated using external calibration (see Supplementary Materials for details).

Study 1B and 2: Face morph task.

Following previous studies (Mujica-Parodi et al. 2009; de Groot et al. 2021), responses that took less than 200 ms ($M_{Caucasian}$ = 0.09%, SD = 0.56%; M_{Asian} = 0.81%, SD = 2.65%) or more than 2,500 ms ($M_{Caucasian}$ = 1.33%, SD = 2.77%; M_{Asian} = 2.68%, SD = 4.29%) were excluded. Any variables with a missingness of 40% or more were median imputed.

The point of subjective equality (PSE) was calculated for each participant. The 6 most ambiguous morphs around the PSE were selected for the main experiment. A sigmoid curve was fitted to each participant's data to determine the PSE:

$$\text{Proportion of face morphs categorized as ''fear''}(y) = a + \frac{b}{1 + e^{(-c*(x-d))}}$$

In this function, the variables represent specific parameters: "a" for the y-offset, "b" for the height of the curve, "c" for the slope, "d" for the inflection point, and "x" for the morph step. Coefficients "a" and "b" were set to 0 and 100, respectively, using R to anchor the lower and upper asymptotes. The values of "c" and "d" were estimated. The PSE was then determined:

$$\text{Estimated Point of subjective Equality}(x) = -\frac{\ln\left(\frac{a+b-50}{50-a}\right)}{c} + d$$

For subjects where a sigmoid curve could not be fitted, we attempted to fit a curve using the means of the closest morph steps. If a curve still could not be fitted, it was because the majority of responses consistently leaned toward one extreme (either above or below 50% fear decisions). In these cases, a PSE of one morph step below or above the smallest or highest morph presented was assigned.

The primer morph task results showed a significant interaction between actor and group, $F(1,65) = 8.57$, $P = 0.005$, $\eta^2 = 0.06$, indicating the need to calibrate the PSE for each individual. Adjusted morph levels based on the primer task were used as stimuli in the main experiment.

To test the effect of fear odor on the receiver's fear perception, a pre-registered repeated measures (RM) ANOVA with factors of Sender Emotion (2 levels: Fear odor vs. Neutral odor), Sender Genotype (2 levels: A-allele vs. G-allele), Morph Level (6 levels: —15% Fear, —9% Fear, —3% Fear, +3% Fear, +9% Fear, +15% Fear), and Receiver Group (2 levels: Asians vs. Caucasians) was conducted on the proportion of face morphs categorized as fearful. To enhance our control over Type 1 errors, we employed the Holm-Bonferroni method (Holm 1979) to adjust for unplanned multiple hypothesis tests. This approach is considered more conservative than the traditional Bonferroni correction because it sequentially adjusts the significance level (α) for each test based on the ranking of *P*-values. *P*-values are ranked smallest to largest. The largest *P*-value gets compared to the original α level (α = 0.05). The smaller *P*-value(s) get(s) compared to an adjusted α level: α'$_{(i)}$ = α/N—i + 1), where α'$_{(i)}$ represents the adjusted α level for the i-th test, and N is the total number of hypothesis tests. If an observed *P*-value is ≤α'$_{(i)}$, we reject the null hypothesis for that specific test.

Study 1B and 2: Sensory tests.

Non-parametric tests were used to analyze odor ratings task data and odor discrimination task data, as they were not normally distributed. The Friedman test compared intensity and pleasantness ratings of body odors (Study 1B) or odorant compounds (Study 2), along with an odorless air condition. The Wilcoxon Signed Rank test compared each odor pairing against a chance proportion of 0.5 in the odor discrimination task. Bayesian Repeated Measures Analysis of Variance (RM-ANOVA) and Mann–Whitney U test were used to accumulate evidence for null hypotheses regarding differences between Receiver Groups (Western Caucasians, East Asians) in perceived body odor intensity, odor pleasantness, and odor

discrimination. Bayes factors (BFs) were calculated and interpreted based on established guidelines (Lee and Wagenmakers 2013).

Study 1B and 2: Outlier detection.

Outliers were detected using Median Absolute Deviation (MAD), a robust procedure for handling outliers (Leys et al. 2013). Outliers were winsorized, meaning their values were replaced with values 1 unit above (or below) the most extreme value that was not an outlier (Field 2013). This procedure helps maintain all data points while reducing the influence of outliers (Liao et al. 2017). As some morph task variables produced MADs of 0 in which case the MAD procedure does not work (it flags virtually all values ± the median as outliers), Z-values ± 1.96 were used instead of MADs to identify outliers. These outliers were also winsorized.

Results

Study 1A: Behavioral experiment (senders)

Before investigating the transfer of fear through body odor from different-genotyped senders to Western Caucasian and East Asian receivers, we first validated the effectiveness of the fear induction compared to a neutral condition in both sender groups: 18 Caucasian self-identified male G-allele carriers and 18 Asian self-identified male AA homozygotes. We used subjective ratings and physiological measures such as skin conductance and armpit sweat quantity to confirm effective fear induction in senders (Fig. 2). Importantly, there were no significant differences in fear experience between the different-genotyped sender groups. Detailed methods and statistical analyses related to this study can be found in the Supplementary Materials. With this validation completed, we then focused on examining whether the fear odors from different genotypes would modulate perceptual/behavioral and physiological responses in receivers.

Study 1B: Behavioral experiment (receivers)

Confirmatory analyses in receivers: Face morph task.

At the perceptual/behavioral level, our expectation was that receivers exposed to fear odor would perceive more fear than disgust in faces morphed between fear and disgust, as indicated by a higher percentage of fear responses on the face morph task. First, we expected that exposure to G-allele fear odor (compared to only G-allele neutral odor) would result in more face morphs being rated as "fearful" in both the Western Caucasian and East Asian Receiver Group (H1). Second, assuming universality, we expected the same effect for A-allele fear odor (compared to only A-allele neutral odor) (H2).

To test these pre-registered hypotheses, we conducted a 2 (Sender Emotion: Fear odor vs. Neutral odor) × 2 (Sender Genotype: A-allele vs. G-allele) × 6 (Morph Level: −15% vs. −9% vs. −3% vs. +3% vs. +9% vs. + 15% Fear) × 2 (Receiver Group: Asians vs. Caucasians) RM-ANOVA, including specific contrasts to directly test our hypotheses. However, the contrasts did not provide support for H1: $t(123.96) = -0.482$, $P = 0.631$, $d = 0.06$, 95% CI [−0.33,0.20], or H2: $t(123.96) = 0.272$, $P = 0.786$, $d = 0.04$, 95% CI [−0.24,0.31]. Splitting the data by Receiver Group did not change these conclusions (t-values < 1.12). For the full RM-ANOVA results, please refer to the Supplementary Table S3.

To test our third pre-registered hypothesis (H3), which stated that different-genotyped senders' fear odor would not affect fear responses differently in Western Caucasian and East Asian receivers, we employed a Bayesian RM-ANOVA. The results provided moderate evidence (close to anecdotal) for the null hypothesis ($BF_{01} = 3.48 ± 1.29\%$).

### Exploratory analyses: Face morph task and physiological responses.

Notably, the main effect of Sender Emotion from the regular RM ANOVA was not significant ($F < 1$). Contrary to our expectation, it was the G-Fear odor that was ineffective in inducing a fear bias, potentially washing out the main effect. To explore the possibility that the G-allele fear odor (G-Fear) might not be effective, while the "odorless" A-allele fear odor (A-Fear) could have an effect, we specifically examined the impact of A-Fear on the percentage of faces rated as fearful. To do this, we conducted a direct comparison between A-Fear and a combination of all other body odor conditions (G-Fear, A-Neutral, and G-Neutral) to determine its fear-biasing effect. Through an unplanned contrast analysis (adjusted using the Holm-Bonferroni method, as described in the Methods section), we found that A-Fear (compared to G-Fear, A-Neutral, and G-Neutral combined) significantly ($\alpha'_2 = 0.05$) increased the percentage of morphs rated as fearful, $t(177.88) = 2.18$, $P = 0.030$, $d = 0.28$, 95% CI [0.02,0.54], irrespective of Receiver Group and across all Morph Levels (Fig. 3A–C).

Furthermore, we explored the same hypothesis using a data-driven approach, specifically focusing on face morphs (FM) 2-4, which were *subjectively perceived* as the most ambiguous based on actual reaction time (RT) data (as detailed below). Analyzing only FM 2-4, the fear-biasing effect of A-Fear (compared to G-Fear, A-Neutral, and G-Neutral) was significant ($\alpha'_1 = 0.025$) with a slightly larger effect size, $t(485.42) = 2.32$, $P = 0.021$, $d = 0.35$, 95% CI [0.04,0.67]. In previous research, a similar fear-biasing effect was found following exposure to (Western Caucasian) fear odor (Zhou and Chen 2009; de Groot et al. 2021).

Another analysis was conducted on the receivers' RTs on the face morph task. The expectation was that rating more visually ambiguous morphs would result in longer RTs, as these stimuli were more difficult to classify as "fearful" or "disgusted." The analysis revealed a significant effect of Morph Level, $F(5, 325) = 14.35$, $MSE = 41037.25$, $P < 0.001$, $\eta_p^2 = 0.18$, 90% CI [0.11,0.23], indicating that participants responded slower to face morphs (FM) 2-4 (FM 2: $M = 993.34$ ms, SD = 277.81 ms; FM 3: $M = 1039.92$ ms, SD = 283.79 ms; FM 4: $M = 1000.00$ ms, SD = 271.02 ms) compared to the other morphs (FM 1: $M = 950.38$ ms, SD = 295.00 ms; FM 5: $M = 934.55$ ms, SD = 237.29 ms; FM 6: $M = 914.67$ ms, SD = 233.94 ms), $t(365) = 7.70$, $P < 0.001$, $d = 0.94$, 95% CI [0.65, 1.23]. Therefore, even though FM 3 (47% fear) and FM 4 (53% fear) may be considered the most ambiguous morphs based on *objective* standards, participants' RT data revealed a slightly different perspective: it was FM 2–4 (41%, 47%, 53% fear) that were *subjectively* perceived as the most ambiguous morphs. Expectedly, the most robust odor-biasing effects are found at the level of subjective ambiguity in perception.

At the physiological level, we explored the hypothesis that fear odor would induce higher skin conductance levels (SCLs)
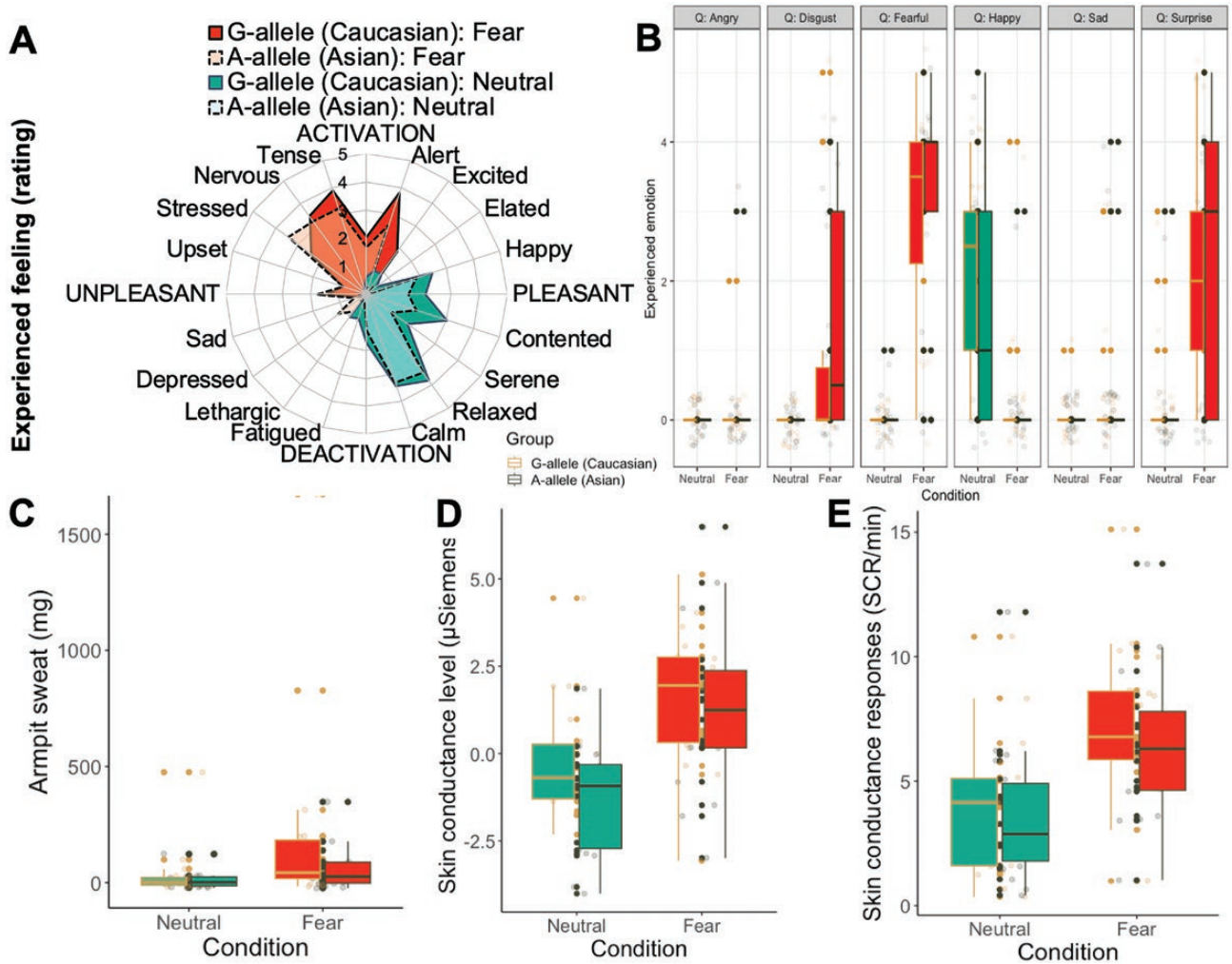
Fig. 2. Validation of effective fear induction in the different-genotyped senders. Note. These results from the body odor collection study (Study 1A) serve as a manipulation check for effective fear induction. The figure displays senders' subjective feelings and physiological responses in the fear-induction condition and neutral conditions, separated by genotype groups: G-allele-Caucasians (n = 18) and A-allele-Asians (n = 18). The panels display self-rated subjective feelings on an affective circumplex (A) and in discrete categories (B), as well as measures of armpit sweat quantity (C), general level of skin conductance minus baseline (D), and mean number of skin conductance responses per minute (E). Dots represent individual data points (jittered).

in both receiver groups. The 2 (Sender Emotion: fear odor vs. neutral odor) × 2 (Sender Genotype: A-allele vs. G-allele) × 2 (Receiver Group: Asians vs. Caucasians) RM-ANOVA indicated a significant main effect of Sender Emotion, $F(1, 57) = 12.44$, $MSE = 0.01$, $P < 0.001$, $d = 0.43$, 95% CI [0.16,0.71], but the effects were in the reverse direction than expected (fear odor: $M = 1.60$, $SD = 0.97$; neutral odor: $M = 1.61$, $SD = 0.97$) (Fig. 3D). The effect of Receiver Group was not significant, $F(1, 57) = 1.45$, $MSE = 3.73$, $P = 0.23$, $\eta_p^2 = 0.02$, 90% CI [0.00,0.12]. There were higher-order interactions, including a significant 3-way interaction: $F(1, 57) = 8.74$, $P = 0.005$, $\eta_p^2 = 0.13$, 90% CI [0.03,0.27], and a significant Sender Emotion × Sender Genotype interaction, $F(1, 57) = 9.16$, $MSE = 0.01$, $P = 0.004$, $\eta_p^2 = 0.14$, 90% CI [0.03,0.27]. Further exploration of the data showed that A-Fear resulted in significantly lower SCLs compared to the other 3 odor conditions ($M_{difference} = -0.05$ μS, $SD_{difference} = 0.14$ μS). However, the negative correlation between A-Fear SCLs and the percentage of faces rated as fearful on the morph task was not significant, $r(57) = -0.08$, $P = 0.562$. These findings suggest that caution should be exercised when drawing

conclusions about fear-specific responding based on SCLs in response to A-Fear.

Sensory tests.

On an explicit level, the 4 different types of body odor were rated equally intense and equally pleasant. They were indistinguishable from each other and differed perceptually from odorless air, which was included in the task to check for potential odor cross-contamination of the tubing, replicating de Groot et al. (2021) (Fig. 3E–G).

Specifically, a Friedman test on odor intensity revealed a significant effect of odor (5 levels: A-Fear, A-Neut, G-Fear, G-Neut, air), $\chi^2(4) = 23.19$, $P < 0.001$, $W = 0.44$. Odorless air was perceived as significantly weaker ($M = 10.16$, $SD = 11.35$) compared to the 4 odor stimuli, $t(136.38) = 13.34$, $P < 0.001$, $d = 1.63$, 95% CI [1.26, 2.00], which ranged from weak to moderately intense (A-Neut: $M = 12.25$, $SD = 10.38$; G-Fear: $M = 16.49$, $SD = 12.47$; A-Fear: $M = 14.00$, $SD = 11.61$; G-Neut: $M = 13.69$, $SD = 9.91$). Another analysis on odor pleasantness showed a significant main effect of odor, $\chi^2(4) = 10.05$, $P = 0.040$, $W = 0.44$. All 4 odors were more
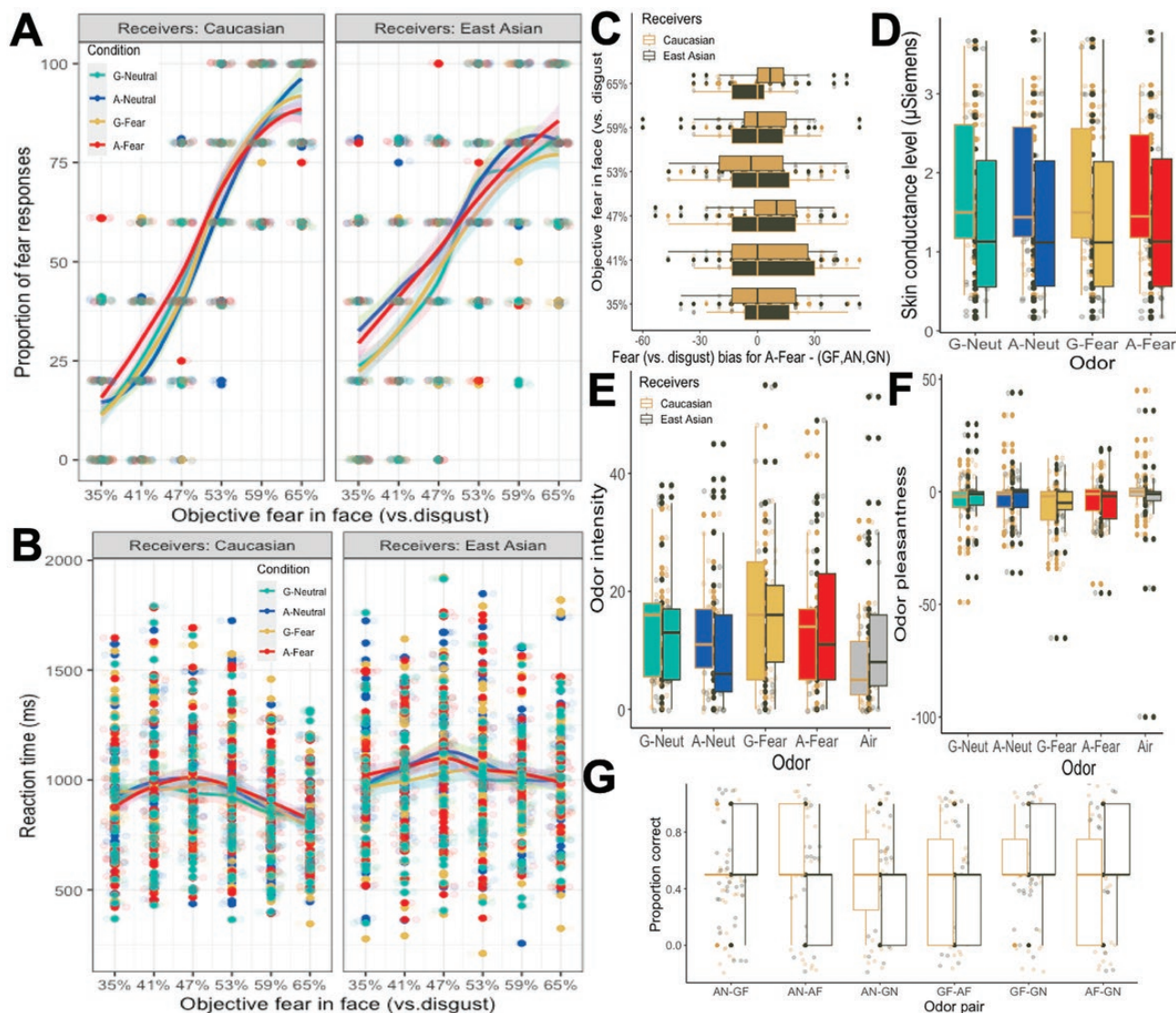
Fig. 3. Effects of different-genotyped senders' fear and neutral odor on receivers. Note. Results of the body odor exposure study (Study 1B). The graphs display the behavioral (A–C) and physiological (D) responses of Western Caucasian receivers ($n = 35$) and East Asian receivers ($n = 33$) after exposure to 4 body odors: fear-induced G-allele senders (G-Fear), fear-induced A-allele senders (A-Fear), neutral state-induced G-allele senders (G-Neut), and neutral state-induced A-allele senders (A-Neut), as well as receivers' performance on sensory tests (E-G). Panel A: percentage of fear responses at different morph levels for each odor condition split per receiver group. Panel B: reaction times split per receiver group. Panel C: relative fear (vs. disgust) responses after exposure to A-Fear minus the other odor conditions (G-Fear, A-Neutral, G-Neutral). Panel D: mean skin conductance levels per odor and receiver group. Panel E: body odor intensity ratings. Panel F: body odor pleasantness ratings. An odorless air control condition was included as a reference. Panel G: proportion of correct discriminations of body odor pairs (chance level = 0.5). Confidence interval bands in AB represent ± 1 SE. Dots represent individual data points (jittered).

disliked (A-Neut: $M = -2.12$, SD = 12.33; G-Fear: $M = -6.69$, SD = 12.52; A-Fear: $M = -4.43$, SD = 11.12; G-Neut ($M = -2.75$, SD = 12.57) compared to odorless air ($M = -1.53$, SE = 16.58), $t(143.31) = 3.53$, $P < 0.001$, $d = 0.43$, 95% CI [0.18,0.69]. When focusing on the 4 body odors, further Bayesian RM-ANOVAs indicated that both receiver groups (Caucasians vs. Asians) did not differ in their intensity (moderate evidence: $BF_{01} = 4.39 ± 0.77\%$) and pleasantness ratings (moderate evidence: $BF_{01} = 4.01 ± 1.64\%$).

Additionally, receivers were unable to significantly discriminate between pairs of body odor above chance levels. This was indicated by a non-parametric one-sample t-test (comparison A-Neut vs. G-Fear: $V = 280.5$, $P = 0.732$, $r_{rb} = 0.06$; A-Neut vs. A-Fear: $V = 351.5$, $P = 0.746$, $r_{rb} = 0.06$; A-Neut

vs. G-Neut: $V = 0.224$, $P = 0.598$, $r_{rb} = -0.10$; G-Fear vs. A-Fear: $V = 296$, $P = 0.511$, $r_{rb} = -0.11$; G-Fear vs. G-Neut: $V = 346.5$, $P = 0.079$, $r_{rb} = 0.31$; A-Fear vs. G-Neut: $V = 418$, $P = 0.253$, $r_{rb} = 0.25$). A Bayesian Mann–Whitney U test provided moderate evidence for the lack of difference in odor discrimination between the two receiver groups ($BF_{01} = 3.37$, $W = 549.5$).

Study 1C: Chemical analysis

We conducted an exploratory chemical analysis using gas chromatography-mass spectrometry (GC-MS; Fig. 4) to examine the 4 sample pools (G-Fear, A-Fear, G-Neut, A-Neut), established from 9 senders each. Due to the limited number of pools, these results are descriptive in nature. The goal of this
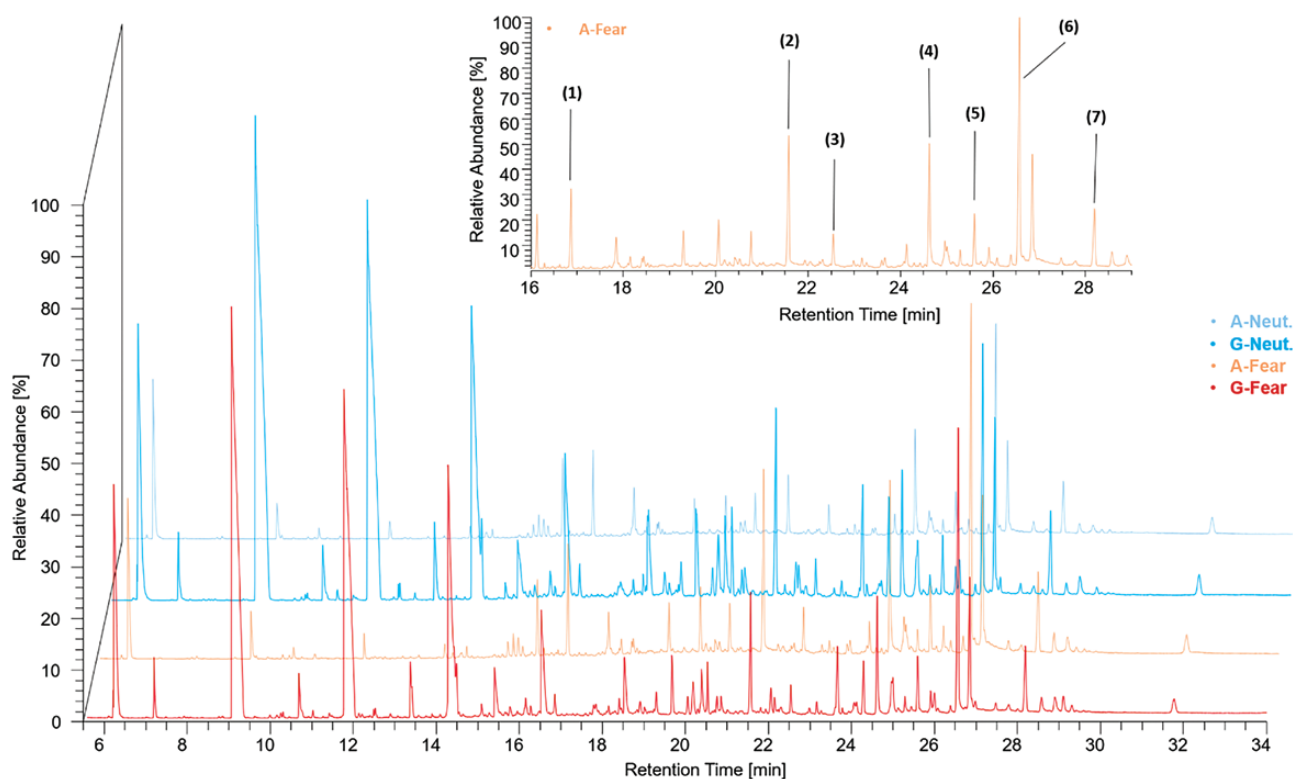
Fig. 4. Chemical analysis: chromatograms of different-genotyped fearful and neutral senders' sample pools. Note. The figure displays the Total-Ion-Count (TIC) chromatograms of the sample pools A-Neut, G-Neut, A-Fear, and G-Fear, obtained using one-dimensional gas chromatography-mass spectrometry using a DB-FFAP column. The bottom section shows the complete chromatograms, while the top right section zooms in on a retention time window between 16 and 29 minutes, where substances selected for quantification elute: dodecan-1-ol (1), hexadecan-1-ol (2), dodecanoic acid (3), tetradecanoic acid (4), pentadecanoic acid (5), hexadecanoic acid (6), and squalene (7). Note that sample pools from G-alleled senders were contaminated by siloxanes, pointing to a use of body care products by these senders either before or within the wash-out phase.

analysis was to assess any obvious qualitative and quantitative differences between the samples. We hypothesized that the chemical compositions would differ between fear and neutral sample pools.

We did not observe any visually detectable differences between the GC-MS profiles of fear and neutral sample pools (Fig. 4). The NIST 20 mass spectral library tentatively identified long-chain alcohols and acids, fatty acid esters, and squalene as the quantitatively most abundant volatiles of the sample pools (Supplementary Table S4). To estimate the concentrations of several quantitatively abundant compounds in the fear and neutral sample pools, we selected specific targets for analysis: dodecan-1-ol, hexadecan-1-ol, dodecanoic acid, tetradecanoic acid, pentadecanoic acid, hexadecanoic acid, and squalene (Table 1). The quantities of most compounds in the fear and neutral sample pools were within one standard deviation of each other, except for hexadecanoic acid, which showed a 44% higher concentration in the fear condition compared to the neutral condition across the different-genotyped samples.

Study 2: Testing individual compounds from sweat samples

Analyzing the percentage of faces rated as fearful using a 4 (odor) × 6 (morph levels) RM-ANOVA, we found a main effect of morph level, $F_{(5,160)} = 197.62$, $P < 0.001$, $\eta^2 = 0.67$. The effect of odor was at the threshold of significance, $F_{(3,96)} = 2.70$, $P = 0.05$, $\eta^2 = 0.003$, while the interaction

Table 1. Estimated concentrations of selected compounds in fear and neutral sample pooled extracts.

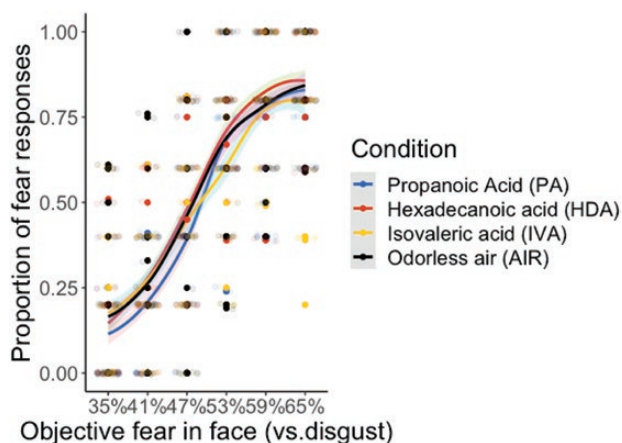| #	Target compound	RI [FFAP]	Fear c [µg/ml]	Neutral c [µg/ml]	Structure
1	Dodecan-1-ol	1937	37 ± 51	30 ± 28	
2	Hexadecan-1-ol	2350	101 ± 35	81 ± 75	
3	Dodecanoic acid	2244	42 ± 11	39 ± 7	
4	Tetradecanoic acid	2656	137 ± 35	114 ± 13	
5	Pentadecanoic acid	2762	87 ± 13	73 ± 17	
6	Hexadecanoic acid	2870	353 ± 58	245 ± 36	
7	Squalene	3056	52 ± 6	50 ± 15	

Average concentrations (±SD) were calculated separately for the Fear and Neutral emotion conditions based on the values obtained from the Asian and Caucasian sample pools. The concentrations were determined using external calibration with GC-MS (for more detailed information, refer to the Supplementary Materials). Concentrations (c) are reported in micrograms (µg) per milliliter (ml) of distillate, and the structures of the substances are provided in the rightmost column. RI = retention index on the DB-FFAP column.

between odor and morph level was not significant, $F < 1$. A contrast comparing HDA with the other conditions (PA, IVA, AIR)

showed a significant fear-biasing effect of HDA, $t(96) = 2.26$, $P = 0.026$, $d = 0.23$ (Fig. 5). In the opposite direction, PA differed significantly from all other odors, $t(96) = -2.13$, $P = 0.036$, leading to a lower number of faces rated as fearful. IVA did not significantly decrease the number of faces rated as fearful, $t < 1$. Analyzing the hypothesis that HDA biases perception, we conducted 2 separate Holm-Bonferroni corrected contrast analyses to investigate whether HDA affected objectively and subjectively most ambiguous face morphs. For the *objectively* most ambiguous morphs, specifically face morphs (FM) 3 and 4 (displaying 47% and 53% fear), our findings indicated that HDA did not significantly ($\alpha'_2 = 0.05$) increase the percentage of faces rated as fearful when compared to the other odors (PA, IVA, AIR), $t(459.06) = 1.95$, $P = 0.051$, $d = 0.34$. However, when we considered a broader range of *subjectively* perceived most ambiguous morphs, FM 2–5 (displaying 41%, 47%, 53%, 59% fear), HDA had a significant ($\alpha'_1 = 0.025$) impact, $t(195.08) = 2.38$, $P = 0.018$, with a moderate effect size, $d = 0.41$, leading to more faces being perceived as fearful. Like in Study 1B, this subjective ambiguity was supported by reaction time (RT) data, where participants had significantly slower RTs when judging FM 2–5 compared to FM 1 and 6, $t(160) = 3.72$, $P < 0.001$, while no significant RT difference emerged between FM 3–4 and FM 2 and 5, $t(160) = 1.75$, $P = 0.083$. Consequently, we concluded that FM 2–5 were subjectively perceived as the most ambiguous morphs, and the strongest odor-biasing effects were observed at the highest levels of subjective (rather than objective) ambiguity in perception.

Regarding odor intensity, a RM-ANOVA showed a significant main effect of odor, $F(3,93) = 9.01$, $P < 0.001$, $\eta^2 = 0.22$. A planned contrast confirmed that odorless air was perceived as less intense ($M = 10.59$, SD = 17.16) compared to HDA ($M = 19.62$, SD = 20.14), PA ($M = 19.22$, SD = 17.52), and IVA ($M = 17.72$, SD = 19.46), $t(93) = 5.10$, $P < 0.001$. A subsequent Bayesian RM-ANOVA provided moderate evidence for iso-pleasantness of HDA, PA, and IVA (BF$_{01}$ = 7.22 ± 1.1%).

Regarding odor pleasantness, there was no significant main effect of odor, $F < 1$. Odorless air did not differ in pleasantness

($M = 2.50$, SD = 12.66) from HDA ($M = 3.62$, SD = 22.17), PA ($M = -0.62$, SD = 20.35), and IVA ($M = 2.81$, SD = 20.29). A Bayesian RM-ANOVA provided moderate evidence for iso-pleasantness of HDA, PA, and IVA (BF$_{01}$ = 6.11 ± 0.5%).

Regarding odor discrimination, a non-parametric one sample t-test indicated that all odorants (HDA, PA, IVA) were significantly distinguishable from odorless air, indicating above 50% chance level discrimination: HDA vs. air ($V = 126$, $P = 0.008$, $r_{rb} = 0.65$); PA vs. air ($V = 126$, $P = 0.008$, $r_{rb} = 0.65$); IVA vs. air ($V = 104$, $P = 0.005$, $r_{rb} = 0.73$). Only HDA and PA were significantly distinguishable from each other, HDA vs. PA ($V = 90$, $P = 0.008$, $r_{rb} = 0.71$). However, HDA and PA could not be reliably distinguished from IVA: PA vs. IVA ($V = 54$, $P = 0.236$, $r_{rb} = -0.29$); HDA vs. IVA ($V = 121$, $P = 0.843$, $r_{rb} = 0.65$), arguably due to the low concentrations.

Discussion

The main objective of this research was to investigate the universality of chemical fear communication. In Study 1A, we first validated the effectiveness of fear induction in sender groups, comparing East Asian ABCC11 AA-allele carriers with Western Caucasian G-allele carriers, and found no significant differences in fear experience among these different-genotyped sender groups. Subsequently, in Study 1B, we examined how senders' fear odor influenced receivers' behavioral and physiological responses. While the original pre-registered hypotheses (H1 and H2) did not yield significant results when examining how fear odor from different-genotyped senders together affected fear responses in receivers, due to the unexpected absence of an effect for G-Fear (explained below), we conducted additional exploratory analyses for A-Fear. These exploratory findings suggest that A-Fear indeed induces a fear bias, highlighting a novel discovery that the presence of a crucial genetic factor—the single nucleotide polymorphism (SNP) 538G → A in the ABCC11 gene, which directly influences body odor—does not hinder the process of chemical fear communication.

Given the absence of any discernible effect from G-Fear odor, contrary to our pre-registered hypothesis that had combined the effects of G-Fear and A-Fear, we conducted exploratory analyses on A-Fear, which were corrected for multiple testing. These analyses aimed to further understand the nuances of the data beyond the pre-registered hypotheses. Specifically, we compared A-Fear to a composite of all other body odor conditions (G-Fear, A-Neutral, and G-Neutral) to assess its capacity to induce a fear bias. Notably, this analysis revealed a significant increase in the percentage of morphs perceived as fearful when exposed to A-Fear, and this effect persisted regardless of the Receiver Group, and occurred particularly for the face morphs subjectively perceived as the most ambiguous, as indicated by reaction time data. These exploratory findings suggest that the *production* of chemical fear cues remains effective despite the presence of a crucial genetic factor, the SNP 538G → A in the ABCC11 gene. This mutation is expressed as a weaker armpit odor and is dominant throughout East Asia (e.g. Martin et al. 2010). Apparently, the lack of body odor caused by the ABCC11 mutation, which spread rapidly across the Far East approximately 30–40,000 years ago due to positive sexual selection pressures (Natsch and Emter 2020), did not eradicate the

Fig. 5. Effects of odorants and controls on face perception. Note. The graph displays the average percentage of fear responses on the face morph task for each face morph level (ranging from 35% to 65% fear versus disgust) in different odor conditions: hexadecanoic acid (HDA), propanoic acid (PA), isovaleric acid (IVA), and odorless air (AIR). Confidence interval band ± 1 SE. Dots represent individual data points (jittered).

adaptive advantages of chemically communicating fear to receivers. These findings add to prior research showing that Caucasian fear odor affected both Caucasian and East Asian *receivers* in an emotion-congruent manner (de Groot et al. 2018). While the present study did not uncover evidence of a fear-biasing effect by the Caucasian G-Fear odor, previous research has extensively demonstrated chemical fear communication in Caucasian samples (meta-analysis: de Groot and Smeets 2017). In conjunction with the outcomes of this research, the collective body of literature leaves open the possibility of a universal mechanism for chemically communicating fear that is shared across the human species.

The lack of fear bias in the face morph task for G-Fear contrasts with abundant evidence in traditional WEIRD samples in which this "odorous" allele is dominant (e.g. Mujica-Parodi et al. 2009; de Groot and Smeets 2017; Maier et al. 2019; Smeets et al. 2020). In contrast to G-alleled individuals, who *do* have a functional ABCC11 membrane transport protein responsible for transporting odor precursors for thiol alcohols and fatty acids onto the skin, A-alleled individuals lack this functional ABCC11 membrane transport protein. Consequently, A-alleled senders have fewer precursors on their skin for the microbiome to transform into odor (Harker et al. 2014). The absence of an effect of G-alleled compared to A-alleled sweat might be explained by the possibility that in G-alleled senders, the microbiome—acting on precursors secreted onto the skin via functional ABCC11 membrane transport protein, thus generating thiol alcohols and fatty acids—partially masks the relevant fear signal that is produced independent of the non-functional A-allele. Another open question is whether odor degradation is partially due to the year-long storage of odor samples (at -30 °C) for Study 1B due to COVID-19 constraints. Importantly, in A-alleled individuals, there may be less microbially produced malodor to potentially mask the fear signal; yet, the exact workings of odorant precursors and bacterial metabolization in dynamically shaping fear odors have yet to be elucidated.

In Study 1C, we identified and semi-quantified a selection of (low) volatile compounds in pooled sample extracts. The exploratory chemical analysis revealed no obvious qualitative nor major quantitative differences between the volatile patterns of fear and neutral samples. It is important to note that the here applied sampling technique discriminates several constituents of axillary odor (Starkenmann 2017). Further analytical work is necessary to elucidate the chemical identity of the fear message, utilizing a larger sample size and advanced data analysis techniques, in conjunction with instruments capable of enhanced volatile separation and detection of odor-active trace compounds, such as comprehensive gas chromatography and gas chromatography-olfactometry, combining solvent and headspace extraction approaches. Peak areas do not necessarily correlate with the actual abundance in the sweat samples, and even quantitative occurrence does not necessarily correlate with bioactivity or smell contribution. Nonetheless quantitatively abundant substances are still a significant part of a sample and can play a contextual role in peripheral and central olfactory processes, which is why we studied them here.

In Study 2, we found evidence for a fear-biasing social communication effect of HDA on receivers compared to odorless air, isovaleric acid, and propanoic acid. It is still unclear what this means, because HDA has been detected in both samples

from the neutral and the fear condition. Previously, HDA has been described to be transferred between humans during handshakes (Frumin et al. 2015). A structurally related C16 compound, hexadecanal (HEX), has been demonstrated to affect human aggression in a dimorphic manner between sexes (Mishor et al. 2021) and to reduce startle reflexes in mainly male participants (Endevelt-Shapira et al. 2018). In mice, it has been shown that the OR37B glomerulus, activated by HEX, is to a significantly lower extent also activated by HDA (Bautze et al. 2012). It is conceivable that HDA and HEX are converted into each other by biotransformation in the nose, even if this has not yet been demonstrated experimentally (see Kornbausch et al. 2022 for a review on this topic).

Sensory tests performed in Study 1B and 2 showed moderate evidence for no difference in subjective odor pleasantness and intensity, and the presented odors could not be discriminated above chance level. The fear bias induced by A-allele fear odor and HDA cannot be explained by malodor alone, although malodor is closely associated with sweat. It appears that both fear odor and HDA have the ability to influence receivers' behavior without their awareness of the *source* of the odor.

The conclusions drawn from this study are limited to self-identified male senders and self-identified female receivers, the most widely used and effective dyad (meta-analysis: de Groot and Smeets 2017): males' larger apocrine sweat glands (Doty et al. 1978) boost fear odor production (Harker 2013), while females are slightly better smellers (meta-analysis: Sorokowski et al. 2019). We assume that the same underlying principles apply to self-identified female senders and self-identified male receivers, with smaller effect sizes, but this requires further investigation.

Another note of caution is that we did not fully complete all steps Wyatt mentions as part of the operational definition of pheromones (2020 Box 1), and therefore cannot assert the existence of a fear-omone. The fundamental basis for the designation of a pheromone is that the respective compound, or combination of compounds, should elicit the same response as the natural stimulus in the bioassay. In Study 2, we observed such a response for HDA. However, what we did not test is whether it acts in this way at natural concentrations. As Wyatt notes (2020), spurious results may occur at high concentrations, as non-pheromones may stimulate (non-olfactory) receptors. We made sure to present the compounds at low concentrations perceived to be equally intense. Also, experiments should demonstrate that *all* compounds in the combination are necessary and sufficient to elicit the full response, and that only this molecule (or combination) elicits the effect rather than other similar molecules. We have detected a much higher number of compounds in the sweat samples, and additional evidence is available from Smeets et al. (2020); HDA was additionally present in both neutral and fear body odor samples, and quantities in the extract were not significantly different. Finally, there should be a credible pathway for the pheromone signal to have evolved. Wyatt describes that to be a signal, both the emission and reception of the pheromone signal should have evolved for a particular function (2020). Fear odor has been found to offer clear advantages to recipients, aiding in threat detection and survival (Darwin 1872/1998; Susskind et al. 2008; de Groot et al. 2012). However, it remains unclear whether senders also benefit from fear odor. Future experiments could explore

whether fear odor triggers affiliative helping behavior or avoidance, as this distinction would determine the sender's potential benefits.

Despite its limitations, the current research achieved two objectives. We added evidence on the composition of fear and neutral body odor samples, and we showed, in a previously untested sample from a population exceeding a billion, that a unique genetic variant eliminating most body odor (e.g. Harker et al. 2014) did not impede the chemical communication of fear. Furthermore, we found that an individual compound present in sweat samples, hexadecanoic acid, replicated the fear-biasing effects in receivers. These findings appear to be consistent with recent research on universal principles in hedonic odor perception (Arshamian et al. 2022); when considered alongside the extensive body of literature on human chemical fear communication, primarily based on Western Caucasian samples (de Groot and Smeets 2017; Loos et al. 2023), our findings leave open a species-wide capacity to chemically communicate fear.

Supplementary material

Supplementary material can be found at http://www.chemse.oxfordjournals.org/

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Data availability

The materials and datasets generated during the study are available in the Open Science Framework repository (https://osf.io/c5wb2/).

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