Journal of Exposure Science & Environmental Epidemiology www.nature.com/jes

INVITED REVIEW

Current knowledge of the degradation products of tattoo pigments by sunlight, laser irradiation and metabolism: a systematic review

Tristan R. Fraser[1], Kirstin E. Ross[1], Ula Alexander[1] and Claire E. Lenehan[1]✉

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The popularity of tattooing has increased significantly over recent years. This has raised concerns about the safety of tattoo inks and their metabolites/degradation products. The photolytic and metabolic degradation of tattoo pigments may result in the formation of toxic compounds, with unforeseen health risks. A systematic literature review was undertaken to determine the current state of knowledge of tattoo pigments' degradation products when irradiated with sunlight, laser light or metabolised. The review demonstrates that there is a lack of knowledge regarding tattoo pigment degradation/metabolism, with only eleven articles found pertaining to the photolysis of tattoo pigments and two articles on the metabolism of tattoo pigments. The limited research indicates that the photolysis of tattoo pigments could result in many toxic degradation products, including hydrogen cyanide and carcinogenic aromatic amines.

Journal of Exposure Science & Environmental Epidemiology (2022) 32:343–355; https://doi.org/10.1038/s41370-021-00364-y

INTRODUCTION

Tattoos are a form of body modification where inks and pigments are inserted into the dermis layer of the skin as a form of self-expression or group affiliation (i.e. tribal and gang tattoos) [1]. The practice of tattooing has existed since ancient times, as evidenced by the 61 tattoos found on the 5300-year-old ice mummy, Ötzi [2]. Traditionally, tattoo inks and pigments were manually deposited into the dermis with the use of a needle or similar object. However, modern tattoo application typically uses a tattoo gun, which allows for the ink to be deposited at more consistent depths and concentrations.

Recently, there has been an increase in the number of people getting tattoos. In a survey undertaken in Australia, it was reported that the number of tattooed individuals rose from 10.1% in 1998 to 14.5% in 2005 and to 25% in 2019, with younger generations driving this increase [3–5]. A similar trend was observed in the United States. In 2006, Laumann and Derick [6] reported that ~24% of the US population aged between 18 and 50 years have at least one tattoo. More recently, in 2019, Kluger and co-workers found that 31.5% of US respondents had at least one tattoo [7]. These two US studies also observed increased numbers of tattoos amongst younger people. In the study by Laumann and Derick [6], 15% of the respondents that were born between 1953 and 1963 reported having a tattoo, which increased to 24% of respondents born between 1964 and 1974, and to 36% of respondents born between 1975 and 1986. Furthermore, the study by Kluger et al. [7] found that 40.2% of 18–24-year-old respondents had at least one tattoo.

Tattoo inks are complex mixtures that may contain multiple pigments, a carrier, preservatives, dispersants and formulants. In addition, the inks often contain by-products or precursors from the production of the pigments, many of which were originally designed for other industries [8]. The pigments can consist of inorganic molecules, such as titanium dioxide, in the case of white inks and many coloured inks as a tinting agent, organometallic complexes, such as phthalocyanines in the case of blue and green inks, or organic molecules, such as azo compounds in the case of yellow-, red- and orange-coloured inks [1, 9], examples of which are given in Fig. 1. As these pigments tend to have a low solubility in the carrier, which is often either water, glycerol, alcohols, witch hazel or some combination thereof, dispersants are often required to avoid aggregation [9]. In addition, the concentration of tattoo pigment that is deposited in the skin can vary significantly, with reported ranges between 0.60 and 9.42 mg/cm^2 [10].

Earlier health concerns regarding tattoos focused on hygiene and the potential for spreading disease. However, increasingly, concerns have been raised about the safety and long-term health effects of tattoo inks and their components as indicated by the drafting of tattoo ink guidelines [11, 12]. Initially, health concerns regarding ink composition focused on metals such as arsenic, lead and mercury that were originally used as pigments [1, 9]. In a study undertaken by the New Zealand Ministry of Health, nine metals were found to be greater than the United States Environmental Protection Agency's recommended concentrations in as many as 24% of the samples tested, though, with the exception of barium and copper, most metals tested were likely impurities. Only cobalt, selenium and chromium VI were found at, or below, the recommended concentration [12]. While inorganic pigments are effective as colourants, in recent years, there has been a demand for colourants with more vibrant colours.

[1]College of Science and Engineering, Flinders University, Bedford Park, SA, Australia. ✉email: Claire.lenehan@flinders.edu.au

Received: 22 October 2020 Revised: 27 June 2021 Accepted: 29 June 2021
Published online: 17 July 2021

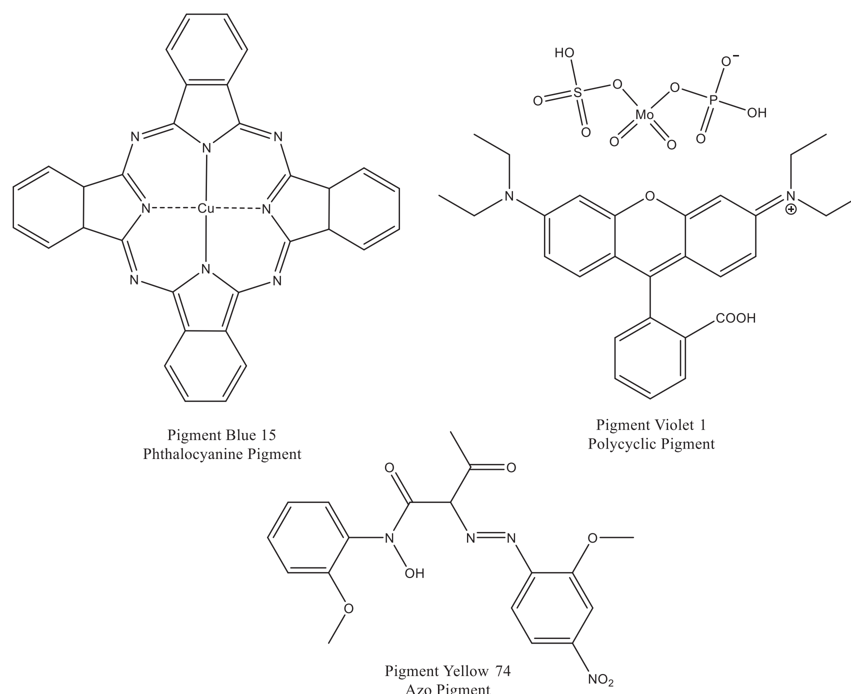

Fig. 1 Example structures of the three primary pigment types. Chemical Structures of Pigment Blue 15, Pigment Violet 1 and Pigment Yellow 74.

Consequently, organic pigments are now used in conjunction with, or replaced, the more traditional inorganic colourants [9]. This has raised health concerns about the organic colourants that are now in greater use. This is demonstrated by the publication of the ResAP (2008)1 Report (Resolution on Requirements and Criteria for the Safety of Tattoos and Permanent Make-up), which lists recommendations of tattoo pigments and impurities that should be absent from tattoo inks due to their carcinogenicity, sensitising properties and mutagenicity [11]. This report became the basis of regulatory legislations around Europe prior to the REACH regulations being published in 2020.

Of those individuals that get a tattoo, roughly a quarter, or 14–26% depending on the demographic, lament their decision [12, 13]. Many choose to undergo tattoo removal. The tattoo removal process can be difficult and costly and often requires multiple sessions. Older methods of tattoo removal include abrasion of the skin (salabrasion or dermabrasion), surgical excision and thermal removal. However, these methods generally leave scars and do not guarantee complete removal of the tattoo [14, 15]. Removal of tattoos is now commonly undertaken using lasers that heat the tattoo pigments with short pulses of light. The laser conditions used depend on the pigments' absorption [14]. The short pulses heat the pigment at a rate greater than the thermal relaxation time of the pigment. This causes thermal degradation of the molecule and heats the tissue in a short range around the pigment. The expansion resulting from heating the surrounding tissue can cause a negative pressure and, consequentially, a shockwave that mechanically destroys the pigment agglomerates, resulting in smaller particles that can undergo phagocytosis [16]. Thus, tattoo laser removal may be achieved by photothermal and photomechanical process. While laser tattoo removal is much more effective than the old methods for tattoo removal and reduces the risk of scarring, many sessions may be required, and complete tattoo removal is still not guaranteed. Furthermore, should the tattoo removal or post-treatment be poorly executed, patients can experience a darkening of the tattoo pigments, scarring or allergic reactions [14, 15].

As a first step to determine the potential health impacts of these degradation products, a systematic review of the current literature was undertaken. This review highlights the gaps in our current understanding, which, once researched and understood, would aid governments and regulators to create informed guidelines and legislation to govern the use and production of tattoo inks and guide the removal processes of tattoos.

Methodology

The databases Medline, Scopus, Web of Science and Scifinder were searched for articles written in English with the keywords; 'Tattoo* AND Dye* OR Pigment* AND Photo* OR Laser* OR Light* OR Tox*' and 'Tattoo* AND Dye* OR Pigment* AND Metabol*' (Fig. 2). As Scifinder uses a 'natural language' search structure, each combination of search terms from the three terminology groups seen in Table 1 were used to search Scifinder. Thus, the first search used the phrase 'Photolysis of Tattoo Pigments', the second used the phrase 'Photolysis of Tattooing Pigments', etc.

The systematic search excluded patents and included all articles published up to July 2020. Articles that were not written in English or German, and were not peer-reviewed articles, were excluded. English and German articles were selected as English is the authors native language and German to English translation was obtainable. A further three could not be accessed using the document delivery service and were also eliminated. Once articles had been excluded according to these criteria, the titles and abstracts of articles were manually reviewed to ensure that they reported either the photolysis, or the metabolism of tattoo pigments. The remaining articles were then read in detail and, for tattoo pigment photolysis, only those that expressly reported the use of a laser, simulated sunlight/UV light or natural sunlight for the photolytic degradation of permanent tattoo pigments or inks, and those that reported the degradation products observed, were included in the systematic review. For the metabolism of tattoo pigments, only those that reported the use of human cell cultures, human subcellular fractions or human enzymes for the metabolism of permanent tattoo pigments and reported that the metabolic products were included in the systematic review. Once

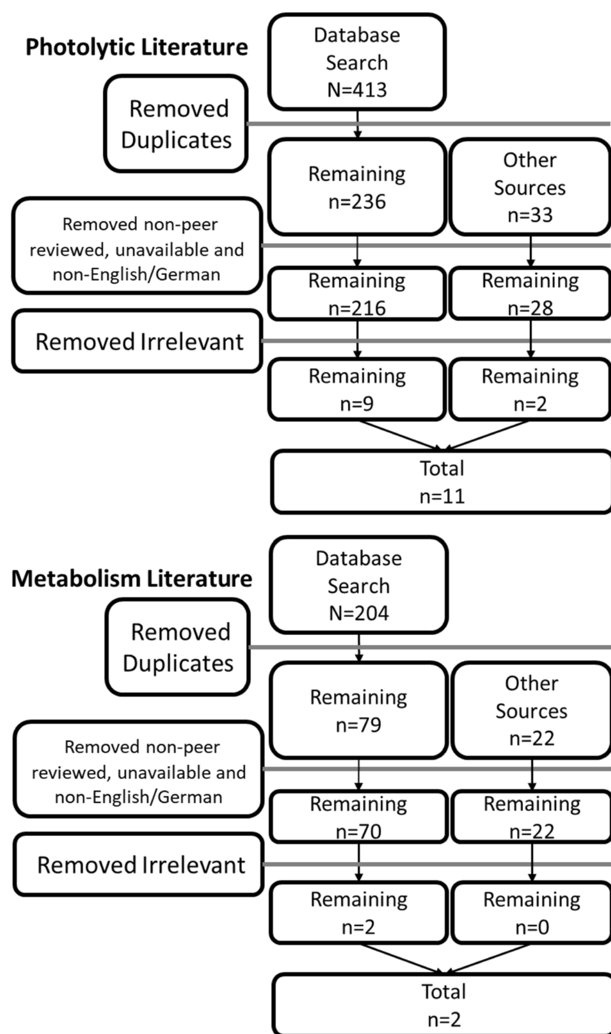

Fig. 2 The literature search protocol. Flow chart of the exclusion processes used to refine the literature search.

the databases had been searched and the results screened, the reference lists of the articles that were included from the search were screened in the same manner as described above. The number of articles that were found in the search, in references, and the number that passed each screening step is presented in Fig. 2. Overall a total of 11 articles, reviews and theses were found concerning the photolysis of tattoo pigments and 2 articles were found pertaining to tattoo pigment metabolism (see Table 2).

RESULTS

Photolysis

As noted above, the search of the databases Medline, Scopus, Web of Science and Scifinder for literature on the photolysis of pigments used in tattoo inks yielded 11 articles of relevance (see Table 2). A total of 24 pigments (see Table 3 for pigment list) were mentioned or examined in these articles. However, 13 pigments are mentioned in only two or less of the 11 articles while Pigment Orange 13 and Pigment Red 22 (PR 22) are mentioned in 5 articles and Pigment Yellow 74 (PY 74) is mentioned in 7 articles. Furthermore, of the 12 pigments that are mentioned in only two of the 11 articles, 7 of them are mentioned once in a review article and once in a research article. Thus, the focus of research on the photolytic degradation of tattoo pigments has been directed at a limited selection of pigments.

Table 1. List of search terms used in the systematic literature review for pigment photolysis and metabolism.

Terminology group 1	Terminology group 2	Terminology group 3
Tattooing	Pigment	Photolysis
Tattoo	Dye	Photolytic degradation
–	–	Photodegradation
–	–	Photolytic decomposition
–	–	Photodecomposition
–	–	Photostability
–	–	Photolytic stability
–	–	Light stability
–	–	Laser irradiation
–	–	Toxicology
–	–	Toxicity
–	–	Metabolism
–	–	Metabolite
–	–	Metabolomics

Terms within a terminology group were separated by OR and whole terminology groups were separated by AND.

The photolysis of pigments by some form of laser is reported in seven of the articles and five of the articles report the photolysis of pigments by natural or simulated solar light. Of these, three articles solely reported the photolysis of pigments under natural or simulated solar light [1, 17, 18]. The Q-switched Nd:YAG laser was the most commonly used laser for experimental photolytic cleavage of tattoo pigments by laser light, although, the Q-switched ruby laser is used alongside the Q-switched Nd:YAG laser [19–21]. Typically, fluences of 3 J/cm^2 up to 5 J/cm^2, a pulse duration of <20 ns and spot sizes of 1–4 mm were used when irradiating tattoo pigments (see Table 4). These laser settings seem to fall within the range of settings used by dermatologists whom tend to start the removal process with a fluence of ~4.2 J/cm^2 and alter it as needed without dropping below 2.9 J/cm^2 or exceeding 9.4 J/cm^2 [22]. As for simulated solar light, Cui et al. [1] used a 6.5-kW xenon-arc lamp in conjunction with a WG320 glass filter to ensure that the light spectrum that the samples were exposed to was consistent with natural solar light [1, 23]. Similarly, a broad band UV lamp operating between 280 and 320 nm and an Atlas Suntest CPS+ Sun simulator operating between 290 and 800 nm have been used to simulate solar irradiation [17, 24]. Conversely, it seems more common for samples to be left near a window or in an area where they will be exposed to natural solar light for several days in order to examine the photolysis of pigments by natural solar irradiation (Table 4 presents a summary of irradiation methods).

A variety of chemical analysis techniques for analysis of the tattoo pigments was reported in the articles concerning tattoo pigment photolysis. The most common analytical technique used was high-performance liquid chromatography (HPLC), which was then commonly coupled with mass spectrometry (MS), conversely, some of the articles used gas chromatography (GC) coupled with MS. Of the six articles that used HPLC systems, two used ultra-high-performance liquid chromatography (UHPLC) [18, 24]. Interestingly, Gaugler [18] used both UHPLC and GCMS to analyse their samples in addition to a HPTLC system coupled with a bioluminescence screening method that used Vibrio fischeri in a novel fashion to assess the toxicity of their samples. Results from pyrolysis–GCMS analysis of unirradiated pigment samples were compared to the GCMS analysis of laser irradiated pigment samples to determine whether pyrolysis could simulate the

Table 2. List of articles found in the systematic database search and in reference lists.

Authors	Year	Title	Reference
Photolysis literature found in the systematic search of databases			
Schreiver I, Hutzler C, Laux P, Berlien H-P, Luch A	2015	Formation of highly toxic hydrogen cyanide upon ruby laser irradiation of the tattoo pigment phthalocyanine blue	[19]
Hering H, Sung AY, Roder N, Hutzler C, Berlien H-P, Laux P	2018	Laser irradiation of organic tattoo pigments releases carcinogens with 3, 3′-dichlorobenzidine inducing DNA strand breaks in human skin cells	[20]
Engel E, Spannberger A, Vasold R, König B, Landthaler M, Bäumler W	2007	Photochemical cleavage of a tattoo pigment by UVB radiation or natural sunlight	[17]
Cui Y, Spann AP, Couch LH, Gopee NV, Evans FE, Churchwell MI, et al.	2004	Photodecomposition of pigment yellow 74, a pigment used in tattoo inks	[1]
Hauri U, Hohl C	2015	Photostability and breakdown products of pigments currently used in tattoo inks	[26]
Vasold R, Naarmann N, Ulrich H, Fischer D, Könlg B, Landthaler M, et al.	2004	Tattoo pigments cleaved by laser light—the chemical analysis in vitro provide evidence for hazardous compounds	[28]
Schreiver I	2018	Tattoo pigments: biodistribution and toxicity of corresponding laser-induced decomposition products	[21]
Engel E, Vasold R, Santarelli F, Maisch T, Gopee NV, Howard PC, et al.	2010	Tattooing of skin results in transportation and light-induced decomposition of tattoo pigments—a first quantification in vivo using a mouse model	[67]
Bauer EM, Scibetta EV, Cecchetti D, Piccirillo S, Antonaroli S, Sennato S, et al.	2020	Treatments of a phthalocyanine-based green ink for tattoo removal purposes: generation of toxic fragments and potentially harmful morphologies	[27]
Photolysis literature found in in reference lists			
Gaugler S	2011	Analysis of bioactive compounds in tattoo inks before and after irradiation with sunlight using HPTLC and in situ detection with *Vibrio fischeri*	[18]
Wezel K	2013	Examination of the behaviour of tattoo inks and pigments under the influence of light	[24]
Metabolism literature found in the systematic search of databases			
Chen H	2006	Recent advances in azo dye degrading enzyme research	[70]
Cui Y, Churchwell MI, Couch LH, Doerge DR, Howard PC	2005	Metabolism of pigment yellow 74 by rat and human microsomal proteins	[25]

degradation resulting from laser irradiation of pigments [19]. Pyrolysis–GCMS was determined to be a suitable predictive method for analysing the possible degradation products resulting from laser irradiation [19]. As previously mentioned, PO 13, PY 74 and PR 22 were the most researched pigments in the literature. All three were azo-based pigments, in fact, 12 of the pigments examined in the photolysis literature are azo pigments, possibly indicating a greater concern about the potential dangers of the degradation of these azo compounds. These concerns seem justified as commonly observed photolytic degradation products include benzonitrile, aniline, benzene and 3,3′-dichlorobenzidine, which are all produced by azo pigments (see Table 5). Benzonitrile was found to be a degradation product from all types of pigments. Aniline was found to be a degradation product from azo and polycyclic pigments and benzene was found to be a degradation product from azo and phthalocyanine pigments. 3,3′-dichlorobenzidine, however, was observed to be produced only from the photolysis of azo pigments. A total of 51 photolysis products were observed in the photolytic degradation of tattoo pigments though, as seen in Table 5, many of these photolytic degradation products are observed to only occur in the degradation of one or two different tattoo pigments. Thus, the potential toxicity of a pigment resulting from laser-induced degradation cannot be estimated from pigments of a similar class as the degradation products appear to vary greatly.

Metabolism

There was little research concerning the metabolism of tattoo pigments by the human body. Only two articles were found in the literature search that investigated the metabolism of tattoo pigments by the human body, one being a research article by Cui et al. [25] examining PY 74 [25], while the second was a review article referencing the research by Cui et al. [25]. The metabolism of other pigments has not been reported. Cui et al. [1] found that PY 74 was metabolised by the cytochrome P450 (CYP450) enzymes, CYP1A1 and CYP1A2. They also reported that the enzymes CYP1B1 and CYP3A4 also metabolised PY 74, but that they did not metabolise as much of the PY 74 as the other two, indicating a lower efficiency. Furthermore, it was found that PY 74 was metabolised in two steps. First, PY 74 is hydroxylated and subsequently it is o-demethylated as presented in Fig. 3.

Microsomes from male F344 rats that had been pre-treated with 3-methylcholanthrene, human liver microsomes, male Sprague-Dawley rat liver microsomes and Human Supersomes® consisting of the cytochrome P450 enzymes 1A1, 1A2, 1B1, 2B6, 2C9, 2D6, 2E1 and 3A4 were used to investigate the metabolism of PY 74 [25]. The metabolites of PY 74 were identified using a HPLC system in conjunction with a photodiode array detector. Subsequently, the structures of the metabolites were identified and confirmed with a 500-MHZ NMR and by coupling the HPLC system with a mass spectrometer [25].

DISCUSSION

Mechanisms of pigment degradation

Tattoo pigment degradation through sunlight, laser irradiation and metabolism, can result in the production of smaller, more toxic molecules, whose composition is dependent on the degradation mechanisms. Whilst sunlight and laser irradiation both use photo-activated mechanisms to cause tattoo degradation, the intensity and duration of the irradiation is different and likely to result in differing compositions and concentrations of the

Table 3. Azo, polycyclic and phthalocyanine pigments—identified using their common pigment names, CAS and colour index (C.I.) numbers—mentioned or analysed in photolysis and metabolism literature.

Pigment type	Pigment name	CAS number	Colour index number	Mentions in literature	References
Azo	Pigment Orange 13	3520-72-7	21110	5	[18, 20, 21, 24, 26]
Azo	Pigment Red 22[a]	6448-95-9	12315	4	[17, 26, 28, 67, 70]
Azo	Pigment Yellow 74[a]	6358-31-2	11741	5	[1, 18, 21, 24–26, 70]
Azo	Pigment Orange 16	6505-28-8	21160	3	[18, 24, 26]
Azo	Pigment Yellow 14[a]	5468-75-7	21095	3	[18, 24, 26, 70]
Azo	Pigment Orange 34	15793-73-4	21115	3	[18, 24, 26]
Azo	Pigment Red 170	2786-76-7	12475	2	[21, 24]
Azo	Pigment Red 9	6410-38-4	12460	2	[1, 26]
Azo	Pigment Yellow 83	5567-15-7	21108	2	[18, 26]
Azo	Pigment Orange 5	3468-63-1	12075	2	[18, 21]
Azo	Pigment Red 112	6535-46-2	12370	3	[18, 21, 24]
Azo	Pigment Yellow 97	12225-18-2	11767	1	[26]
Azo	Pigment Orange 62[a]	52846-56-7	11775	0	[70]
Phthalocyanine	Pigment Blue 15	147-14-8	74160	3	[19, 21, 26]
Phthalocyanine	Pigment Green 36**	14302-13-7	74265	3	[21, 26, 27]
Phthalocyanine	Pigment Green 7	1328-53-6	74260	3	[21, 26, 27]
Polycyclic	Pigment Red 202	3089-17-6	73907	2	[24, 26]
Polycyclic	Pigment Red 122	16043-40-6	73915	2	[21, 26]
Polycyclic	Pigment violet 19	1047-16-1	73900	1	[21]
Polycyclic	Pigment Violet 1	1326-03-0	45170	3	[18, 21, 26]
Polycyclic	Pigment Violet 23	6358-30-1	51319	2	[21, 26]
Polycyclic	Pigment Violet 37	5797-98-9	51345	1	[26]
Other	Pigment Red 254	84632-65-5	56110	2	[21, 24]
Other	Pigment Yellow 138	30125-47-4	56300	2	[20, 21]

[a]Indicates that the pigment was mentioned or analysed in an article pertaining to pigment metabolism.
**Reference [21] mentioned PG 36 but did not analyse its photolysis or metabolism.

degradation products. The acute radiation dose from high-intensity laser irradiation over short nano second pulses has the potential to quickly degrade the pigments into smaller molecules. In contrast to lasers, the acute radiation dose from sunlight energy is significantly lower and may produce fewer acute degradation products. However, tattoos are exposed to sunlight for substantially longer durations than they are to lasers, thus, the degradation may, overall, be greater, albeit, over a much slower time frame. Thus, high-intensity laser light may result in acute high-level concentrations of certain degradation products whereas exposure to sunlight is likely to result in low-level degradation and chronic exposure. As would be expected, the higher intensity of the laser irradiation may result in degradation products of pigments that are not observed when the same pigments are irradiated with sunlight. For example, phthalocyanine pigments have been proposed to be stable when exposed to sunlight [26] but have been observed to produce products such as hydrogen cyanide when irradiated with a laser [19, 21, 27]. Conversely, the laser irradiation performed by Vasold et al. [28] and the solar irradiation performed by Engel et al. [17] on the azo-based pigment PR 22 found that both irradiation types produced 2-methyl-5-nitroanaline, 4-nitrotoluene and naphthol AS. Thus, while the intensity of laser irradiation allows it to degrade some pigments that the low intensity of solar irradiation cannot achieve, there are other pigments, such as azo pigments, that can be degraded into the same products by both irradiation types. Although, this research identified little to no work examining the mechanistic aspects of the photodegradation of pigments in tattoos, it is known that the azo-based compounds will undergo reductive cleavage, forming primary aromatic amines, or loss of the azo-bridge [20, 26, 29]. Further work is needed to expand our understand of the degradation mechanisms for all pigment classes as this understanding could be used to help determine which pigments should be considered potentially toxic.

Photolysis of pigments within the epidermis

Azo-based dyes such as PY 74 have been scrutinised in the literature most often, as demonstrated in Table 3. These types of pigments are used to make yellow-, orange- and red-coloured inks, thus, the focus on azo pigments is justified as, excluding black, red is the most common colour used in tattoos; followed by yellow, blue and green [30, 31]. The survey conducted in German speaking countries found that 50% and 14% of tattoos contained black and red ink, respectively. 9.6% and 9.1% of tattoos contained blue and green ink, respectively, while yellow was found in 8.2% of tattoos [30]. Compared with these colours, other colours had a combined prevalence of 6.9% in tattoos. Brady et al. [32] also found that the most observed colour in tattoos was black (90.3%), followed by red (36%), blue (30.3%) and Green (28%). Brady et al. [32] also found that yellow and orange were observed in 21% and 12% of tattoos, respectively. The major focus of investigations being those into azo-based pigments is justified when observing these case reports by Kluger and Koljonen [33] concerning melanomas, carcinomas and similar tumours or tumour type conditions arising in tattooed areas of skin. They found that in 32 of the 65 case reports, the condition arose in a location of the tattoo that contained a red ink. Furthermore, only 3 of those 32 case reports indicated that the affected area contained multiple colours other than red. The review also found that black-tattooed areas of skin were the second most common areas for these

Table 4. Methods of irradiation used to investigate the photolysis of tattoo pigments.

Laser irradiation			Natural and/or simulated solar irradiation		
Reference	Laser	Settings	Reference	Light source	Settings
[21]	Nd:YAG	5 J/cm^2, 4-mm spot size, >20-ns pulse duration	[1]	Xenon-arc lamp	6.5 kW with WG320 glass filters
[21]	Ruby	3–5 J/cm^2, 4-mm spot size, >20-ns pulse duration	[17]	Broad band UV	280–320-nm wavelengths, intensity of 0.0015 W/cm^2, 25-cm distance, 4-h exposure
[28]	Nd:YAG	8-ns pulse duration, 1-mm spot size, 2 J/cm^2, 10 Hz for 10-min repitition, 15-mJ light impulse	[17]	Natural sunlight	110 days exposure
[67]	Nd:YAG	2.5 J/cm^2, wavelength of 532 nm	[24]	Atlas Suntest CPS+	290–800-nm wavelengths, intensity of 750 W/m^3, 35 °C, exposure durations of 20–40–60 h 1–2 weeks
[19]	Nd:YAG	1064 nm and 532-nm wavelengths, 5 J/cm^2, 3-mm spot size	[18]	Natural sunlight	Exposure stopped after 12 and 14 days (March–April), 30, 35 and 41 days (April–May) and 39 and 40 days (April–June)
[19]	Ruby	694-nm wavelength, 5 J/cm^2, 5-mm spot size			
[24]	Nd:YAG	3-mm spot size, 4-J/cm^2 intensity, 10-ns pulse, 532-nm wavelength			
[20]	Nd:YAG	532 nm, 5 J/cm^2, 4-mm spot size, >20-ns pulse duration			
[20]	Ruby	694 nm, 3–5 J/cm^2, 4-mm spot size, 20-ns pulse duration			
[27]	Nd:YAG	532 nm, 0.525 J/cm^2, 4-mm spot size			
[24]	Nd:YAG	532 nm, 10-ns pulse duration, 4 J/cm^2, 3-mm spot size			

condition to affect [33]. After reviewing these cases, Kluger and Koljonen [33] concluded that the association of the cutaneous malignancies needs to be considered as a coincidental as they report that the number of cases does not exceed the standard rate of cutaneous malignancies.

It can be seen from Table 5 that information on polycyclic- and phthalocyanine-based pigments is lacking, though it has been hypothesised by Hauri and Hohl [26] that these types of pigments have a greater stability than the azo-based pigments due to the lack of reported degradation products when irradiated with solar light. However, during tattoo laser removal, Q-switched lasers have successfully been used to remove or, at the very least, severely fade tattoos containing phthalocyanine pigments [14], indicating that there is some interaction between laser light and the pigment, either through direct interaction or indirect interaction mediated by the inter- and intra-cellular environments. This is demonstrated by the research by Schreiver [21], Schreiver et al. [19] and Bauer et al. [27] indicate that the higher irradiation intensities of laser irradiation causes phthalocyanine pigments to degrade, producing compounds such as hydrogen cyanide, chlorobenzonitriles and benzenedicarbonitriles.

Exposure of tattoo pigments to sunlight within the epidermis

Many of the articles in this review attempted to mimic the natural light conditions to which tattoo pigments are exposed. That is, sunlight is comprised of ~6% ultraviolet radiation (UVR), ~52% visible light (λ 400–760 nm) and ~42% infra-red radiation (λ 760–10^6 nm). UVR is further divided into UVC (λ 100–290 nm), UVB (290–320 nm) and UVA (λ 320–400 nm) though UVC is absorbed by the atmosphere and does not reach the ground [34]. However, as the wavelength of light lengthens, its ability to penetrate the skin increases and therefore shorter wavelengths that are defined as UVB radiation only penetrate skin to a depth of ~20 µm. This indicates that the portion of UVB radiation that reaches the dermis layer of our skin to participate in the photolysis of tattoo pigments is very limited whereas UVA radiation can reach the subcutis layer of the skin and, consequentially, has a greater contribution in tattoo pigment photolysis [34, 35]. Thus, in order to simulate the light conditions experienced by pigments, two articles exposed samples of tattoo inks or pure pigments to sunlight by sitting them next to a window or on a similar surface. Three articles were published that exposed samples to UV lamps.

The exposure conditions of tattoo pigments to the broad band UV lamp used by Cui et al. [1] appear most appropriate as they modelled their exposure conditions from the work performed by Howard et al. [23]. Howard et al. [23] measured the UVR spectrum of terrestrial sunlight at noon in the USA and compared it with the spectra produced by a fluorescent sun lamp with, and without, a cellulose triacetate filter and a 6.5-kW xenon-arc lamp that was filtered using a Schott WG320 filter. They reported that the xenon-arc lamp was the most representative, though it was found to emit low levels of light below 290 nm that is not observed in terrestrial sunlight. Wezel [24] also used a filtered Atlas Suntest CPS+ xenon lamp to simulate terrestrial light. The Suntest CPS+ xenon lamp is commonly employed in the standard testing of cosmetic light fastness and sunscreen sun protection factor rating [36] and the comparison of the lamp's UV spectrum with a spectrum of terrestrial light recorded by the International Commission of Illumination shows that it generates reasonable approximation of terrestrial light.

Engel et al. [17] used a broad band UV lamp that emits in a range of 280–320 nm. The UV spectrum of the broad band lamp that Engel et al. [17] used was compared to the absorption spectrum of PR 22 and terrestrial light and this showed that the emission peak of the lamp was ~40–50 nm lower than the earliest peak observed in the terrestrial light spectrum. Furthermore, the work cited by Engel et al. [17] for the terrestrial light spectrum was reported on a company website that could not be accessed and,

Table 5. List of photolytic degradation products, the parent pigment and the parent pigment type.

Degradation product	Parent pigment type	Parent pigment	Irradiation type	Reference
N-(2-methoxyphenyl)-3-oxobutanamide	Azo	PY 74	Simulated solar	[1]
1-Phenyl-3-methyl-5-pyrazolone	Azo	PO 13	Laser	[21]
2-Aminobenzonitrile	Azo	PO 13	Laser	[21]
2-Chloroaniline	Azo	PO 13	Laser	[21]
3,3-Dichlorobiphenyl-4-amine	Azo	PO 13	Laser	[21]
o-Phentidine	Azo	PR 170	Laser	[21]
1-Amino-2-Naphthol	Azo	PR 112	Laser	[24]
2-Methylacetoacetanilide	Azo	PY 14	Laser	[24]
4-Chloro-2,5-dimethoxyaniline	Azo	PY 83, 98	Laser	[24]
Acetoacetanilide	Azo	PO 16	Laser	[24]
o-Toluidine	Azo	PY 14, PR 112	Laser	[24]
1,2-Dihydroxynaphthalene	Azo	PO 5	Natural solar	[18]
1-Phenyl-2,3-dimethyl-4-aminopyrazolon	Azo	PO 13	Natural solar	[18]
B-naphthol	Azo	PO 5	Natural solar	[18]
Trans-o-coumaric acid	Azo	PO 5	Natural solar	[18]
2-(Hydroxyimino)-N-(2-methoxyphenyl)-3-oxobutanamide	Azo	PY 74	Simulated solar	[1, 26]
N,N″-bis(2-methoxyphenyl)urea	Azo	PY 74	Simulated solar	[1, 26]
3,3′-Dichlorobenzidine	Azo	PO 13, 34, PY 14	Laser	[20, 21, 24, 26]
4-Aminobenzamide	Azo	PR 170	Simulated solar, laser	[21, 24, 26]
Benzamide	Azo	PR 170	Simulated solar, laser	[21, 24, 26]
1-4-Dichlorobenzene	Azo	PR 9	Laser	[26, 28]
2-5-Dichloroaniline	Azo	PR 9	Laser	[26, 28]
4-Nitrotoluene	Azo	PR 22	Simulated and natural solar, laser	[17, 26, 28, 67]
2-Methylformanilide	Azo	PY 14, PR 112	Simulated solar, laser	[24, 26]
3,3′-Dichlorodiphenyl	Azo	PO 13, PY 14	Simulated solar, laser	[24, 26]
3,3′-Dimethoxydiphenyl	Azo	PO 16	Simulated solar, laser	[24, 26]
Formanilide	Azo	PO 16	Laser	[24, 26]
o-Acetoacetanisidine	Azo	PY 74	Simulated solar, laser	[24, 26]
2-Methylacetanilide	Azo	PY 14, PR 112	Simulated solar, laser	[24, 26]
4-Hydroxybenzamide	Azo	PR 170	Simulated and natural solar, laser	[18, 24, 26]
Naphthol AS	Azo	PR 170	Simulated and natural solar, laser	[18, 24, 26]
Phenyl isocyanate	Azo	PO 13	Laser	[20, 21]
2-Methyl-5-nitroaniline	Azo	PR 22	Simulated and natural solar, laser	[17, 28, 67]
4-Chloro-2,5-dimethoxyacetoacetanilide	Azo	PY 83, 97	Natural and simulated solar	[18, 24]
Hydrogen cyanide	Azo, phthalocyanine	PO 13, PB 15	Laser	[19, 21, 26]
Benzene	Azo, phthalocyanine, other	PO 13, PB 15, PY 138	Laser	[19–21]
Benzonitrile	Azo, phthalocyanine, polycyclic, other	PO 13, PR 170, 254, PB 15, PV 19, PY 138	Laser	[19, 21]
1-Cyanonaphthalene	Azo, polycyclic	PR 170, PV 19	Laser	[21]
Biphenyl	Azo, polycyclic	PO 13, PV 19	Laser	[21]
Aniline	Azo, polycyclic	O 13, 16, PR 170, V 19	Laser	[20, 21, 26]
Chlorobenzene	Azo, phthalocyanine, other	PO 13, PR 254	Laser	[21]
3-Chlorobenzamide	Other	PR 254	Laser	[21]
3-Chlorobenzonitrile	Other	PR 254	Laser	[21]
4-Chlorobenzonitrile	Other	PR 254	Laser	[21]
Pentachlorobenzene	Other	PY 139	Laser	[21]
Hexachlorobenzene	Other	PY 138	Laser	[20, 21]

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Table 5 continued

Degradation product	Parent pigment type	Parent pigment	Irradiation type	Reference
Xylene	Other	PY 140	Laser	[20, 21]
Biphenyldicarbonitrile	Phthalocyanine	PB 15	Laser	[21]
2-Butanone	Phthalocyanine	PB 15	Laser	[19]
Benzenedicarbonitriles	Phthalocyanine	PB 15	Laser	[19, 21]
3,4-Dimethyl-2-hexene	Phthalocyanine	PG 7	Laser	[27]
2,4-Dimethyl-2-hexene	Phthalocyanine	PG 7	Laser	[27]
2,4-Dimethyl-1-hexene	Phthalocyanine	PG 7	Laser	[27]
5-Methyl-1-heptene	Phthalocyanine	PG 7	Laser	[27]
4-Methyl-1-heptene	Phthalocyanine	PG 7	Laser	[27]
3,5-Dimethyl-2-hexene	Phthalocyanine	PG 7	Laser	[27]
Heptilhexylether	Phthalocyanine	PG 7	Laser	[27]
1,2-Dimethyl-cyclohexane	Phthalocyanine	PG 7	Laser	[27]
2,2′-(Methylenebis(oxy))bispropane	Phthalocyanine	PG 7	Laser	[27]
2,2′-(Ethylidenebis(oxy))bispropane	Phthalocyanine	PG 7	Laser	[27]
1,2,3,5-Tetrachlorobenzene	Phthalocyanine	PG 7	Laser	[27]
3,4,5,6-Tetrachloro-1,2-benzonitrile	Phthalocyanine	PG 7	Laser	[27]
2,4,5,6-Tetrachloro-1,3-benzonitrile	Phthalocyanine	PG 7	Laser	[27]
2,4,6-Trichloro-1-benzonitrile	Phthalocyanine	PG 7	Laser	[27]
Pentachlorobenzene	Phthalocyanine	PG 7	Laser	[27]
Diethylphthalate	Phthalocyanine	PG 7	Laser	[27]
2,3,4,5-Tetrachlorobenzonitrile	Phthalocyanine	PG 7	Laser	[27]
1,3,7-Trichloronaphthalene	Phthalocyanine	PG 7	Laser	[27]
1,2,3,5-Tetrachloro-4-ethoxy benzene	Phthalocyanine	PG 7	Laser	[27]
2,3,5,6-Tetrachloro-1-phenol	Phthalocyanine	PG 7	Laser	[27]
Pentachlorobenzonitrile	Phthalocyanine	PG 7	Laser	[27]
4-Chloroaniline	Polycyclic	PR 202	Laser	[24, 26]

thus, is unverifiable. Consequentially, the photolysis of PR 22 performed by Engel et al. [17] using the broad band lamp can be said to be an important representative degradation of PR 22 by UV radiation but is not necessarily representative of the photolysis of PR 22 under the types of conditions that it would be expected to experience in a tattoo. Ultimately, Engel et al. [17] did not rely on the broad band lamp as a model of the expected conditions experienced by a tattoo, rather, they used it as a comparison to the photolysis of PR 22 that was exposed to terrestrial light.

Metabolism of pigments

Unlike the photolysis of tattoo pigments, there is a significant a lack of research on the metabolism of tattoo pigments by the human body. The results obtained by Cui et al. [25] found that cytochrome P450 enzymes, especially the 1A1 and 1A2 enzymes, could metabolise PY 74 display a need to further our understanding of tattoo pigment metabolism. This is further supported by considering the distribution of cytochrome P450 enzymes in the human body reported by Yengi et al. [37]. They performed an assay on the mRNA of CYP450 enzymes in skin samples from 27 human volunteers. Their results showed that CYP1A1 was present in low levels (mean of 0.004 attograms/18S mRNA), CYP1B1 was consistently present in high levels (2.46 femtograms/18S mRNA) and CYP3A4 was detected in moderate levels (mean of 1.10 femtogram/18S mRNA). CYP1A2 was below the limits of detection [37]. Rolsted et al. [38] found that CYP3A4 was present specifically in the dermis of the human skin, however, there was no evidence for the presence of CYP1A1 in the human dermis. This lack of enzymatic activity from CYP1A1 was hypothesised to be a result of the enzymatic product being below limits of detection [38]. In contrast, research by Saeki et al. [39] identified mRNA coding for CYP1A1 in addition to mRNA coding for CYP1B1 in skin fibroblasts from the human dermis. The results for CYP1A1, CYP1B1 and CYP3A4 are further supported by Wiegand et al. [40] and Luu-The et al. [41] which, thusly, supported the hypothesis put forward by Rolsted et al. [38]. Finally, minor quantities of RNA coding for CYP1A2 has also been identified in the human dermis, although it was in greater abundance within the epidermis [41]. As such, the metabolism of pigments within the dermis layer seems to be a possible method for them to be removed from the skin, causing tattoos to fade and, possibly, as a process for producing harmful metabolic products.

Light microscopy and transmission electron microscopy studies of mice that had been tattooed with either a black or red ink showed that the pigments could be collected within the Kupffer cells within the liver [42]. A number of research and case reports have also indicated that tattoo pigments are also relocated into lymph nodes within the area of the tattoo [33, 42–45]. The lymphatic system is responsible for draining the lymph into the circulatory system so that waste can be excreted from the body, which suggests that the tattoo pigments are transported into the liver via the blood [42]. Thus, while Sepehri et al. [42] only observed tattoo pigment collection within the liver, it is possible that collections of tattoo pigments within other organs may be possible. If such a process does occur, then pigments are likely to be exposed to xenobiotic metabolism not only in the skin, but also within the liver, kidneys and respiratory tract. Within the liver, cytochrome P450 enzymes have been found to have a concentration between 1.0 and 1.5 nmol per mg of protein while the kidneys have only been found to possess CYP450 enzymes in a concentration between 0.1 and 0.2 nmol per mg of protein [46]. Within the kidneys, CYP1A1 and CYP3A4 have been identified,

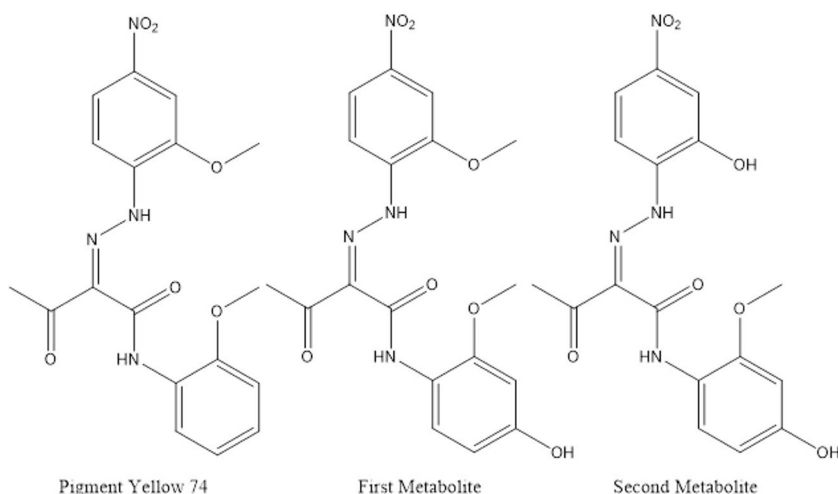

Fig. 3 Reported Metabolites of Pigment Yellow 74. Metabolites of pigment yellow 74 formed by cytochrome P450 enzymes [26].

which suggests that there is the possibility for PY 74 to be metabolised [47, 48]. Similarly, CYP1A1, CYP1A2, CYP1B1 and CYP3A4 have all been identified within lungs and colon while the stomach expressed CYP1A1, CYP1A2 and 3A4 and the small intestines expressed CYP1A1, CYP1B1 and CYP3A4 [49]. Zhao and Allis [50] investigated the activity of CYP1A2 and CYP3A4 enzymes present within the liver and Persson et al. [51] quantified the concentration of mRNA coding for CYP1A1, CYP1A2 and CYP3A4 in human liver samples. While CYP1B1 was not reported in the previously mentioned articles concerning the CYP450 isoenzyme content of liver cells, it is undoubtedly present as Chang et al. [52] not only quantified the mRNA of CYP1A1 and CYP1A2, but also quantified the mRNA coding for CYP1B1 demonstrating its presence within the liver. Given the above, it is likely that PY 74 could undergo metabolism in the liver, lungs, kidney or gastrointestinal tract. It is necessary, however, to further assess the distribution and metabolism of tattoo pigments to confirm that pigments other than PY 74 can be metabolised and to determine whether they are distributed throughout the body rather than remaining stationary in the skin or lymph nodes.

Toxicity of the degradation products of tattoo pigments

As noted above, various pigments have been found to undergo photolysis and PY 74 has been found to undergo metabolism. Given the quantities of CYP450 enzymes present within the skin and the current knowledge on the distribution of pigments throughout the body, however, it seems unlikely that the metabolism of pigments would occur quickly. Thus, metabolism is most likely to cause damage only through the accumulation of metabolites that the body cannot excrete. Conversely, the photolysis of pigments may well pose a larger threat to human health, especially when it occurs due to laser tattoo removal treatments. As seen in Table 6, many of the photolysis products are reported to cause harm, however, many of the toxicological studies for these photolysis products have not been performed to account for exposure to them via their production in the dermis.

Benzonitrile, aniline, benzene and 3,3′-dichlorobenzidine are some of the preponderantly observed photolysis products. The Chemwatch® material safety data sheet (MSDS) for benzonitrile rates it as harmful and the ECHA has rated both the dermal and oral toxicity as Acute Class 4, harmful [53, 54]. Benzonitrile is believed to cause systemic damage when absorbed into the bloodstream via cuts or abrasions and results in a fatality if 150 g is ingested, though 150 g is a significant dose to ingest and seems unlikely to occur during tattoo removal [55]. When the cytotoxicity of benzonitrile was investigated using hamster and pig cells and human kinesin expressed in *E. coli*, it was found that benzonitrile caused an incorrect distribution of chromosomes within daughter cells [56]. Aniline is also considered to possess a high toxicity hazard and a moderate body contact hazard as inhalation and ingestion can cause methemoglobinemia while contact can result in skin irritation (including redness, swelling and blistering), eye irritation and respiratory irritation [57–59]. The mean lethal dose of aniline is reported as being somewhere between 15 and 30 g [57, 60]. Although 3,3′-dichlorobenzidine is considered to only have moderate toxicity and body contact hazards, it is considered to be an extreme chronic hazard in its ChemWatch [61] MSDS. Similar to aniline, 3,3′-dichlorobenzidine may cause methemoglobinemia when inhaled or ingested, though it is more concerning that 3,3′-dichlorobenzidine has been found to be carcinogenic and impacts fertility with chronic exposure [51, 61, 62]. While the lethal dose exceeds 8 g/kg, when 3,3′-dichlorobenzidine is irradiated with light, the carcinogenic effects can be observed at concentrations of 2 µM in *Salmonella typhimurium* and cytotoxicity and genotoxicity in Human Jurkat T-cells is observed from concentrations between 2 and 100 µM [61, 63]. Considering that the average concentration of pigment injected into the skin when tattooing is 2.53 mg/cm^2 and that the pigment is not converted into any one of these products at a ratio of 1:1 or higher, it seems unlikely that any of these three photolysis products could be fatal [10]. However, there is the possibility for adverse health effects to occur. Zeilmaker et al. [64] found that 3,3′-dichlorobenzidine, for example, has a concentration of 340 µg/g within an airbrush ink. They calculated that aerosol exposure leads to a lifetime daily absorption through the skin into the body of 5 ng/day, but the exposure calculations were not undertaken for direct dermal injection. In contrast to airbrush inks, tattoo inks have been reported to contain much lower levels of 3,3′-dichlorobenzidine (median concentrations 1.7 µg/g), even after reductive splitting [65]. Whilst these levels are much lower, the tattoo inks are directly injected into the skin, rather than being absorbed from the air and still may result in a toxicological impact. Indeed, Sabbioni and Hauri [66] calculated that direct exposure to 3,3′-dichlorobenzidine and o-toluidine from the degradation of a 400-cm^2 tattoo (20 × 20 cm) over 20 years containing 1.0 g of PY 14 increases the lifetime (70 years) cancer risk to tattooed individuals. They report an expected additional 4.5 cancer cases per 10,000 people with tattoos, which is significantly higher than the US-EPA tolerance of one additional case per one million people. Furthermore, 3,3′-dischlorobenzidine is an intermediate used during the synthesis of azo pigments, which is often found as an impurity in tattoo inks comprised of azo pigments [63].

Table 6. List of photolytic degradation products and their human toxicity.

Degradation product	Toxicity
Benzonitrile	Harmful when contacted with skin, ingested.
Aniline	Toxic when swallowed, contact with skin, inhaled. Possible genotoxicity, carcinogenicity. May damage organs and eyes.
3,3′-Dichlorobenzidine	Harmful on contact with skin. Possible skin allergen, carcinogen.
Benzene	Skin and eye irritant. Fatal when swallowed, inhaled. Possible carcinogen, genotoxicity. May damage organs.
2-Methylacetanilide	Harmful when ingested. Skin irritant. Possible respiratory irritant. Eye damage.
2-Methylformanilide	–
3,3′-Dichlorodiphenyl	May damage fertility. May damage organs.
4-Chloro-2,5-dimethoxyacetoacetanilide	
4-Chloro-2,5-dimethoxyaniline	Harmful when ingested. Respiratory, skin and eye irritant. May cause organ damage. Possible carcinogen.
o-Toluidine	Possible carcinogen [71].
1-Cyanonaphthalene	Harmful if ingested, contact with skin, inhaled. Serious eye, skin, respiratory irritant.
Biphenyl	Skin, eye, respiratory irritation.
Chlorobenzene	Harmful if ingested or inhaled.
Hydrogen cyanide	Fatal if swallowed, inhaled or in contact with skin.
2-Methyl-5-nitroaniline	Toxic if swallowed, inhaled, in contact with skin. Possible carcinogen. Causes serious eye and skin irritation [72].
1,2-Dihydroxynaphthalene	Skin, eye, respiratory irritation. Possible allergic skin reaction.
1-4-Dichlorobenzene	Causes eye irritation. Possible carcinogen. Harmful when swallowed.
1-Amino-2-naphthol	
1-Phenyl-2,3-dimethyl-4-aminopyrazolone	Harmful if swallowed, causes eye, skin and respiratory irritation.
1-Phenyl-3-methyl-5-pyrazolone	Harmful if swallowed, causes eye, skin and respiratory irritation.
2-(Hydroxyimino)-N-(2-methoxyphenyl)-3-oxobutanamide	–
2-5-Dichloroaniline	Toxic when swallowed, inhaled and in contact with skin. Repeated or prolonged exposure may cause organ damage.
2-Aminobenzonitrile	Causes eye, skin and respiratory irritation. Harmful when swallowed, inhaled and in contact with skin. Possible carcinogen.
o-Chloroaniline	Causes eye, skin and respiratory irritation. Repeated or prolonged exposure may cause organ damage. Toxic when swallowed, inhaled and in contact with skin.
3,3-Dichlorobiphenyl-4-amine	–
3,3′-Dimethoxydiphenyl	–
4-Aminobenzamide	Causes eye, skin and respiratory irritation. Harmful when inhaled or swallowed.
4-Hydroxybenzamide	Causes eye, skin and respiratory irritation.
p-Nitrotoluene	Genotoxic. Toxic when swallowed, inhaled and in contact with skin. Repeated or prolonged exposure may cause organ damage.
Benzamide	Harmful if swallowed. Possible of causing genetic defects.
B-naphthol	Harmful if inhaled or swallowed
Formanilide	Harmful if swallowed. May cause an allergic skin reaction.
N-(2-methoxyphenyl)-3-oxobutanamide	Harmful if swallowed
N,N″-bis(2-methoxyphenyl)urea	–
Naphthol AS	May cause an allergic skin reaction.
o-Phenetidine	–
Phenyl isocyanate	Causes eye, skin and respiratory irritation. May cause an allergic skin or breathing reactions. Possible carcinogen. Harmful if swallowed or inhaled. Repeated or prolonged exposure may cause organ damage.
Phenyl isocyanate	Causes eye, skin and respiratory irritation. May cause allergic or asthma symptoms upon inhalation. May cause allergic skin reaction. Possible carcinogen. Fatal when inhaled. May cause organ damage upon prolonged/repeat exposure.
Trans-o-coumaric acid/2-hydroxycinnamic acid	Causes eye, skin and respiratory irritation. Harmful if swallowed. May cause allergic skin reaction.
2-Butanone/methyl ethyl ketone	Causes eye and respiratory irritation. May cause drowsiness or dizziness. May cause skin drying and cracking upon repeat exposure.

Table 6 continued

Degradation product	Toxicity
3-Chlorobenzamide	–
3-Chlorobenzonitrile	Causes eye, skin and respiratory irritation. Toxic when swallowed. Harmful upon skin contact. May cause allergic skin reaction.
4-Chloroaniline/p-chloroaniline	Causes serious eye irritation. Possible carcinogen. Toxic if swallowed or inhaled and when in contact with skin. May cause allergic skin reactions.
4-Chlorobenzonitrile	Causes eye, skin and respiratory irritation. Harmful if inhaled, swallowed or in contact with skin. May cause allergic skin reactions.
Benzenedicarbonitriles	N/A (Constitutes a large group of molecules)
4,4′-Biphenyldicarbonitrile	Harmful if inhaled, swallowed or in contact with skin.
Hexachlorobenzene	Prolonged or repeat exposure causes organ damage. Possible carcinogen.
Pentachlorobenzene	Harmful if swallowed.
Xylene	Harmful when inhaled or in contact with skin. Causes skin and eye irritation. May cause dizziness or drowsiness.

The toxicity data for compounds with no reference were taken from ChemWatch [53].

Experimental solvents and wavelengths
When analysing the photolytic degradation of pigments, the solvent and laser wavelength are important considerations in the experimental design. The solvents used in the various literature of tattoo pigment photolysis are water [19, 20, 24], tetrahydrofuran (THF) [1, 17], acetonitrile [28], propan-2-ol [27] and dichloromethane (DCM) [17, 18]. The primary limitation of using these solvent suspensions for in vitro studies is that the degradation products formed may not be truly representative of the degradation products formed in vivo. While using solvents can aid in suspending the pigment for simplified analysis, they may play a role in the laser degradation processes by forming adducts or altering the mechanistic pathway when compared with interstitial fluid and skin tissue. As a result, irradiated samples may lack degradation products that are actually produced during the photolysis of pigments within the skin or contain products that would not normally be observed in a skin matrix. THF, dioxane, chloroform and DCM were chosen by Engel et al. [17] for their UV irradiation experiments, as they were reported to be stable to irradiation. However, the interaction between solvent and pigment and their likely effect on the degradation was not explored. Alternatively, work using a mouse model, such as in the work of Engel et al. [67], or post-mortem pig skin samples, such as in the work of Hering et al. [20] and Gaugler [18] are more likely to see degradation products that are reflective of those that may be expected in human laser tattoo removal. In their mouse model work Engel et al. [67] confirmed the presence of napthol-AS, 2,5-MNA, and 4-NT that were observed in their solution studies. However, they limited their report to these three compounds, thus it is difficult to say if there were significant differences between the in vitro solution studies and the in vivo mouse model work.

The most common laser and wavelength used in literature studies of laser tattoo pigment degradation is the Q-switched Nd;YAG laser operating at 532 nm (Table 4). However, Q-switched Nd;YAG, ruby and alexandrite laser are used in tattoo removal procedures at wavelengths of 532, 1064, 694 and 755 nm, with the choice of laser dependent on the colour of the pigment [14, 68]. Using a 532-nm laser is not ideal for all studies, as the pigments may not absorb light strongly at this wavelength and, within a typical practice, is only used for light colours such as yellow, red and orange. When examining azo dyes, Herring et al. [16] found that, for PY 138 and PO 13, the 532-nm Nd:YAG laser wavelengths resulted in higher concentrations of degradation products than the 1064-nm Nd;YAG or 694-nm Ruby laser wavelengths. For those articles examining the photolysis of phthalocyanine pigments (PB 15, PG 7 or PG 36), the ideal laser to use would be either the 694-nm Ruby or 755-nm alexandrite laser as these pigments have low absorptions at 532 and 1064 nm [27, 69]. Thus the work of Schreiver [21] and Schreiver et al. [19] who used both the ruby and the Nd:YAG lasers for irradiation of the phthalocyanine class of pigments is likely to be the most reflective of laser removal procedures for these pigments. Using a 532-nm laser, Bauer et al. [27] observed the presence of chlorinated analogues of 1,2-benzenedicarbonitrile upon the photolysis of PG 7. This pigment has a similar chemical structure to PB 15 studied by Schreiver et al. [19] with the PG 7 being a chlorinated analogue of PB 15. Both studies resulted in similar products, with irradiation of PG 7 resulting in chlorinated analogues of the degradation products of PB 15. For example 1,2-benzenedicarbonitrile was found by Schreiver [21], and a chlorinated analogue of 1,2-benzenedicarbonitrile was observed by Bauer et al. [27]. This indicates that the larger degradation compounds may have been produced by similar degradation pathways, however, further study is necessary to confirm such a hypothesis. Interestingly, Schreiver et al. [19] showed that the 1064 and 532-nm lasers resulted in minimal degradation of PB 15, whilst 532 nm was successfully used by Bauer et al. [27]. This may be a result of differing laser fluence and frequency along with differing concentrations of pigments used by the two groups. These differences highlight the need to examine the photolysis of pigments with all laser wavelengths and conditions that they are likely to be subjected to during laser tattoo removal procedures. The absence of a degradation product may not be reflective of what occurs under the conditions of tattoo removal.

CONCLUSION
At first glance, there is a large variety of tattoo pigments that have been analysed to determine their photolytic degradation products (see Table 3), however, most of these pigments are only examined by one group of researchers and those pigments that are investigated in multiple articles are primarily azo-based pigments. This has resulted in a lack of information in the literature concerning the photolytic degradation of polycyclic- and phthalocyanine-based pigments. Similarly, there is lack of information concerning the metabolism of all types of pigments by human enzymes as Cui et al. [25] were the only researchers found to have analysed pigments' metabolic products. The photolytic degradation of non-azo base pigments is important as laser tattoo removal procedures, and exposure to solar radiation, cause tattoos to fade [14]. Furthermore, the resultant products from pigments that have been found to undergo photolytic degradation are often reported as having potential to cause harm to an individual's health, such as in the case of aniline

or 3,3′-dichlorobenzidine, both of which are, at the least, suspected carcinogens and cause methemoglobinemia [51, 57, 58, 60, 61, 63]. The metabolism of tattoo pigments should be further investigated, as the work by Cui et al. [25] has demonstrated the CYP1A1, CYP1A2, CYP1B1 and CYP3A4 enzymes found in the human skin can metabolise the azo-based pigment PY 74. Reports of tattoo pigments in lymph nodes and in the liver indicate that tattoo pigments may be more mobile throughout the body than previously thought [42, 43, 45]. This further emphasises the need for research into the metabolism of tattoo pigments, as the CYP450 enzymes that were found to metabolise PY 74 are generally found within the liver, small intestines, lungs and kidneys, in addition to the dermis layer of the skin [46–52]. By better understanding of the toxicity of photolytic and metabolic products of tattoo inks public health will be better protected and reduce the risk to the future generations that seek to express themselves through body art.

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ACKNOWLEDGEMENTS

The authors would Like to acknowledge Deborah Westland (CEO of the Australian College of Laser Therapy) for discussing the tattoo laser removal process. The authors would also like to acknowledge Associate Professor Ingo Koeper and Maximillian Mann from Flinders University (Bedford Park, South Australia) for their aid in translating articles written in German. The authors would like to thank the Australian Government Department of Education, Skills and Employment for providing an Australian Research Training Program Scholarship to TRF.

COMPETING INTERESTS

The authors declare no competing interests.

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Correspondence and requests for materials should be addressed to C.E.L.

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