Individual shedder status and the origin of touch DNA

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ABSTRACT

Due to improved laboratory techniques, touched surfaces and items are increasingly employed as sources of forensic DNA evidence. This has urged a need to better understand the mechanisms of DNA transfer between individuals. Shedder status (i.e. the propensity to leave DNA behind) has been identified as one major factor regulating DNA transfer. It is known that some individuals tend to shed more DNA than others, but the mechanisms behind shedder status are largely unknown. By comparing the amounts of DNA deposited from active hands (i.e. used “as usual”) and inactive hands (i.e. not allowed to touch anything), we show that some of the self-DNA deposited from hands is likely to have accumulated on hands from other parts of the body or previously handled items (active hands: 2.1 ± 2.7 ng, inactive hands: 0.83 ± 1.1 ng, paired t-test: p = 0.014, n = 27 pairs of hands). Further investigation showed that individual levels of deposited DNA are highly associated with the level of DNA accumulation on the skin of the face (Pearson’s correlation: r = 0.90, p < 0.00001 and Spearman’s ranked correlation: rₚ = 0.56, p = 0.0016, n = 29). We hypothesized that individual differences in sebum secretion levels could influence the amount of DNA accumulation in facial areas, but no such correlation was seen (Pearson’s correlation: r = −0.13, p = 0.66, n = 14). Neither was there any correlation between DNA levels on hands or forehead and the time since hand or face wash. We propose that the amount of self-DNA deposited from hands is highly influenced by the individual levels of accumulated facial DNA, and that cells/DNA is often transferred to hands by touching or rubbing one’s face.

1. Introduction

Handled items and surfaces are commonly encountered at crime scenes and are often used as sources of DNA evidence. Due to the development of improved laboratory techniques, it is possible to analyse traces containing just a few cells transferred by touching [1, 2]. This, in turn, leads to another challenge: how to determine if a minute DNA trace found at a crime scene is relevant for the police investigation. The term “shedder status” is an important factor in deciding whether it is likely that a collected DNA trace is the result of a direct contact or secondary transfer [3–5].

Shedder status refers to an individual’s propensity to leave behind genetic material on touched items and surfaces. Numerous studies report that some individuals tend to “shed” more DNA than others [3, 6–11]. While it has been proposed that there are “good” and “poor” shedders, i.e. individuals that consistently deposit more or less DNA [3, 11–13], there is also evidence that the quantity of deposited DNA varies from time to time for most persons [10, 14]. This suggests that although some factors appear to steadily influence the shedder status of a person over time, conditional factors such as behaviour, environment and activity also have an impact in a given situation. Biological factors affecting the quantity of deposited DNA are largely unknown, but there is coherent support that men in general shed more DNA than women [6, 10, 11, 15]. It is also reported that younger men shed more DNA than older men, but no such age-related trend has been found among women [14]. Certain activities, like wearing gloves or washing hands decrease the amount of deposited DNA [9, 11, 16], while touching another body part or one’s mobile phone appears to increase DNA deposit levels [10]. Altogether, the underlying mechanisms controlling DNA shedding, and the impact of genetic versus various non-genetic conditional factors, are not yet clear.

Initially, it was assumed that the DNA recovered from touched items
originated from epidermal cells, sloughed off from the outermost layer of the skin, and it has been suggested that individuals with a naturally high skin cell turnover are the “good” shedders [9,17]. More recently this view has been challenged, and it has been suggested that a major part of DNA transferred through touching comes from body fluids such as saliva, sweat and sebum [18–20]. The possibility that both nucleated cells and cell-free DNA can be transferred to the hands from other parts of the body or from previously handled items, has also been considered [10,21]. Following the study by Zoppis et al. [20], where it was shown that detection of touch DNA is dependent on previous DNA transfer from a sebum-rich area of the skin to the hand, a correlation between shedder status and individual sebum secretion levels has been proposed [21].

We examine if the shedder status of an individual at a given occasion can be coupled to the DNA levels on the facial skin, which would imply that a majority of deposited touch DNA from hands originates from touching the face or other parts of the body. We investigate this hypothesis by 1) comparing deposited DNA levels from “inactive” hands (i.e. thoroughly washed hands that did not touch anything prior to deposition) and “active” hands (i.e. thoroughly washed hands operating “as usual” prior to deposition), and 2) correlating deposited DNA amounts from the active hands of an individual and DNA quantities simultaneously collected from a sebum-rich area of the face. We also examine the relation between deposited DNA levels and individual facial sebum secretion levels.

2. Materials and methods

2.1. Experimental design

This study was approved by the Swedish Ethical Review Authority (approval no 2021-00643).

2.1.1. DNA deposition from active versus inactive hands

First, we investigated whether deposited DNA from hands were more likely to originate from shed cells/DNA from the hands themselves, or from extrinsic sources, i.e. from touching other body parts or previously handled objects that carry residual cells/DNA. The participants (n = 9) were assigned to attend a weekly one hour seminar, three weeks in a row. During the seminars, the participants were instructed to keep one of their hands “inactive”, meaning that this hand could not touch anything. The other “active” hand was allowed to operate “as usual”, i.e. to touch objects, handle phones, scratch the nose etc. Prior to the seminars, all participants were instructed to thoroughly wash both hands with soap and water for 15 s and let their hands air-dry. Which hand to keep active at each trial was assigned by the research leaders, ensuring that both dominant and non-dominant hands were active/inactive in the different trials. At one additional occasion, both hands of the participants were assigned to be active during a one-hour trial, operating as usual in the working environment (office and laboratory work) but with instructions not to wear gloves or wash hands again after the initial thorough hand wash.

After the 1 h trials (± 5 min), all participants deposited DNA by holding a plastic tube in each hand (see details in Section 2.2), a method that was recently evaluated by Johannessen et al. [13].

2.1.2. Correlation between DNA amounts from hands and face

To elucidate whether the levels of deposited DNA from the hands of an individual is dependent on the DNA levels from sebum-rich areas in the face of the same person, we conducted an investigation with 15 participants, including the nine persons from the first trial. At two different occasions (one week apart), the participants were asked to deposit DNA from both hands by holding plastic tubes, and at the same occasion also provide two DNA samples from their forehead (see details in Section 2.2). The participants were unaware of the exact date and time they would be asked to deposit DNA, thus the time passed since the last hand and face wash was not controlled and highly individual. This was done to mimic real-life situations where perpetrators touch objects with bare hands. After each occasion, the participants answered a questionnaire stating the time since the last hand wash (< 30 min, 30–60 min, 1–2 h or > 2 h), face wash (the same morning, the day before or ≥ 2 days ago), their recent major activities (e.g. office work, laboratory work, meeting) and whether they had been in direct physical contact with anyone during that day. Participant #15 participated at the first occasion only.

2.1.3. Sebum levels on face

Next, facial sebum levels of the participants were measured to investigate whether the intrinsic sebum level is an underlying factor of shedder status. A SebumScale (Delfin Technologies Oy, Kuopio, Finland) was used to assess the intrinsic level of sebum secretion of each participant (n = 14, same persons as in previous investigation except for participant #12 who did not participate). Their forehead sebum levels were measured at two separate occasions (around one week apart). At 1.5 h prior to each measurement, all participants were instructed to thoroughly wash their foreheads with soap and water for 15 s, then let their face air-dry. In a previous pilot study, we found that directly after face wash the sebum levels were very low (1–2 µg/cm², n = 2). During the subsequent 1.5 h, the participants were not allowed to touch their forehead or let anything (e.g. hats, caps or hair) come in contact with the area to be measured. This way, the measured sebum level at 1.5 h after face wash reflects the individual production rate of sebum, i.e. sebum secretion rate. In addition, the forehead sebum levels of the same persons were measured without the face wash prior to measurements, at two separate occasions (except participant #15 who did not participate, and #2 and #5 who were measured only once). The last experiment was designed to reflect the accumulated forehead sebum level of the participants, to account for individual habits such as frequency and thoroughness of face wash. All measurements took place during normal working hours and none of the participants had performed any exercise on the day of the sebum level measurements.

2.2. DNA sampling procedure

DNA depositions from hands were collected by instructing the participants to firmly hold a sterile 50 ml plastic tube (Sarstedt, Helsingborg, Sweden) in each hand for 30 s (Fig. 1a). Within 1 h, the deposited DNA was collected from the tubes applying a single cotton swab (Sefola Cotton swab, ref 120788, OneMed Sverige AB, Malmö, Sweden) moistened with 60 µL of 0.9% NaCl (Nordkrim, Helsingborg, Sweden) [22]. The tube walls were thoroughly swabbed by sweeping the upper tip of the cotton head in parallel movements while the swab was rotated slowly (Fig. 1b, also see [23]). The tube cap was not swabbed. Five negative process controls, i.e. plastic tubes that had not been touched, were swabbed and analyzed for the presence of any contaminating DNA.

DNA collection from faces was performed by applying a cotton swab (Sefola) directly onto the forehead. The swab was moistened with 100 µL of 0.9% NaCl prior to sampling, and two 2 cm × 2 cm squares on each side of the forehead defined by a template cut from a DNA free cloth (Fig. 1c) were swabbed applying one swab per square. Each square was swabbed by sweeping the tip of the swab applying medium-hard pressure across the defined area while rotating the swab slowly, covering the entire square twice. The swabbing procedure was performed by the same person throughout each experiment, protecting the samples from contaminating DNA by wearing lab coat, gloves, face mask and hair net, and carefully applying a standardized swabbing technique [23].

After DNA collection, the tip of the cotton swab was cut off just above the shaft, with a clean pair of scissors, and placed into a microtube tube and stored at −20 °C until further processing.

2.3. DNA extraction

The DNA extraction was performed using a Chelex-based method...

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DNA was retrieved from the tubes by thoroughly rubbing a cotton swab against the tube walls (excluding the cap) while slowly rotating the swab. c) DNA from the forehead was collected from two 2 cm × 2 cm squares on each side of the forehead, defined by a template cut from a DNA free cloth.

2.4. DNA quantification and STR analysis

DNA quantification was performed on a LightCycler Nano instrument (Roche Diagnostics, Basel, Switzerland) with the LightCycler Nano Software 1.1, applying the R1 qPCR assay [25] with 10X Immobuffer (Bioline Reagents, London, UK), 0.2 mM dNTP (Roche Diagnostics), 4 mM MgCl₂ (Roche Diagnostics), 0.3 μM of each primer (RBI_80F and RBI_235R, Life Technologies, New York, USA), 0.2 μM hydrolysis probe (Life Technologies), 2 μg BSA (Roche Diagnostics) and 1 U Immolase DNA polymerase (Bioline Reagents) in a total volume of 20 μL (10 μL sample). The qPCR protocol included DNA polymerase activation at 95 °C for 10 min, followed by 45 cycles at 95 °C for 10 s, 60 °C for 20 s and 72 °C for 30 s. For absolute quantification, 1:5 dilutions of 2800 M control DNA (Promega Corporation, Madison, WI, USA) (0.001 – 2 ng/μL, limit of quantification (LOQ) 0.001 ng/μL) were included. Short tandem repeat (STR) analysis was performed using PowerPlex ESX 16 Fast System (Promega Corporation), HID Veriti Thermal Cycler, ABI 3500 Genetic Analyzer (36 cm capillary array; POP4; injection parameters: 13 s/1.2 kV; analytical thresholds: blue channel 30 rfu, green 45 rfu, black (yellow) 70 rfu and red 75 rfu) and GeneMapper ID-X Software v1.3 (Thermo Fisher Scientific, Waltham, MA, USA). All samples from hand depositions were STR profiled, regardless of whether the DNA concentrations were below 0.001 ng/μL (LOQ). Out of in total 58 samples from foreheads, 23 samples were randomly selected for STR profiling, along with reference (buccal swab) samples from all participants to acquire their DNA profiles. No Template Controls (NTCs) were included in all PCR runs (quantification and STR analysis), to ensure the absence of DNA contamination during PCR preparation. The electropherograms were examined for donor (self DNA) and foreign (non-self DNA) STR alleles, respectively. All stutter alleles below the thresholds set by the manufacturer (Promega Corporation) were removed. Donor alleles were defined as all alleles corresponding to the STR DNA profile of the participant. The maximum number of successfully typed donor STR alleles is 30. An allele peak above the analytical threshold in a known homozygote locus was counted as two alleles. Up to three stochastically distributed foreign alleles in an electropherogram were viewed as drop-in events in accordance with Hansson and Gill [26], and were thus removed. Electropherograms with four or more unknown STR alleles were considered to contain foreign DNA. If the peak heights of the foreign STR alleles were less than 20% of the donor allele peak heights, then any alleles in donor allele stutter positions (i.e. exceeding the stutter filter) were removed. If the foreign STR allele peaks were higher than 20% of the donor allele peaks, remaining alleles in stutter positions were included in the foreign DNA profiles. In this way, stutter alleles were not allowed to contribute to neither an overestimation nor an underestimation of the amount of foreign DNA in the samples. Naturally, any foreign STR alleles overlapping with alleles from the donor could not be detected as foreign, potentially leading to a slight underestimation of foreign DNA.

2.5. Data analysis

DNA yields from hands and face are presented as total DNA amounts (ng per extracted sample) in scatter plots and bar chart with mean total DNA yields ± standard deviations. The total DNA yield, the percentage of donor DNA and number of successfully typed donor alleles for each sample are available in the supplement. The percentage of donor DNA versus foreign DNA for each sample was calculated as (TPHdonor + foreign alleles) × 100, where TPH is the total sum of STR allele peak heights. Differences in DNA deposition between active and inactive hands were tested statistically with a paired t-test (i.e. pair-wise comparisons of inactive and active hands), while differences between dominant and non-dominant hands were tested with independent two sample t-test assuming equal variances. One-way ANOVA followed by Tukeys post-hoc test was applied with R software [27] to examine statistical differences between the participants regarding deposition of foreign alleles from hands (active and inactive, respectively) and sebum secretion levels. All the data sets submitted to ANOVA met the criteria of approximately normal distributed data (evaluated by inspection of residuals histograms) and approximately equal variances (tested by applying Levene’s test in R package car [28]). Due to unequal variances, the data sets of total DNA yields from hands and foreheads were not subjected to ANOVA. Instead, participants that consistently (for all samples) deposited higher or lower DNA yields compared to the median yield of all samples were identified.

Amounts of foreign DNA were estimated by multiplying the total DNA yield with TPHforeign alleles/TPHdonor + foreign alleles. Pearson’s correlation coefficient (r) was determined to examine the strength of the linear relationship between 1) DNA yields on inactive versus active hands, 2) DNA yields from hands and face for each participant at a given occasion, 3) DNA yields from hands or face and time since last hand or face wash, and 4) sebum levels and DNA yields from hands or face for each participant. Prior to analysing the correlation between DNA amounts on hands or face and time since last hand or face wash, the indicated times in the questionnaire were converted as follows: < 30 min = 1, 30–60 min = 2, 1–2 h = 3, > 2 h = 4 and the same day = 1, 1 day ago = 2, ≥ 2 days ago = 3. Additional analysis with Spearman’s rank order correlation test was performed to confirm the association between DNA amounts from hands and face.
3. Results

3.1. DNA deposition from active versus inactive hands

By comparing the mean DNA yields deposited from active and inactive hands of nine individuals, it was clear that the active hands in general accumulated more DNA (2.1 ± 2.7 ng) than inactive hands (0.83 ± 1.1 ng, paired t-test \( p = 0.014 \), Fig. 2 and Table S1). A higher level of DNA was retrieved from the active hand in 23 of the 27 pairs of hands, but the differences between active and inactive hands varied considerably. In 16 pairs of hands, the DNA amount on the inactive hand was ≤ 50% of the DNA amount retrieved from the active hand (< 20% in 12 of these pairs). The inactive hands in the remaining seven pairs harboured between 51% and 87% of the DNA amounts found on the corresponding active hand. In four pairs of hands (participants #1, #2, #4 and #8), a higher amount of DNA (2–4 times more) was deposited from the inactive hand. STR profiling revealed that most deposited DNA originated from the donor for both active hands (96 ± 10%, \( n = 45 \)) and inactive hands (86 ± 22%, \( n = 27 \)). Four of the inactive hand samples gave 0 ng DNA in the quantification, while the STR profiling resulted in 6–8 detected donor alleles (Table S1). This is likely due to stochastic effects, where a multiplex PCR (STR analysis) has a higher chance of detecting low amounts of DNA compared to a single target assay such as the applied quantification system.

The amounts of deposited DNA differed substantially between individuals for active hands (ranging from 0.62 ± 0.56 ng (participant #9) to 4.7 ± 5.0 ng (participant #8)) as well as for inactive hands (ranging from 0.080 ± 0.14 ng (participant #6) to 2.5 ± 1.4 ng (participant #1)). The intra-individual variation was high and only two participants consistently deposited higher (#5) or lower (#7) levels of DNA from active hands (\( n = 5 \)) compared to the median DNA yield (1.3 ng) for all active hand deposits (\( n = 45 \)). Considering inactive hands, participants #1, #3 and #8 provided higher DNA yields (\( n = 3 \) per participant) than the median DNA yield for all inactive hands (0.41 ng, \( n = 27 \)), while #5, #6 and #9 gave lower DNA yields.

No correlation between DNA levels on active and inactive hands was found (\( r = -0.056, p = 0.78, n = 27 \)), implying that individuals with higher levels of DNA on their inactive hands were not consistently the same as those with higher DNA levels on their active hands. Further, no significant differences in DNA deposition between dominant and non-dominant active hands were seen (1.6 ± 1.1 ng and 2.6 ± 3.8 ng, \( p = 0.24 \)). None of the negative process controls (swabbed clean plastic tubes, \( n = 5 \)) carried any detectable DNA, and all NTCs were negative.

3.2. Correlation between DNA amounts from hands and face

Next, we show that there is a strong correlation between DNA amounts deposited from hands and DNA concomitantly collected from the face (Pearson’s correlation: \( r = 0.90, p < 0.00001 \) and Spearman’s ranked correlation: \( r_s = 0.56, p = 0.0016, n = 29 \), Fig. 3 and Table S2). In line with the results in Section 3.1, most of the deposited DNA from hands was from the donor (93 ± 14%). DNA profiling of randomly
selected face samples (n = 23) revealed that the DNA from the face in general consisted entirely of donor DNA (99.6 ± 1.2%). It was thus assumed that all DNA from foreheads originated from the donor. The mean deposited DNA amounts differed substantially between the participants, ranging from 0.11 ± 0.11 ng (participant #9) to 6.5 ± 4.2 ng (participant #1) for hands and from 0 ng (participant #12) to 67 ± 85 ng (participant #1) for faces, and the intra-individual variation was often high. Eight of the 15 participants consistently deposited higher (#1-3 and #8) or lower (#6, #9, #10 and #11) levels of DNA from hands compared to the median DNA yield for all hand deposits (0.44 ng). Similarly, participants #1-3 consistently provided higher DNA yields from face while participant #11-13 and #15 provided lower DNA yield compared to the median DNA yield of all face samples (2.9 ng). Although the correlation analysis points to a strong statistical relation between DNA amounts from hands and face, a few of the participants deviate from this pattern. Participants #4, #7 and #10 provided high DNA levels from face while participant #12 provided lower DNA amounts from face, although the DNA amounts from their hands deviate from this pattern. Participants #4, #7 and #10 provided higher DNA amounts compared to inactive hands.

No correlation was found between DNA levels on hands or forehead and the time since hand or face wash, respectively (hands: r = 0.0015, p = 0.99, and forehead: r = -0.18, p = 0.34, Table S3). Neither were there any indications that a specific activity prior to DNA deposition (i.e. office work, meeting, lab work etc) affected the DNA yield. None of the negative process controls (swabbed, clean plastic tubes, n = 5) carried any detectable DNA, and all NTCs were negative.

3.3. The prevalence of foreign DNA

Foreign STR alleles were found in 49% of the samples from active hands (n = 101) and in 37% of the samples from inactive hands (n = 27). However, in only a few samples the foreign DNA constituted more than 20% of the total DNA (10 out of 101 active hands and 6 out of 27 inactive hands). Even fewer samples harboured a majority of foreign DNA: only one sample from active hands (55% of foreign DNA, #1, Table S2) and three samples from inactive hands (56%, #4; 61%, #7; 70%, #9; Table S1). It should be noted that participants #4, #7 and #9 also had the lowest DNA deposits of all participants. However, no differences in the amounts of deposited foreign DNA were observed between the participants for neither active (F (8,56) = 0.99, F_{crit} = 2.2, p = 0.46) nor inactive hands (F (8,18) = 0.77, F_{crit} = 2.7, p = 0.63). Further, there was no correlation between the amounts of donor and foreign DNA on active hands in either trial (r = 0.016, p = 0.92 and r = 0.055, p = 0.78, respectively). This suggests that the prevalence of foreign DNA on hands is a stochastic phenomenon, dissociated from the propensity to deposit donor DNA or personal habits (hygiene routines etc.). Rather, foreign DNA appeared in samples from different individuals at random occasions. However, no apparent increase of foreign DNA was seen when the participants stated that they had been in direct physical contact with another person the same day (Table S3).

3.4. Association between sebum levels and amounts of deposited DNA

Sebum secretion levels (as measured 1.5 h after face wash) differed significantly between the participants (F (13,14) = 20, F_{crit} = 2.5, p < 0.0001), ranging from 4.5 ± 2.1–59 ± 3.5 µg/cm² (Fig. 4). The amounts of accumulated sebum (without face wash prior to the measurements) were higher than the secretion levels for all participants, except for #2. There was a strong correlation between the accumulated levels and sebum secretion levels (r = 0.82, p = 0.00068), indicating that the sebum level of the forehead at any time is highly dependent on the intrinsic sebum secretion level. However, there was no correlation between individual facial sebum secretion and the amount of shed DNA from hands (r = -0.026, p = 0.93, n = 14) or the DNA amounts collected from the face (r = -0.13, p = 0.66, n = 14).

4. Discussion

It is widely accepted that there are individual variations in shedder status, but whether this is due to physiological mechanisms, personal habits or other circumstantial factors is largely unknown [3,6-11,29]. Our results suggest that in many cases the majority of touch DNA is not derived endogenously from the hands. Rather, it seems to be transferred to the hands from elsewhere. This was demonstrated by comparing the DNA depositions from active hands (used “as usual”) and inactive hands (not allowed to touch anything) of participants who had washed their hands thoroughly one hour prior to DNA deposition. Higher amounts of DNA were retrieved from the active hand in 23 of 27 pairs of hands, where around 50% of the active hands had DNA levels that were more than five times higher compared to the corresponding inactive hands. In some pairs, the difference in DNA amounts were more modest, with less than the double amount of DNA on the active hands. In four pairs of hands from different participants, two to four times more DNA was found on the inactive hand. This may be explained by loss of accumulated DNA due to actions such as handling of objects. Despite this, there is statistical support for the finding that active hands in general harbour higher DNA amounts compared to inactive hands.
One reason for the higher DNA retrieval from active hands may be physiological differences between an inactive and active hand, such as a higher sweat production in an active hand. It has previously been shown that sweat may act as a vector for cell-free DNA, which has been shown to be present in touch DNA [19]. However, since the participants attended a seminar during the hour between hand wash and DNA deposition, it is not likely that a higher sweat production in active hands could explain the five times higher DNA amounts for around 50% of the active hands. Rather, these higher DNA amounts are more likely due to DNA accumulation on the active hands from exogenous sources. Much of the DNA may end up on the hands through touching other body parts (e.g., face, arms etc.) or personal objects that harbor residual self-DNA (e.g., cell phone and keyboard). This is in line with other studies, where it has been shown that unwashed hands carry higher amounts of DNA than washed hands [16], and that the amounts of deposited DNA increased when the participants had touched another body part or their mobile phones prior to deposition [10].

We saw great differences in the mean DNA deposition levels among the participants, supporting the existence of individual variations in shedder status. It was not consistently the same individuals who deposited high DNA levels from both their active and inactive hands. This is likely due to the different mechanisms for DNA accumulation on active and inactive hands. The DNA accumulating on active hands is dependent on both behavior (e.g., the frequency of scratching and touching other body parts or items), and physiological mechanisms. The DNA on inactive hands is bound to originate from the hands, and inter-individual levels may depend on physiological mechanisms such as keratinocyte turnover rate [9,17,30]. No correlation between DNA deposition and time since last hand wash was observed in our study, but higher DNA amounts are more likely due to DNA accumulation on active hands. Rather, these higher DNA amounts are more likely due to DNA accumulation on the active hands from exogenous sources. Much of the DNA may end up on the hands through touching other body parts (e.g., face, arms etc.) or personal objects that harbor residual self-DNA (e.g., cell phone and keyboard). This is in line with other studies, where it has been shown that unwashed hands carry higher amounts of DNA than washed hands [16], and that the amounts of deposited DNA increased when the participants had touched another body part or their mobile phones prior to deposition [10].

When collecting DNA from hands and face on the same occasion, a strong correlation in amounts of DNA was found. This implies that the amount of DNA on hands often is dependent on the amount of DNA on the skin of the face. The fact that the time since last face wash and facial DNA levels were uncorrelated suggests that mainly physiological factors affect the abundance of DNA on facial skin. The transfer of DNA from face to hands may depend on personal habits such as the frequency of touching or scratching facial parts. It has been reported that people touch their face on average 50 times per hour [34]. Thus, even if some individuals tend to touch their face less than others, it seems that independently of such differences a person with a high release of DNA from the face also is more likely to deposit high amounts from the hands. However, there will always be individuals that deviate from this pattern, as observed here.

It is possible that the physiological mechanisms behind DNA release from the face are different from those behind accumulation of endogenous DNA originating from hands. For example, inter-individual differences in sebum secretion levels would elegantly explain the different levels of accumulated DNA in the face and thus individual shedder status. Sebum is a lubricating substance consisting of cellular debris from fully mature sebocytes that is produced in the sebaceous glands in the skin, abundant in the face of humans. Sebum production is under hormonal control, and especially androgens in males stimulate the proliferation of sebocytes in facial sebaceous glands [35]. In agreement, men have been shown to secrete significantly higher levels of facial sebum than women [36], which would be a plausible biological basis behind the higher levels of deposited DNA by men. However, measuring facial sebum secretion levels of the participants revealed that there was no correlation between the sebum secretion level and the accumulation of DNA on the forehead or on the hands. Individual habits such as frequency and thoroughness of face washing or showering had no significant impact on the facial sebum levels, as the accumulated levels of sebum (over time) on the forehead skin were proportional to the sebum secretion levels (measured 1.5 h after a thorough face wash). In all, this suggests that the individual sebum secretion level is not a major factor behind shedder status.

Other body fluids like sweat, saliva, urine, nasal and eye fluids have also been suggested as potential vectors for touch DNA [21]. While very low amounts of DNA-containing cells have been detected in nasal lavage and eye fluids [31], saliva is indeed a potent vector for transfer of DNA [18]. Sweat has been shown to constitute a major component of touch DNA [38], and may be a source of accumulated DNA that is dependent on physiological traits of an individual. The function of sweat glands differs between individuals and is known to cause a great variation in eccrine secretory capacity among individuals [39]. Sweat gland activity is also regulated by hormones [40,41], and men have been shown to begin sweating sooner and in higher volume with activity or heat than women [42]. Thus, it is plausible that an individual with a higher sweating rate accumulates more DNA on the skin which is subsequently transferred to the hands by touching other body parts.

Possibly, several physiological factors contribute to the accumulation of DNA on the skin with unequal strengths for different individuals. An increased keratinocyte turnover may be the major factor behind DNA shedding in individuals with "dry" hands or skin diseases [43,44], while for others a high sebum or sweat secretion may give rise to more DNA on the skin. If this is the case, the search for a correlation between DNA shedding and a single factor controlling this ability is bound to be
5. Conclusions

Our results suggest that in many cases, the majority of DNA deposited on items and surfaces does not originate from the hands themselves, but may have been transferred to the hands by touching, rubbing or scratching other body parts or handling personal objects. The individual levels of deposited DNA are highly associated with the level of DNA accumulation on the skin of the face, but no correlation between amounts of deposited DNA and facial sebum secretion was seen. Nevertheless, the strong association to facial DNA accumulation sug-

gests that physiological mechanisms rather than differences in personal habits dictate the individual shedder status.

CRediT authorship contribution statement


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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.fsigen.2021.102626.

References