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Evaluation of the Efficacy of Adhesive Film in Securing Biological Trace Evidence and the Isolation of Human Cells by Laser Microdissection
Applicability of Adhesive Tapes in Securing Biological Stains and Direct Laser Microdissection of Human Cells – Evaluation Study

Summary

Adhesive tapes are successfully applied in forensic casework for lifting and securing biological stains. The tape-deposited stains can be directly visualized in order to identify human cells. Until recently, the lack of efficient methods of cell isolation have restrained forensic scientists from selectively genotyping biological stains, despite satisfactory visualization. The implementation of laser microdissection technology in forensic biology opened unparalleled possibilities for micromanipulation and isolation of cellular structures. The tapes containing biological stains can be mounted directly as microdissection-ready specimens. Additionally, the dissected stains are amenable to standard analytical methods of DNA profiling, yielding good-quality profiles, which confers applicability of this technology in forensic casework.

Hereby, we present the results of a study undertaken to assess the compatibility of several commonly used laser microdissection tapes with technology. Based on evaluation of several parameters, the tapes were selected with outstanding performance in terms of effectiveness of stain collection as well as predispositions that laser-assisted micromanipulations.

Keywords biological stains, forensic tapes, adhesive tapes, laser microdissection, STR polymorphism

Introduction and purpose of the article

The adhesive properties of forensic tapes are successfully used for taking and preserving trace evidence, including fibre, hair, and fingerprints [1, 2, 3].

In the field of forensic biological research, use of adhesive film is documented as an effective and efficient method for the sampling of biological material deposited on flat surfaces, such as tissue fragments, single cells, or encrusted stains of body fluids [4, 5]. The scope of the use of adhesive film is not limited only to evidence [6, 7]. It has also been suggested that this type of material should be used as an alternative method to more invasive methods of collecting comparative biological samples [8]. An example of the adhesive film normally used in forensic biological laboratories is HAT adhesive tape (*hydrophilic adhesive tape*). The material used for the production of HAT tape used in the isolation of DNA from biological material, dissolves under the influence of water [8]. Human cells secured on the surface of the tape, after its dissolution, are immediately released into a lysis solution, which allows avoidance of mechanical cell separation and contributes to minimizing the loss of nuclear DNA during the isolation. The genetic material removed

by extraction of HAT tapes maintains quality, which allows the performance of genetic profiling using the method of polymorphism analysis of microsatellite loci (*Short Tandem Repeats*, STR), normally used by genetic labs [8, 9, 10, 11]. The mentioned properties of HAT tapes have contributed to their prevalence as a convenient complement to traditional methods of securing and profiling biological trace evidence.

However, the use of HAT tapes, especially on large surface areas of evidence is not without drawbacks. The research carried out in the Biological Research Division of the ISA Forensic Laboratory (unpublished results) shows that not in every case the complete dissolution of the hydrophilic strip is possible, and its residues can significantly hinder the process of DNA isolation. Furthermore, with an increasing size of a tested surface, the likelihood of depositing a greater number of inhibitors of DNA amplifications on the tape (e.g., dyes, humic acid), and/or mixtures of cells from different individuals, and consequently difficulties in obtaining and/or interpretating DNA profiles, also increases. Finally, the larger size of the tape parts causes problems, both of a technical (insufficient capacity of the tubes normally used) and economical nature (large reagent consumption), in the context of the nucleic acid isolation.

Laser Microdissection

In recent years, laser microdissection technology was introduced to forensics, allowing a precise acquisition of material in the form of single cells for molecular research before the due DNA analysis [12, 13, 14]. Historically, the development of laser microdissection was a response to the demand for accurate, fast, preventing contamination methods for the isolation of cells or tissue fragments from preparations. This led to the emergence of automated micromanipulators, then described by Emmert-Buck in 1996 and others [15], of the LCM (*Laser Capture Microdissection*) system, working on the basis of the laser cutting method using an infrared radiation laser beam.

Currently, the advanced systems of laser microdissection are equipped with laser modules, emitting IR or UV radiation, integrated with a high-end microscope with a motorized table [16]. The whole unit is complemented by a computer system that allows to control the cutting process using software, camera and computer monitor [17].

Originally designed for practical use in oncology, the microdissection system was quickly applied in a variety of other fields of biological sciences, including forensics [12, 13, 16, 18]. In Poland, the first implementation of laser microdissection in a forensic laboratory took place in the Forensic Laboratory of the Internal Security Agency in 2011, while the first forensic opinion based on the LCM method was issued in the same laboratory on 22 November 2013.

The use of laser microdissection in forensic sciences opens up many new opportunities, details of which are beyond the scope of this publication, including controlled isolation of certain fragments of biological material, isolation of single human cells, elimination of potential inhibitors of PCR amplification from the sample, the possibility of separation of cell mixtures coming from different individuals or different tissues, a significant increase in sensitivity analyses, etc.

As for securing forensic trace evidence, laser microdissection offers an interesting alternative to the comprehensive isolation of DNA from a swab sample (often with substrate fragments). After performing the microscopic preparation, the same trace may be subjected to selective isolation of the desired biological material by using laser microdissection. In the original application, recommended by the manufacturers of laser microdissection systems, the cell preparations are made on special polymer substrates, recommended by the manufacturer (Fig. 1a). Isolated cells or tissue fragments are then separated from the remainder of the preparation together with a part of the membrane using a laser beam. The advantage of such solution is the standardization of the preparation substrate and the possibility of selecting polymeric materials compatible with the microdissection system being in use. In the case of forensic trace evidence, normally secured with swabs or forensic adhesive films, the drawback of the above solution is connected with the necessity of transferring each time the material to the surface of the polymer membrane, which in particular poses technical problems and can lead to the loss of the material.

ISA Forensic Laboratory started a pilot research into assessing the suitability of laser microdissection system equipped with a prototype laser of increased power (see Research Method) for the isolation of human cells directly from the adhesive film used to secure the biological trace evidence. This publication is a summary of the research work which was taken to evaluate standard tapes used for forensic purposes in the aspect of the efficiency of transferring biological trace evidence from the surface of the material evidence and compatibility with laser microdissection, particularly in microscope preparations and susceptibility to laser beam cutting. During the study, a number of parameters which determine the suitability of the film surface such as viscosity, selective adhesiveness, transparency, thickness, flexibility, shearing and separation of the cut

portions were tested. Then the separated fragments of the adhesive film including human cells were used as input material in the standard procedure for gene profiling which is used in forensic laboratories, called the STR polymorphism analysis.

As a result of the research, an adhesive film of optimum performance, of high and selective adhesiveness and compatibility with the technology of laser microdissection, was selected. It was also proved that in the tested range, separated with the use of microdissection, fragments of the tape contained human cells containing genetic material that met both quantitative and qualitative requirements of standardized methods of forensic analysis, allowed for which the generation of high-quality DNA profiles.

Materials and methods

Research Material

In the research, 12 positive and negative transparent fingerprint films were examined, which are normally used for securing fingerprints, 4 adhesive mini-films for sealing 96-well reaction plates (used, inter alia, in a GeneAmp 9700 thermocycler) and, additionally, an anti-reflective film used for the protection of screen tablets. A detailed list of the test films and tapes are shown in Table 1.

Protecting Biological Trace Evidence and Making Preparations

The evidence investigated in the research, which is the subject of this publication, was a glass with visible trace evidence in the form of lip prints and fingerprints, which was supposed to contain biological trace evidence deposited by the user in the form of human cells derived from epithelial (skin) lips as well as intra- and extracellular genetic material accompanying sweat and fatty substances forming fingerprints [19]. To move the biological trace on an adhesive surface,

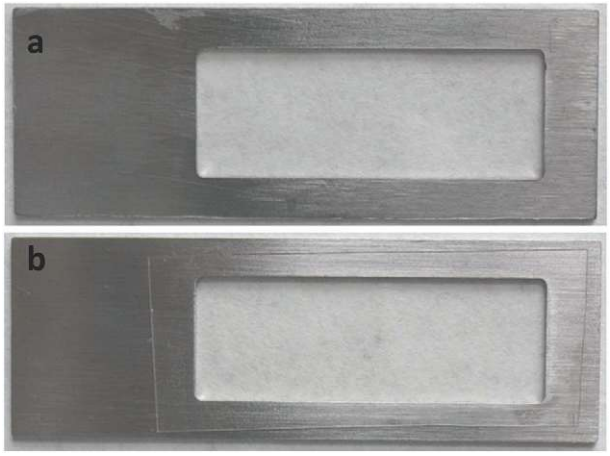

Fig. 1. Laser microdissection specimen frames with original MMI membrane (a) and user-mounted adhesive tape (b).

Table 1

Tested adhesive films and tapes. Adhesive tapes evaluated in the study

Seq. No.	Type of tape/film	Sort of tape/film	Manufacturer/proper name
1	fingerprint film	the negative transparent gel	FOTON
2	fingerprint film	the negative transparent gel	BVDA/Keep Cool
3	fingerprint film	positive transparent	BVDA
4	fingerprint film	positive transparent	STANIMEX
5	fingerprint film	positive transparent (for micro-traces)	STANIMEX
6	fingerprint film	positive transparent	FILMOLUX
7	fingerprint film	positive transparent	TESA/Film Special
8	fingerprint film	positive transparent	Neschen
9	fingerprint film	positive transparent	Neschen/FILMOLUX 609
10	fingerprint film	positive transparent	Neschen/S50
11	fingerprint film	positive transparent	Neschen/FILMOLUX Soft
12	fingerprint film	positive transparent	Sirchie
13	adhesive film	transparent	Eppendorf/Adhesive PCR film
14	adhesive film	transparent	Eppendorf/Adhesive Storage Film
15	adhesive film	transparent	Life Technologies/MicroAmp Optical Adhesive Film
16	adhesive film	transparent	PlateMax/Genuine Axygen Quality
17	anti-glare film	transparent electrostatic	3M/Natural View Fingerprint Fading Screen Protector

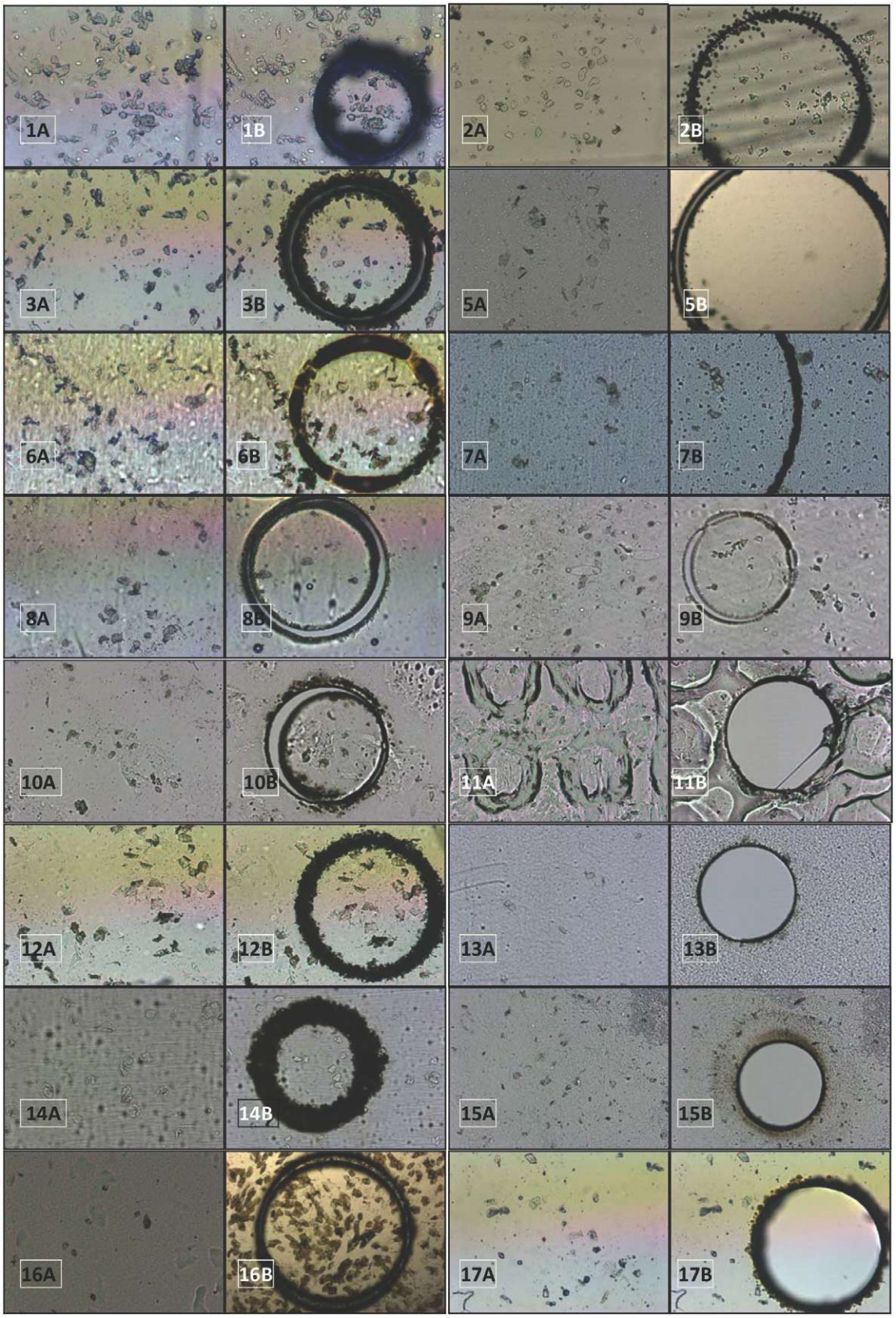

Fig. 2. Microscopic images of the tested tapes. The numbers that correspond with those given in table 1. a – images before and b – after laser microdissection.

each of the tested films were cut into size 3 × 1 cm, glued to the surface of the glass in a conspicuous place of use for about 10 seconds, then it was torn with a vigorous movement. Then the fragment of the film with the transferred biological trace was attached, by bonding the adhesive side of the tape to the edge of the metal frame MMI (Molecular Machines & Industries) of the length of edge of 7 x 2.5 cm, and the dimensions of the working aperture of 45 x 16 mm (Fig. 1b).

Microscopic Imaging and Laser Microdissection

The imaging of specimens was carried out using an optical microscope equipped with a confocal Nikon A1-R and NIS-Elements software (Nikon). The specimens were observed in transmitted light, using black-and-white CCD digital camera Nikon QI-1 with cooling of the resolution of 1280 x 960, 10x Plan Fluor lens N.A.0,30, w.d. 16 mm and 20x Plan Fluor NA.0,45, w.d. 1.0 mm. Microscopic images of each film (except for film no. 4 – see Results and Discussion) is shown in Figure 2.

Cutting fragments of the adhesive films was performed by using the laser microdissection module of Molecular Machines & Industries (MMI) integrated with Nikon microscope A1-R. The module was equipped with a pulse, picosecond solid-state laser of the power of 4 mW, emitting radiation in the UV spectrum range (of the wavelength of 355 nm). The frames with film fragments were attached to the motorized microscope stage operating as an internal control module software of CellCut microdissection integrated with MMI CellTools platform.

The laser microdissection of specimens was carried out in the transmitted light, using the high-internal digital CCD MMI FI1C camera of the resolution of 1032 x 776, in which the MMI CellCut module is equipped, and a 10x Plan Fluor NA.0,30, w.d. lens 16 mm and 20x Plan Fluor NA.0,45, w.d. 1.0 mm, using the variable parameters of the laser power and the focal length of the laser beam. The separation of fragments followed under the influence of the laser beam due to the effect of cold ablation. The separate fragments were placed in a non-contact manner in 0.2 ml tubes, using proprietary technology of automatic feeder ADF developed by MMI company for the microdissection module line of CellCut type (www.molecular-machines.com).

Criteria for the Evaluation of Adhesive Films for Forensic Purposes

The film evaluation included the following criteria: transparency/visibility of cells, selective adhesion (film performance in terms of adhesion of human cells), viscosity, flexibility, cutting susceptibility and the thickness. The first two parameters were evaluated by microscopic observation. Viscosity and elasticity was estimated in the organoleptic manner. The cutting susceptibility resulted from type of response of the film surface to the laser beam. The measuring of the thickness of each film (expressed in mm) was made using an electronic thickness gauge LITEMATIC VL-50 of Mitutoyo Company, of the resolution of 0.01 microns, equipped with the measuring system that minimizes the error "Abbe" and the mechanism for reducing the measurement pressure. Due to the subjective nature of most of the implemented criteria, a point rating scale was adopted of the 0-5 point range, where "0" expressed the value of the worst, such as the lack of cutting susceptibility of the film, while "5" the best value, e.g. very high susceptibility to cutting (Fig. 3). By summing the points for each parameter, those films were considered unsuitable which, at least for one of the criteria, scored less than 3 points, while the remaining were ranked in terms of the usefulness, in agreement with their final scores. No points were given for thickness, which was expressed in the metric values.

STR Polymorphism Analysis

To determine the STR polymorphisms from biological material (mouth swab), deposited on the three selected films: 3M/Natural View Fingerprint Fading Screen Protector, PlateMax/Genuine Axygen Quality, positive transparent/Neschen, which received the highest rating point (Table 3), the researchers separated, with the use of laser microdissection, fragments containing about 20 cells. After microscopic inspection of the adhesion lid and verification of the number of cells, the material in the cut portion was subjected to the DNA isolation process, taking advantage of the phenol method. Isolated DNA was suspended in the total volume of 5 mL H_2O, and the mixture was used as DNA template in the amplification reaction. DNA amplification was performed using the SELECT AmpFISTR ® NGM™

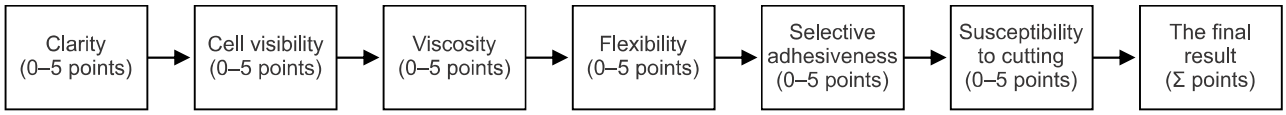

Fig. 3. Evaluation criteria applied to assessment of tapes usability.

Table 2

The results of the point evaluation of the adhesive film parameters
Results of evaluation for scored criteria of adhesive tapes

Seq. No.	Type of film	Clarity	Visibility of cells	Viscosity	Thickness [mm]	Flexibility	Selective adhesiveness	Susceptibility to cutting	Sum of points
1	FOTON gel negative transparent	4	5	4	0.56	2	5	0	**20**
2	BVDA, Keep Cool gel negative transparent	5	5	4	0.59	2	5	3	**24**
3	BVDA positive transparent	5	5	2	0.12	1	2	1	**16**
4	STANIMEX positive transparent	0	0	4	0.15	3	0	0	**7**
5	STANIMEX positive transparent for micro-traces	4	5	5	0.14	1	4	4	**23**
6	FILMOLUX positive transparent	3	3	5	0.11	5	3	1	**20**
7	TESA Film Special positive transparent	3	3	4	0.07	4	3	3	**21**
8	Neschen positive transparent	4	5	4	0.09	4	4	4	**25**
9	Neschen FILMOLUX 609 positive transparent	4	3	4	0.13	4	4	4	**23**
10	Neschen S50 positive transparent	5	4	3	0.05	5	3	3	**23**
11	Neschen FILMOLUX soft positive transparent	2	2	3	0.11	5	2	4	**18**
12	Sirchie positive transparent	4	4	4	0.15	2	4	3	**21**
13	Adhesive PCR film Mini-film	3	3	2	0.11	5	2	4	**19**
14	Adhesive Storage Film Mini-film	3	4	4	0.21	4	4	1	**20**
15	MicroAmp Optical Adhesive Film Mini-film	3	3	3	0.11	4	3	4	**20**
16	Axygen Genuine Quality (PlateMax) Mini-film	5	4	3	0.09	4	5	4	**25**
17	3M/Natural View Fingerprint Fading Screen Protector	5	5	4	0.17	3	4	4	**25**

PCR Amplification Kit (Applied Biosystems), using 31 cycles of reproduction, i.e. the amount higher for samples with low DNA content (called Low Copy Number - LCN). The total volume of the reaction mixture was 12.5 μl. The amplicon division by the method of capillary electrophores was performed on the ABI 3130 sequenator. STR polymorphism analysis was performed using the GeneMapper ID-X software. The obtained DNA profiles were compared with the reference profiles (DNA_{ref}) which formed

those known, previously identified, involving the use of standard methods (DNA column isolation of the mouth swab), profiles of three donors of the biological trace evidence used in the experiments.

Determination of the Amount of DNA

In order to assess the DNA content in the isolates derived from human cells after laser microdissection, the whole isolate, obtained from 20 isolated cells (in the volume of 2 μl), was evaluated in terms of the DNA amount, using Quantifiler Duo DNA Quantification Kit system from Applied Biosystems, the 7500 Real Time PCR System Applied Biosystems and the SDS Software v1.2.3 software from Applied Biosystems, according to the manufacturer's instructions. The research was carried out in three repetitions, in the presence of the positive control in the form of DNA of a given concentration (AmpFISTR® Control DNA 007), diluted to working concentration of 100pg/ul.

Results and Discussion

The evaluation results of particular parameters with the detailed scores was shown in Table 2. The microscopic images of the film surfaces used to secure biological trace evidence are shown in Figure 2. The exception is the film no. 4 (positive fingerprint film - STANIMEX), in which case the microscopic images could not be obtained, because of the minimum transparency.

The adequate transparency of the film and good cell visibility are some of the key criteria for determining the suitability of the adhesive film for forensic purposes for the direct use as the microscopic specimen subjected to imaging and laser microdissection. The specimens were visualized in transmitted visible light to determine the required light beam permeability for the film surface. In order to ensure good permeability, transparent films were selected for the research, which in most cases resulted in satisfactory transparency of specimens and good visibility of the secured cells. The exceptions were the films no. 4 and 11, which received less than 3 points in both evaluation criteria, which is below the threshold of acceptability. A particularly favourable surface structure, good clarity and cell visibility were the features of all negative films, positive films no. 3, 5, 8 and 10, mini-film no. 16 and anti-reflection film no. 17.

Another key parameter determining the suitability of the film in the laser microdissection technology is its susceptibility to laser beam cutting (column: Cutting susceptibility – table 2). There was no observable relationship between cutting susceptibility and other parameters being the matter of the evaluation. This made it possible to conclude that the key factor for this parameter is fibre composition of the material of which the film was manufactured. Due to the fact that detailed lists of the components of particular films are usually covered by corporate secret, it was not possible to determine the optimal material for laser microdissection. Therefore, the research focused on the evaluation of the reaction of film surface to laser radiation. The highest score was given in the case of separation of the film fragment by minimal amount of repetitions of the laser beam, being led with the minimal laser power, with the narrow cut edges and, simultaneously, in the absence of the adverse effects of "bubbling" of the dissolved glue, cutting edge scorching and melting of the film surface by the laser beam. Such separation was possible to achieve for films no. 5, 8, 9, 11, 13, 15, 16 and 17. The lack of the highest score (5) resulted from the fact that in none of the tested films it was possible to entirely separate the cut area by the application of a single laser beam.

The viscosity criterion relates to the overall adhesive properties of the film, i.e. the durability of its adhesion to various types of surfaces. In this study, glass and paper (with no traces of biological deposits) surfaces were evaluated, while the given scoring is the average score for both types of surfaces. In the case of paper, the films with the highest score, which in a sustainable manner (i.e. without falling off) adhered to the evaluated surfaces, while at the same time were relatively easy to peel off, i.e. without breaking the lignin fibres of the paper. The highest scores in this category were received by the positive films no. 5 and 6; however it was not a characteristic feature of the entire category. The scoring below the threshold of acceptability was obtained by films no. 3 and 13.

The film flexibility does not directly determine its suitability for laser microdissection. However, this parameter was included in the total score due to the variety of shapes and physical features of the material evidence encountered in practice, making the manipulation of less flexible film difficult. This parameter also directly translates into the ease of preparing the microscope specimen. The worst scoring was obtained by the positive gel films (no. 1 and 2) and the positive films no. 3, 5 and 12, which undoubtedly is related to the large thickness of these films.

The selective adhesiveness of forensic films for, i.e. the ability to bind to human cells, determines the validity of its use as a method of preserving biological trace evidence. This parameter is not directly conditioned by the viscosity of the overall film, but rather due to the affinity of the adhesive substance

Table 3

The list of films rated above the threshold of acceptability for each of the tested parameters
A list of tapes with scoring exceeding acceptance threshold in all criteria

Film No.	Type of film	Sort of film	Manufacturer/Model	Sum of points
17	anti-glare film	transparent electrostatic	3M/Natural View Fingerprint Fading Screen Protector	25
16	adhesive film	transparent	PlateMax/Genuine Axygen Quality	25
8	fingerprint film	positive transparent	Neschen	25
9	fingerprint film	positive transparent	Neschen/FILMOLUX 609	23
10	fingerprint film	positive transparent	Neschen/S50	23
7	fingerprint film	positive transparent	TESA/Movie Special	21
15	adhesive film	transparent	Applied Biosystems/MicroAmp Optical Adhesive Film	20

used for organic matter, including human cells. Most efficient for the transfer of biological material were gel negative films (1 and 2) and mini film no. 16. It should be noted that in the research carried out, the selective adhesiveness was of a relative parameter nature, i.e. the obtained evaluation depended on the score in the categories of *transparency* and *visibility of the cells.* This can particularly be noted in film no. 4, in which, due to low transparency (0 pts) and visibility of the cells (0 pts), evaluation of the selective adhesiveness was not possible (0 pts). For the purposes of this research, having the aim of assessing the range of films for their practical use in laser microdissection technology, the inability to asses a parameter (e.g. *selective adhesiveness* – film no. 4) translated into the lowest score, which meant a film incompatibility with the technology.

In addition to the scoring parameters, two additional criteria were determined, such as the film thickness and surface structure. In the case of the thickness, its value had no direct impact on the usefulness of film, remaining at the same time, in indirect relationship with other parameters, such as flexibility, transparency and susceptibility to cutting. On that account, this parameter, expressed in a metric scale, was inserted into the tests for information only. The structure of the surfaces of tested films was evaluated based on microscopic images juxtaposed in Figure 2. Due to the large variety, this parameter proved very difficult to copy in a point scale. Also, it was found that the usefulness of the film is much more accurately expressed by scoring parameters of transparency and cell visibility.

Table 3 shows the films that were rated above the threshold of acceptability, i.e. 3 or more points for all the test criteria. The results of the performed tests allowed the conditional classification of seven out of all the tested films as being useful to secure biological trace evidence and being compatible with laser microdissection. This group contained were positive films (7–10), mini-adhesive films (15, 16) and anti-reflection film (17). Both the type and nature of the film, however, does not determine its suitability for microdissection. Attention should be paid to the Neschen products which in three out of the four cases were found to be compatible. However, when choosing for the further research was considered, solely a detailed assessment of all tested parameters expressed by the total amount of points scored by the different film models was taken into account.

For the purpose of the research which consisted in genotyping of the biological material secured on the surface of the film and then separated by means of laser microdissection, three types of film were classified to receive the highest number of points in the usability test, i.e. foils no.17, 16 and 8 (indicated by the grey shade in Table 3). Then, from each of the classified film, fragments comprising a total of approximately 20 of human cells (Fig. 4) were separated, in three repetitions representing biological material from three different donors. The

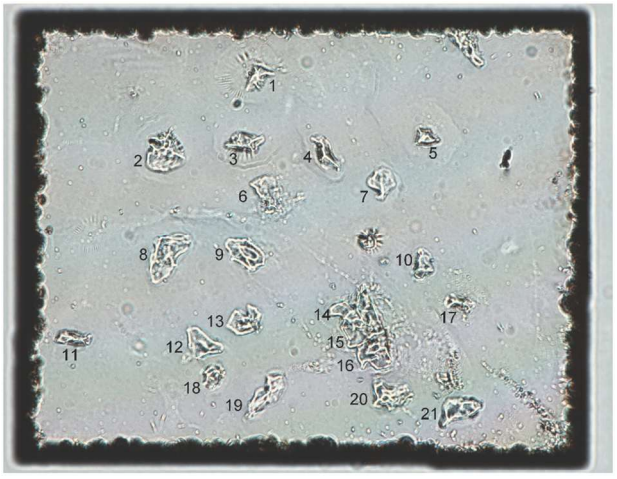

Fig. 4. Microscopic image of the microdissected forensic tape (3M) with epithelial cells. The numbers enumerating the individual cells with entire perimeter within the field of view.

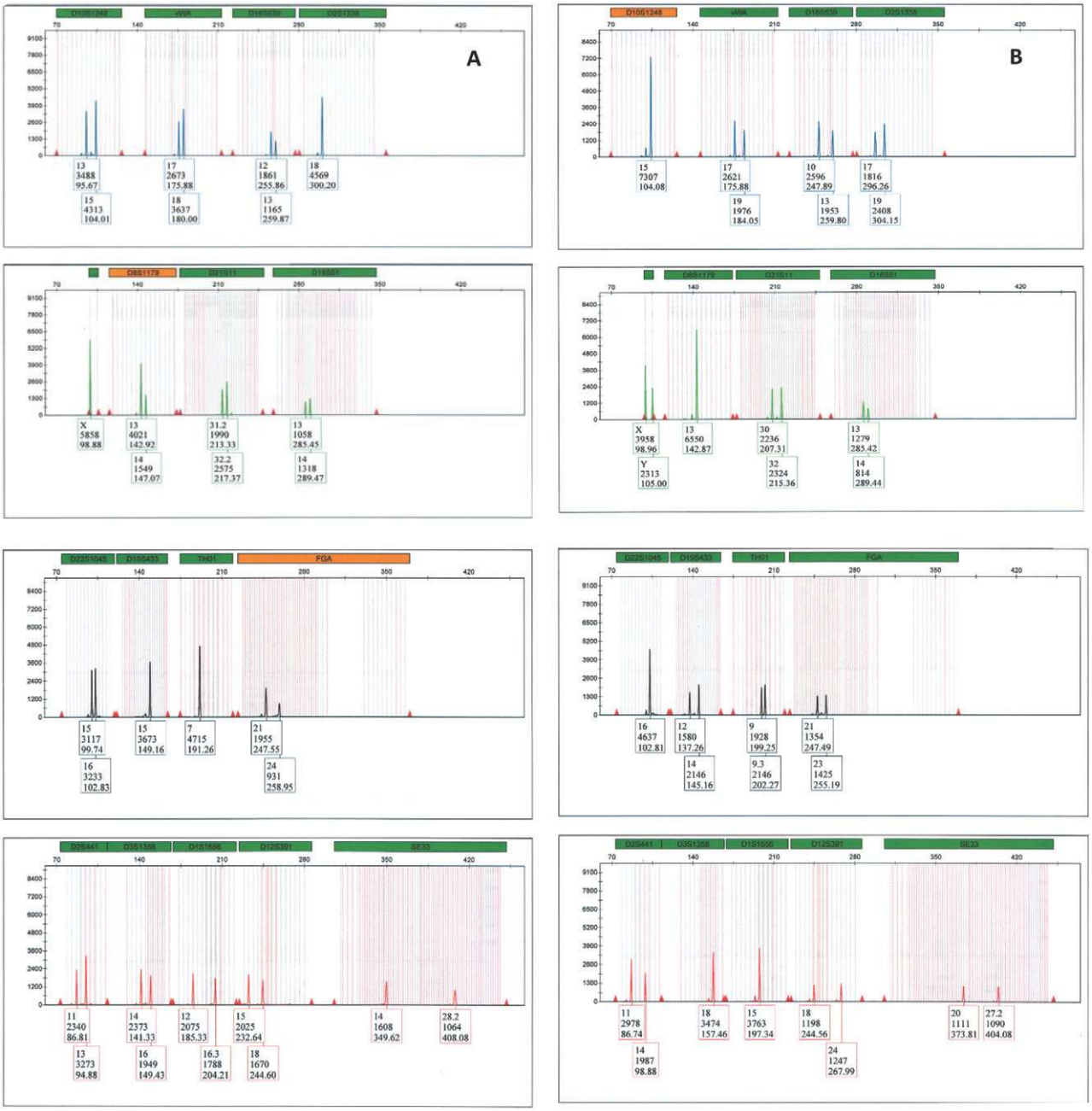

Fig. 5. Genetic profiles Obtained from biological stains containing buccal cells, collected about a Natural View Fingerprint Fading Screen Protector (3M), laser microdissection is subjected, phenol DNA isolation protocol and DNA amplification with NGMSElect ™. The Y-axis scaled in rfu units. Panels A and B present exemplary profiles of two different donors.

obtained fragments were used as the input material in the standard procedure of DNA isolation, PCR amplification and capillary electrophoresis used by forensic laboratories (procedure described in the Materials and Methods section).

As a result of measuring the amount of DNA in three independent samples from different donors, each containing about 20 cells, the values were obtained, respectively, 40 pg, 66 pg, 30 pg DNA. These values were lower by about half of the expected values, resulting from the literature data with regard to the content of DNA in the cell nucleus. According to the accessible knowledge, 20 diploid cell nuclei comprises DNA in the amount of 132 pg. Thus, taking into account the 30% loss of genetic material during the isolation of phenol, isolates should contain approximately 90 pg of DNA. The lowered value of the measurement was also obtained for the control sample (the expected

value resulting from the applied dilution - 100 pg, the measured value - 50 pg). The observed discrepancies may result from the fact that the concentration of DNA being the matter of the analysis oscillate in the range of the lowest measuring point of the standard curve of Quantifiler Duo DNA Quantification Kit set, which means that the sensitivity of the method may be insufficient for a reliable measurement. In addition, according to the authors' knowledge, there are currently no methods of measurement that could allow to precisely estimate the amount of DNA of several or anywhere from ten to twenty of picograms. As the consequence of the above-mentioned results of the experimental research into profiling the single human cells, separated with the use of laser microdissection, the quantitative designation by Real Time PCR were abandoned. For the purpose of estimating the amount of DNA in the isolates of 20 cells, it was assumed that they contained between 90 pg (6.6 pg x 20 cells - 30% of losses during isolation) and 45 pg of DNA (average value of the indications resulting from the method of the Real Time PCR).

In the analysed quantitative scope, the combination of the laser microdissection technique with the use of the selected films no. 17, 16 and 8, enabled to obtain full STR profiles that meet the analytical requirements resulting from the intra-laboratory validation of the Research Office of Forensic Research of The Internal Security Agency and the recommendation of the International Society for Forensic Genetics – IFSG (the sample profiles for 3M film were shown in Fig. 5). Furthermore, for all the analysed films and repetitions, the compatibility of the labelled profiles with the DNA_{ref} donors' reference profiles was found, in the full extent of the analysed STR loci included in the NGMSElect™ set. The obtained results confirm that the films, used for securing biological trace evidence, can successfully be used, directly as microscope specimens that are compatible with the laser microdissection system, and with standard methods of gene profiling.

Summary

Implementation of laser microdissection to forensic sciences created new possibilities for genetic profiling of biological trace evidence traditionally secured on swabs and forensic adhesive films. In addition, besides currently used global DNA isolation from secured material, experts have gained the opportunity of typing and selective sourcing of certain components of trace evidence, especially human cells, in an automated manner and free from contamination. In order to further simplify the process of genotyping biological trace evidence, a number of the adhesive films was tested in terms of their suitability for securing evidence and their compatibility with laser microdissection technology. As a result of these tests, films of optimum parameters were selected, ensuring effective transfer of organic material, in particular human cells, from the surface of evidence, the possibility of direct microscopic imaging of the cells and susceptibility to separation of the specimen fragments with the use of laser beam. Regarding the growing interest in laser microdissection technology, the presented results of the research can significantly contribute to the popularization of the use of the forensic adhesive films, with the directly secured material as microscopic specimens adapted for microdissection. The proposed method of using the adhesive film in forensics will enable selecting and sourcing any areas and components of the specimen, and consequently, more conscious, controlled and selective genotyping of biological material derived from secured trace evidence.

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Source

Fig. 1–5: authors
Tab. 1–3: own elaboration

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