

**Ministry of Environment and Food of Denmark** Environmental Protection Agency

## Test method for PAAs in tattoo and PMU inks

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Sources must be acknowledged.

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### 1. Background

The aim of this project was to develop and validate one or two analytical test method(s) for free primary aromatic amines in tattoo and PMU inks in relation to the restriction dossier on chemicals in tattoo and PMU inks, currently under evaluation of the European Chemical Agency (ECHA). The test method(s) shall comply with the requirements for test methods of the Forum for Exchange of Information on Enforcement (Forum), with the aim to be listed in the Compendium of analytical methods, which content recommended analytical methods by the Forum to check compliance with REACH XVII<sup>1</sup> restrictions (ECHA 2016b). The test methods developed in this project do not address azo-pigments, only free PAAs.

<sup>&</sup>lt;sup>1</sup> Existing restrictions of chemicals in REACH (EF no. 1907/2006) are listed in REACH appendix XVII

# 2. Selection of the PAAs in this project

Piccinini et al. (2015) have identified 67 azo-colorants that are used in tattoos and PMU inks. In a study (DEPA, 2017) these 67 azo-colorants have - based on their chemical structures been analyzed with respect to a theoretical decomposition by cleavage of the azo bond and amide hydrolysis. The result of the analysis shows that 24 PAAs may theoretically occur in tattoo and PMU inks due to cleavage of the azo-bond, due to amide hydrolysis or as an impurity from the production of the azo-colorant (DEPA 2017).

The Dossier Submitter of the restriction dossier on tattoo and PMU ink, currently under scrutiny of ECHA, used the results of the study by the Danish EPA (DEPA, 2017) and applied an approach selecting PAA's, with a harmonized classification as carcinogenic or as skin sensitizing, to be included in the restriction dossier using the following criteria

- 1. have been found in tattoo inks or inks for PMU on the market;
- may be present in tattoo inks due to either cleavage of azo-bond or amide hydrolysis of an azo colorants used in tattoo inks or originate from the production of the azo colorants used in tattoo inks;
- may be present in tattoo inks due to reductive cleavage of azo bond of one of the azo colorants listed in the CoE ResAP(2008)1; or
- may be present in tattoo inks either due to reductive cleavage of azo bond or due to Amide hydrolysis of one of the azo colorants restricted in Annex XVII entry 43 of REACH in various textiles.

Using the above-mentioned criteria, the Dossier Submitter identified 27 PAA's with a classification as carcinogenic or as skin sensitizing. The negative list of aromatic amines which should neither be present in tattoos and PMU products nor released from azo-colorants in the CoE ResAP(2008)1 was almost identical with the PAA's identified by the Dossier Submitter.

However, later during the development of the dossier, additional two PAA's was included in the restriction proposal: 4-amino-3-fluorophenol (CAS no. 399-95-1) and 2,6-xylidine (CAS no. 87-62-7). At that time, the development of a chemical analytical methodology was already initiated and the two additional PAA's are for this reason not included in this project. Further, two of the azo-colorants listed in the CoE ResAP(2008)1, was also proposed to be included in the restriction dossier, due to the fact that they could both be classified as azo-colorants and PAAs: 6-amino-2-ethoxynaphthaline (CAS no. 293733-21-8) and 2,4-xylidine (CAS no. 95-68-1). However, it was concluded that these two azo-colorants should not be included in the dossier and are for this reason not included in this project.

In Table 1, the initially 27 selected PAAs are listed. These are the PAA's which have been selected in this project. TABLE 1. PAAs selected in the dossier proposal and included in this project.

CAS no.	PAA
95-53-4	o-toluidine
106-49-0	p-toluidine
95-69-2	4-chloro-o-toluidine
137-17-7	2,4,5-trimethylaniline
120-71-8	6-methoxy-m-toluidine
62-53-3	Aniline
106-50-3	p-Phenylenediamine
106-47-8	4-chloroaniline
90-04-0	o-Anisidine
99-55-8	5-nitro-o-toluidine
95-80-7	4-methyl-m-phenylendiamine
615-05-4	4-methoxy-m-phenylenediamine
91-59-8	2-naphthylamine
92-67-1	p-Phenylaniline
5417-63-0	3-amino-2-naphthol
60-09-3	4-Aminoazobenzene
101-77-9	4,4'-methylenedianiline
101-80-4	4,4'-oxydianiline
92-87-5	Benzidine
97-56-3	4-o-tolylazo-o-toluidine
838-88-0	4,4'-methylene-di(o-toluidine)
119-93-7	o-tolidine
139-65-1	4,4'-Thiodianiline
101-14-4	4,4'-methylenebis[2-chloroaniline]
91-94-1	3,3'-dichlorobenzidine
119-90-4	3,3'-dimethoxybenzidine
121-57-3*	4-aminobenzenesulphonic acid

\* 4-aminobenzenesulphonic acid could not be analyzed with the developed method, se below

4-aminobenzenesulphonic acid (CAS no. 121-57-3) could not be analyzed applying the developed method, as it was not possible to dissolve the chemical in a relevant concentration. A literature survey for dissolution of this compound gave very mixed and contradictive suggestions for possible dissolving solvents. Several solvents were tried.

Polar solvents: methanol, acetone, water, basic and acidic.

Nonpolar solvents: hexane, dichloromethane, acetonitrile, cyclohexane, toluene etc.

The compound is probably slightly soluble; however, the concentration in the calibration solution would be too low for identification.

### 3. Development of the method

The methods were developed with solutions of known PAA's, followed by a test with a tattoo ink (without azo colorants) in which PAA's in known concentrations were added: The ink-sample was mixed thoroughly by shaking the bottle and a subsample was taken out. The tattoo ink was then spiked with at stock solution of the PAA's to obtain a concentration of approximately 10  $\mu$ g/g (10 ppm). A few tattoo inks with azo colorants were also analyzed.

#### 3.1 Extraction method

Two methods have been tried:

- The standard method for extracting PAA's from ink (DEPA 2012, Council of Europe, Resolution ResAP(2008)1) based on liquid extraction but without addition of dithionite and
- A solid phase extraction method (SPE) (Aznar 2009) by cation-exchange column.

The SPE methods is far the easiest and fastest, however, recovery and reproducibility were not as good as the other method based on liquid extraction. A further validation of the SPE method revealed that the derivatization was less efficient, possible due to the solvent used for elution. Thus, the approach based in liquid extraction was chosen for further development.

#### 3.2 Analytical method

Two methods were considered and tested for analysis:

- GC-MS (Chen et al. 2018), and
- LC-MS-MS (Mortensen et al. 2005).

Due to separation issues and detection limits the GC-MS methods was chosen for further validation. For description of the method: See the analytical protocol, Annex 1.

#### 3.3 Derivatization

Especially for the smaller PAA's (single aromate), separation and identification can be an issue as they have similar structures, see Annex 2. This is not the issue for larger PAA's (the double aromates). Derivatization was therefore tested before quantification.

The PAA's were tested for derivatization by two methods:

- Silylation with N,O-Bis(trimethylsilyl)trifluoroacetamide (BSTFA) (Halket & Zaikin 2003), and
- Benzalde-hyde/formic acid (Chen et al. 2018).

Both methods are simple with only one-step, and the corresponding derivatives can be directly injected into the GC–MS, without any additional pretreatment. A third method for derivatization with iodination were also considered (Smith et al 1998), however, this method required multiple step and were therefore rejected.

The results showed that the benzaldehyde/formic acid was the best method for the smaller PAA's (the single aromates) but not for the larger PAAs (the double aromates). The bigger

molecules gave at better identification without derivatization. This was caused by that the derivation made the molecules too big, which resulted in that the big molecules came out in the chromatogram as one big bump.

It was for this reason evaluated that, to obtain the best analytical result of the PAA's in the ink, the method had to be divide into two; one for the smaller PAA's (with derivatization) and one for the bigger (without derivatization).

The same temperature program was used for both methods. The temperature program was optimized for the separation of the various PAA's. The same program appeared to give a good separation for the smaller as well as the larger PAA's.

#### 3.4 Homogeneity and subsamples of the tattoo ink

In order to test the homogeneity of the tattoo inks a simple dry matter test was performed. However, as the dry matter content were less than 1% for eight selected tattoo inks it was not possible to use this method for testing homogeneity. It was planned to use the dry matter test to investigate different methods for taking out subsamples but that was not possible either. Other inks might have higher dry matter content depending on color or producer.

Both shaking and steering of the tattoo-ink-bottle was tested before taking out a subsample. The shaking and stirring was done by manually for approximately 10-20 seconds. As the tattoo-ink-bottles were small, less than 50 mL, mechanical shaking and stirring was not considered relevant. The subsamples were analysed for selected PAAs expected to be found in the ink used for this test. The different methods showed very low variation in the concentrations compared to the uncertainty of the analytical method and to get a representative homogeneous sample was not considered an issue.

### 4. Method performance

The performance of the test method(s) shall comply with the requirements for test methods of the Forum for Exchange of Information on Enforcement (Forum), with the aim to be listed in the Compendium of analytical methods, which content recommended analytical methods by the Forum to check compliance with REACH XVII<sup>2</sup> restrictions (ECHA 2016b).

The verification of the methods have been performed on tattoo-ink spiked with a solution of known PAA's. One tattoo-ink without azo colorants was spiked with at stock solution of the PAA's to obtain a concentration of approximately 10  $\mu$ g/ml. Then the sample was mixed thoroughly and a subsample was taken out. A few inks containing azo-colorants were also analyzed with and without spiking. It should be noted that the reproducibility is "within-laboratory".

#### 4.1 Applicability of the method

The specific free PAA's in tattoos and PMU ink included in the analytical methods are listed in table 2. The concentration range for the analysis is from LOD up to approximately 50 µg/g.

Limit of detection (LOD) is determined as three times signal to noise on the GC-MS.

Recovery is determined as the fraction of the analyte that is recovered after addition of a known amount of PAA's.

Reproducibility, which is the analytical variation between identical samples, is normally estimated only from measurement results produced by different laboratories. The reported reproducibility is instead intermediate precision (also called within-laboratory reproducibility), which can be used to provide an estimate of the between-batch variability of the results and thereby a more realistic estimate of the long-term variability of measurement results in the laboratory.

Measurement uncertainty is based on reproducibility (Rep) and recovery (Rec) using the Nordtest Method (2012). The uncertainty for the spiking procedure  $(u_{sp})$  was estimated to 0.1% and is therefore insignificant. The reported values are expanded uncertainty, U, two times the calculated uncertainty.

$$U = 2 \cdot \sqrt{Rep^2 + u_{sp}^2 + (100 - Rec)^2}$$

The performance of the methods can be found in table 2. At this stage the method does not complete fulfill the requirement to the Forum Compendium (ECHA 2016) as it still needs to be tested within different laboratories.

<sup>&</sup>lt;sup>2</sup> Existing restrictions of chemicals in REACH (EF no. 1907/2006) are listed in REACH appendix XVII

CAS no.	PAA	LOD µg/g	Rec %	Rep %	U %
95-53-4	o-toluidine	0,2	95	10	22
106-49-0	p-toluidine	0,2	97	7	15
95-69-2	4-chloro-o-toluidine	0,4	102	7	15
137-17-7	2,4,5-trimethylaniline	0,3	95	10	22
120-71-8	6-methoxy-m-toluidine	0,2	90	8	26
62-53-3	Aniline	0,2	95	12	26
106-50-3	p-Phenylenediamine	0,2	80	6	42
106-47-8	4-chloroaniline	0,1	98	10	20
90-04-0	o-Anisidine	0,2	97	8	17
99-55-8	5-nitro-o-toluidine	0,4	101	15	30
95-80-7	4-methyl-m-phenylendiamine	0,2	95	5	14
615-05-4	4-methoxy-m-phenylenediamine	0,4	98	5	11
91-59-8	2-naphthylamine	0,4	98	7	15
92-67-1	p-Phenylaniline	0,3	97	8	17
5417-63-0	3-amino-2-naphthol	0,2	95	6	16
60-09-3	4-Aminoazobenzene	0,6	90	10	28
101-77-9	4,4'-methylenedianiline	0,3	92	5	19
101-80-4	4,4'-oxydianiline	0,7	97	6	13
92-87-5	Benzidine	0,4	96	5	13
97-56-3	4-o-tolylazo-o-toluidine	0,3	92	5	19
838-88-0	4,4'-methylene-di(o-toluidine)	0,4	90	4	22
119-93-7	o-tolidine	0,4	97	4	10
139-65-1	4,4'-Thiodianiline	0,3	102	7	15
101-14-4	4,4'-methylenebis[2-chloroan- iline]	0,3	95	10	22
91-94-1	3,3'-dichlorobenzidine	0,2	95	6	16
119-90-4	3,3'-dimethoxybenzidine	0,5	100	8	16

**TABLE 2.** Performance of the analytical method. Limit of detection (LOD), Recovery (Rec), Uncertainty (U)

### 5. Test of tattoo ink

A minor survey of some of the largest recognized suppliers of inks showed that only a few azo colorants in general were used at the moment. The data are based on Material Safety Data Sheets (MSDS)'s for the inks<sup>3</sup>. Table 3 shows a list of colorants and the PAA's that theoretically might be present in tattoo inks due to either cleavage of azo-bond, amide hydrolysis or as impurities from the production. It should be noted that the survey cannot be considered as complete, as its objective was to find inks that could be used for testing and validating the developed methods.

CAS no.	PAA	Azo colorant
119-90-4	3,3'-dimethoxybenzidine	PO16
62-53-3	Aniline	PR269, PO16, PR 22, PR 2, PR 146, PY 97
90-04-0	o-Anisidine	PR 210, PY 65, PY74
91-94-1	3,3'-dichlorobenzidine	PY 14, PY 83, PO 13
95-53-4	o-toluidine	PY 14, PY 83, PO 13
99-55-8	5-nitro-o-toluidine	PR 22

TABLE 3. Colorants and PAA's that theoretically might be present in tattoo ink

According to different investigations, azo colorants can be degraded by light exposure. The azo pigments may degrade, for example under solar or laser irradiation, to the original primary aromatic amines or other decomposition products (Piccinini et al. 2015). Investigations have also shown that the azo pigments can be degraded by enzymatic and microbial metabolic reductive cleavage releasing aromatic amines. This degradation might also occur on the skin due to the influence of skin bacteria. The primarily enzyme responsible for the azo cleavage is azo reductase, which have been found in both bacteria and in mammals, including humans. Bacterial degradation has also been found to be both aerobic and anaerobic (Piccinini et al. 2015, Environment Canada 2012).

An investigation of the stability is beyond the scope of this report. However, minor single tests with sunlight and bacteria have been performed:

A bottle of tattoo ink (Color: orange) was left in the sun approximately 5 days in its original container. The tattoo ink was analyzed before and after exposure to sunlight: Aniline and 3,3'-dimethoxybenzidine was detected in small amounts in the tattoo before and after. The differences were insignificant.

Human bacteria (from a sneeze) were concentrated, added to a container of tattoo ink (Color: orange), and left for room temperature for approximately 4 days. o-anisidine and 3,3'-dimethoxybenzidine were detected in small amounts in the tattoo both before and after. The differences were insignificant.

<sup>3</sup> MSDS links: e.g. <u>https://www.intenzeproducts.eu/msds-sds/#.XHfbYuR8C7Q;</u> <u>https://www.worldfamoustattooink.com/safety-data-sheets/, https://shop16300.hsta-tic.dk/upload\_dir/docs/PDF/Starbrighte---Combine-Result.pdf</u> The measured concentrations were: 2  $\mu$ g/g o-anisidine 2  $\mu$ g/g and 0.4  $\mu$ g/g 3,3'-dimethoxybenzidine. The results indicate that neither sunlight nor bacterial degradation is a source to PAA's in tattoo ink during ordinary handling of the inks in relation to the chemical analysis of the PAA's in the laboratory. However, this was as mentioned only minor tests. Examples of measurements of other tattoo inks can be seen in table 4.

Tattoo ink	o-anisidine	3,3'-dimethox- ybenzidine	o-toluidine	aniline
Orange 1	2.0	0.4*	ND	ND
Orange 2	0.5	1.1	ND	0.3
Yellow 1	ND	ND	2.0	0.4
Green	7.6	ND	ND	ND
Red	0.5	ND	2.0	0.6

TABLE 4. Analysis of selected tattoo inks ( $\mu$ g/g). ND: not detected

\*Below detection limit

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# Appendix 1. Analytical protocol

**TABLE 1.** PAAs included in the analytical method. The table is separated in PAAs for derivatization and without derivatization for the GC-MS methods, see below.

CAS no.	PAA
	With derivatization, single aromate
95-53-4	o-toluidine
106-49-0	p-toluidine
95-69-2	4-chloro-o-toluidine
137-17-7	2,4,5-trimethylaniline
120-71-8	6-methoxy-m-toluidine
62-53-3	Aniline
106-50-3	p-Phenylenediamine
106-47-8	4-chloroaniline
90-04-0	o-Anisidine
99-55-8	5-nitro-o-toluidine
95-80-7	4-methyl-m-phenylendiamine
615-05-4	4-methoxy-m-phenylenediamine
	Without derivatization, double aromate
91-59-8	2-naphthylamine
92-67-1	p-Phenylaniline
5417-63-0	3-amino-2-naphthol
60-09-3	4-Aminoazobenzene
101-77-9	4,4'-methylenedianiline
101-80-4	4,4'-oxydianiline
92-87-5	Benzidine
97-56-3	4-o-tolylazo-o-toluidine
838-88-0	4,4'-methylene-di(o-toluidine)
119-93-7	o-tolidine
139-65-1	4,4'-Thiodianiline
101-14-4	4,4'-methylenebis[2-chloroaniline]
91-94-1	3,3'-dichlorobenzidine
119-90-4	3,3'-dimethoxybenzidine

#### Solutions and solvents:

Stock solutions: 100 mg of each PAA is dissolved in 10 mL methanol (10 mg/mL). For two PAAs, it is necessary to add acetone. 3-amino-2-naphthol (MeOH/Acetone: 5/5 mL) and 4,4'- oxydianiline (MeOH/Acetone: 7/3 mL).

The stock samples must be stored in the freezer.

Two calibration mixtures is prepared: one for the smaller PAAs (single aromate) and one for the larger PAAs (double aromate), see table 4 above.

250  $\mu$ l of each stock solution is added to 50 mL volumetric flask and filled up with methanol (concentration 50  $\mu$ g/mL of each PAA).

Seven calibration solutions from 0.1 to 25  $\mu$ g/mL must be prepared: 0.1, 0.05, 0.5, 1, 5, 10 and 25  $\mu$ g/mL MeOH.

Labelled compounds: D5-aniline, D6-3,3'-dichlorobenzidine in (20 µg/mL) MeOH. Citric buffer solution: Citrate/NaOH (0.06 molar/L, pH 6) preparation:

NaOH (4 molar): 80 g NaOH in 300-400 ml of water, after dissolution transfer to 500 volumetric flask and fill up with water.

Citric acid buffer: weigh 6,303 grams (precision 1 mg) of citric acid monohy-drate in approx. 400 ml of water, adjust to pH 6.0 with NaOH (4 molar), trans-fer to 500 ml volumetric flask and fill up with water.

Derivatization solutions: Benzaldehyde and formic acids solutions: 100  $\mu I$  in each 10 mL Methanol

#### Sample preparation and extraction

Liquid extraction: 0.5 g Tattoo ink is weighted added labelled standards (0.5 mL) and added 5-10 mL citric acid buffer (pH 6) at room temperature. Sonification bath, 60 minutes. This mixture is then extracted with 5-10 mL MTBE 3 times. The extracts are combined. Filtered with either 0.2 $\mu$ m syringe filter, Na<sub>2</sub>SO<sub>4</sub> column or through EXtrelut® NT 20 column with additional 5-10 ml MBTE in order to remove solid pigments and evaporated almost dryness and dissolved in 1 mL methanol.

#### GC-MS analysis

Column: VF-200ms capillary column (5% trifluoropropyl substituted methyl polysiloxane, length: 30 m, ID: 0,025 mm)).

Temperature program: 60°C held 2 min, 5°C/min up to 190°C, 20°C/min up to 280°C, held for 3 min.

Injector temperature: 280°C

Injection: 1 µI

Two methods are used for the analysis: For the single aromate molecules, derivatization was used and for the double aromate molecules, direct analysis on GC-MS without derivatization. Table 1 above show which PAAs that are analyzed in which method.

Derivatization: 150  $\mu$ l sample + Derivatization: 30  $\mu$  benzaldehyde and 20  $\mu$ l formic acid solution, was mixed in a GC vial. Heated in 30 minutes at 40°C. The same is done for the calibrations solutions.

Note: the mixtures with derivatization in the GC vials become unstable over time, so the samples and calibration solutions should be analyzed on GC-MS within 24 hours after derivatization.

In table 2 are the quantification ions (m/z) and approximately elution time (RT) shown.

CAS no.	PAA	m/z	RT
	With derivatization, single aromate		
95-53-4	o-toluidine	118	15.6
106-49-0	p-toluidine	194	18.1
95-69-2	4-chloro-o-toluidine	229	19.6
137-17-7	2,4,5-trimethylaniline	146	19.7
120-71-8	6-methoxy-m-toluidine	225	19.9
62-53-3	Aniline	180	22.6
106-50-3	p-Phenylenediamine	284	26.4
106-47-8	4-chloroaniline	115	26.8
90-04-0	o-Anisidine	211	26.9
99-55-8	5-nitro-o-toluidine	240	31.9
95-80-7	4-methyl-m-phenylendiamine	298	34.4
615-05-4	4-methoxy-m-phenylenediamine	314	35.6
	Without derivatization, double aromate		
91-59-8	2-naphthylamine	143	21.7
92-67-1	p-Phenylaniline	169	25.2
5417-63-0	3-amino-2-naphthol	159	27.4
60-09-3	4-Aminoazobenzene	92, 197	30.4
101-77-9	4,4'-methylenedianiline	198	30.6
101-80-4	4,4'-oxydianiline	200	30.8
92-87-5	Benzidine	184	31.1
97-56-3	4-o-tolylazo-o-toluidine	106, 225	31.6
838-88-0	4,4'-methylene-di(o-toluidine)	226	31.9
119-93-7	o-tolidine	212	32.2
139-65-1	4,4'-Thiodianiline	216	32.5
101-14-4	4,4'-methylenebis[2-chloroaniline]	231	32.7
91-94-1	3,3'-dichlorobenzidine	252	32.9
119-90-4	3,3'-dimethoxybenzidine	244	33.1

TABLE 2. GC-MS analysis. Quantification ion (m/z), approximately elution time (RT)

Calibration curves: Linearity, R2: 0.95-0.99 with derivatization, 0.94-0.98 without derivatization

### Appendix 2. Chemical structures of PAAs

#### TABLE 1. Chemical structures included in the analytical method

CAS no.	PAA	Structure
90-04-0	o-Anisidine	OCH3
95-53-4	o-toluidine	CH <sub>3</sub>
95-80-7	4-methyl-m-phenylendiamine	H <sub>2</sub> N NH <sub>2</sub>
106-47-8	4-chloroaniline	NH <sub>2</sub> Cl
99-55-8	5-nitro-o-toluidine	O <sub>2</sub> N CH <sub>3</sub>
95-69-2	4-chloro-o-toluidine	CI
62-53-3	Aniline	NH <sub>2</sub>
106-49-0	p-toluidine	NH <sub>2</sub> CH <sub>3</sub>

615-05-4	4-methoxy-m-phenylenediamine	H <sub>2</sub> N NH <sub>2</sub>
120-71-8	6-methoxy-m-toluidine	NH <sub>2</sub>
106-50-3	p-Phenylenediamine	H <sub>2</sub> N NH <sub>2</sub>
137-17-7	2,4,5-trimethylaniline	NH <sub>2</sub>
91-94-1	3,3'-dichlorobenzidine	
119-90-4	3,3'-dimethoxybenzidine	H <sub>3</sub> CO H <sub>2</sub> N H <sub>2</sub> N
119-93-7	o-tolidine	H <sub>2</sub> N
139-65-1	4,4'-Thiodianiline	H <sub>2</sub> N
91-59-8	2-naphthylamine	NH <sub>2</sub>
92-87-5	Benzidine	H <sub>2</sub> N-NH <sub>2</sub>
5417-63-0	3-amino-2-naphthol	OH NH <sub>2</sub>
92-67-1	p-Phenylaniline	NH <sub>2</sub>
97-56-3	4-o-tolylazo-o-toluidine	N <sub>NN</sub> NH <sub>2</sub>
101-77-9	4,4'-methylenedianiline	H <sub>2</sub> N NH <sub>2</sub>



TABLE 2. Chemical structures of PAAs not included in the method according to chapter 2.

CAS no.	PAA	Structure
121-57-3	4-aminobenzenesulphonic acid	H <sub>2</sub> N H <sub>2</sub> N
399-95-1	4-amino-3-fluorophenol	OH NH <sub>2</sub> F
87-62-7	2,6-xylidine	H <sub>3</sub> C CH <sub>3</sub>
293733-21-8	6-amino-2-ethoxynaphthaline	H <sub>2</sub> N O CH <sub>3</sub>
95-68-1	2,4-xylidine	H <sub>2</sub> N H <sub>3</sub> C CH <sub>3</sub>

#### Test method for PAAs in tattoo and PMU inks

The aim of this project was to develop and validate one or two analytical test method(s) for free primary aromatic amines in tattoo and PMU inks in relation to the restriction dossier on chemicals in tattoo and PMU inks, currently under evaluation of the European Chemical Agency (ECHA). The test method(s) shall comply with the requirements for test methods of the Forum for Exchange of Information on Enforcement (Forum), with the aim to be listed in the Compendium of analytical methods, which content recommended analytical methods by the Forum to check compliance with REACH XVII restrictions (ECHA 2016b).

The test methods developed in this project do not address azo-pigments, only free PAAs.



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