STANDARD TWINNING PROJECT FICHE

1. Basic Information

- 1.1 Publication notice reference: EuropeAid/ 137-991/IH/ACT/HR
- 1.2. Programme: Transition Facility IPA/2013/24986 (Annex of C(2013) 8057 final); Institution Building Envelope
- 1.3 Twinning Number: HR 14 IPA JH 05 16
- 1.4 Title: Implementing Next Generation Sequencing (NGS) technology in DNA forensic science laboratory (CRO NGS)
- 1.5 Sector: Justice and Home Affairs
- 1.6 Beneficiary country: Republic of Croatia

2. Objectives

2.1 Overall objective:

Capacities of the Ministry of Interior enhanced to effectively combat crime within the EU and international environment, in line with the related EU policies and strategies.

2.2 Project purpose:

Procedures for implementing and using of NGS technology developed and forensic DNA experts trained in order to upgrade efficiency of Forensic Science Centre (FSC) "Ivan Vučetić" of the Ministry of Interior (MoI) to effectively combat crime.

2.3 Contribution to Accession Treaty/Relevant national documents:

Accession Treaty

Following the signature of the Accession Treaty on 9 December 2011 and its ratification procedure in the Member States, Croatia joined the European Union on 1 July 2013 as the 28th Member State. This Twinning project is in compliance with general objectives set in the Accession Treaty. The project purpose is in line with one of the specific commitments undertaken by the Republic of Croatia in the accession negotiations (Annex VII of the Accession Treaty): To continue to ensure a sustained track record of substantial results based on efficient, effective and unbiased investigation, prosecution and court rulings in organized crime and corruption cases at all levels including high level corruption, and in vulnerable sectors such as public procurement.

The Treaty on the Functioning of the European Union

This Twinning project is in line with the objectives of the Treaty on the Functioning of the European Union: The Union shall establish police cooperation involving all the Member States' competent authorities, including police, customs and other specialized law enforcement services in relation to the prevention, detection and investigation of criminal offences. (...) European Parliament and the Council (...) may establish measures concerning:

(a) the collection, storage, processing, analysis and exchange of relevant information;

- (b) support for the training of staff, and cooperation on the exchange of staff, on equipment and on research into crime-detection;
- (c) common investigative techniques in relation to the detection of serious forms of organized crime.

Strategic plan for the Ministry of Interior and other institutions involved in protection and rescue for the period 2016 - 2018

This Twinning project is in accordance with the objective *Decrease criminal behavior* which inter alia states the following goals: to reduce the risk of criminal behaviour, to improve prevention of criminal behaviour and to improve crime prevention. Among methods of achieving the abovementioned goals are: improving the work of the criminal police, improving the effectiveness of combating corruption and organized crime and strengthening cooperation between the police and the judiciary.

Through implementation of up-to-date practices and methods, this Twinning project will increase the quality of forensic work in FSC "Ivan Vučetić" thus ensuring more effective crime combat within both national and EU environment.

3. Description

3.1 Background and justification:

In 1980s DNA analysis was introduced in forensic laboratories. Soon it has become an indispensable tool in forensic identification and an inevitable component of criminal investigations. Although standard methods of short tandem repeat (STR) typing are improving continuously, the basic principle of DNA identification in forensics remained unchanged until recently, when NGS technology was introduced. Today, numerous EU Member States possess necessary equipment and work on protocol optimization with the final goal of implementing NGS technology in forensic science and eventually routine casework. In that respect, establishing NGS procedures and training of employees would enable Croatia to participate and adequately contribute to the EU efforts in fighting crime.

FSC "Ivan Vučetić" is equipped with MiSeq desktop sequencer upgraded with FGx (forensic genomic system), which enables forensic DNA testing *via* simultaneous typing of 27 autosomal STRs, 24 Y-chromosome STRs, 7 X-chromosome STRs, 94 identity single nucleotide polymorphisms (SNPs), 22 phenotypic SNPs, and 56 biogeographical/ancestry SNPs. Research platform of the mentioned instrument enables, among other applications, sequencing of either whole mtDNA genome or its D-loop hypervariable regions, which is an acknowledged tool in forensic identification. The aim of this project is therefore the transfer of knowledge to resident DNA experts in order to capacitate them to fully employ NGS instrument and protocols for forensic typing of STRs, SNPs and mtDNA in scientific DNA laboratory. It will enable the Republic of Croatia to join the other EU Member States in global efforts of introducing NGS technology into routine forensic practice. Moreover, implementation of NGS procedures will enable building of national reference databases to which comparison of casework profiles will be made and the rarity of profiles determined.

Forensic DNA analysis is confronted with several challenges, including low-copy-number template, highly degraded and contaminated samples, the need of high accuracy and reproducibility, as well as time and cost considerations. Currently, forensic profiling is mostly based on polymerase chain reaction (PCR) and capillary electrophoresis (CE) - fragment analysis; methods employed to detect length variations in short tandem repeat (STR) markers. At the moment, more than 60 countries worldwide have established forensic STR DNA databases that grow continuously. To avoid random matches between unrelated individuals, it is recommended to incorporate more STR markers within databases (currently, up to 15 STR markers are routinely used). However, simultaneous detection of more STR markers would be very difficult due to technical limitations of fluorescence-based CE instruments that are currently used. Namely, alleles of identical or similar length, but of different

sequence cannot be distinguished. This problem occurs in everyday routine work, especially with STR mutations in complex paternity cases and analysis of complex DNA mixtures containing DNA from more than two individuals.

NGS technology has many advantages over PCR-CE based approach in forensic identification: high throughput, low cost, simultaneous detection of large number of relevant loci (SNPs in addition to STRs), detection of nucleotide variants within STR sequences, the ability to distinguish alleles of similar length and to extract maximum information from highly degraded DNA. NGS technology can significantly facilitate the identification of mixed DNA samples and analysis of complex kinship cases, thus increasing the efficiency and cost-effectiveness of legal cases. In addition to STR typing, NGS technology enables detection of SNPs which are informative for individual's identity, phenotype and ancestry. Moreover, it enables simultaneous analysis of different genetic targets such as autosomes, sex chromosomes and mtDNA, and also has the potential of introducing applications in other aspects of research, such as monozygotic twin studies, body fluid determination, species identification and forensic animal and plant analysis.

Y chromosome STRs (Y-STRs) are commonly used to distinguish male component of DNA mixtures when a high female background is present or to reconstruct paternal relationships between male individuals. Likewise, X chromosome STRs also have the potential to complement the analysis of other genetic markers, especially in kinship testing and solving complex forensic cases.

SNP is the simplest type of polymorphism characterized by a single base difference in the DNA sequence. SNPs have recently become the marker of choice for many genetic studies. In forensic cases when neither a suspect nor a hit exists, additional information from evidence samples might provide valuable clues regarding the phenotype of the offender. To be able to predict skin, eye and hair color, even a facial structure based only on biological material left behind at a crime scene or from a disaster victim, is one of the major expectations from forensic genetics in the future. Combined data from genotyping multiple SNPs can yield exceedingly high power of discrimination. Most importantly, since SNP genotyping is beyond standard PCR-CE workflow, NGS technology will enable introduction of this type of analysis into FSC "Ivan Vučetić" laboratory practice.

Demands for mitochondrial DNA (mtDNA) testing are constantly increasing due to a large number of missing person cases and natural disasters. It has proved to be a useful tool in cases involving low amount or degraded genomic DNA or wherein the maternal lineage needs to be investigated, due to its characteristics: small genome size, multiple copies, maternal inheritance, high mutation rate and lack of recombination. The old system of Sanger sequencing is slow and has a low throughput. Additional polymorphic loci are required to increase the power of discrimination in identification. Research has shown that whole mitochondrial genomes can resolve some cases where common mtDNA types cannot distinguish between individuals. Therefore, NGS technology has the potential to greatly assist the analysis of whole mitochondrial sequences, detecting low level of mtDNA heteroplasmy and facilitate interpretation of mixtures containing biological material from more than one person.

Twinning project activities are designed in order to result with fully operating NGS forensic laboratory, including instrument validation, human mtDNA protocol validation and ForenSeq DNA protocol validation. Within the Twinning project on-the-job trainings of the DNA experts will be focused on validation procedures, sequencing protocols, preparation of both reference (buccal swab, blood) and challenged casework samples for sequencing, analysis and interpretation of sequencing data. Moreover, it is planned that internships for four resident DNA experts will be organized in laboratories with the fully established NGS workflow. Internship within the environment where the forensic STR / SNP and mtDNA methods are validated and fully employed, will give the experts necessary insight into overall NGS process, from DNA isolation to data interpretation. It will give them the opportunity for integral training *in situ* with the team of experts experienced in different fields, covering entire NGS workflow.

FSC "Ivan Vučetić" is fully equipped for the above mentioned activities (instruments for DNA extraction and purification, instruments for DNA quantification, thermal cyclers, gel electrophoresis equipment with gel imager, MiSeq desktop sequencer with FGx upgrade and all necessary consumables). Resident DNA experts are experienced forensic and molecular biologists, proficient in standard molecular biology techniques.

The project activities will enable complete and successful implementation of NGS methods into forensic science DNA laboratory which will ensure obtaining the maximum amount of information from challenging evidence samples, or known reference samples, thus improving the work of the criminal police and effectiveness in combating crime.

3.2 Linked activities:

Transition Facility "Biometric face identification"

This Twinning light project is currently in preparation and it is expected to be circulated in 1st quarter of 2016. The purpose of this project is to increase capacities of Forensic Science Centre (Department of dactyloscopy and identification) and its forensic experts in the field of biometric face identification.

Transition Facility "Advanced biostatistics in routine forensic DNA casework"

This Twinning light project is currently in preparation and it is expected to be circulated in 2nd quarter of 2016. Project purpose is to develop procedures for implementing and using of advanced statistical programs for DNA mixtures and familial/relatives search and train forensic DNA experts in order to upgrade efficiency of forensic science DNA laboratory (MoI) to effectively combat crime and to consequently increase cooperation between police and judiciary.

IPA 2013 "Strengthening capacities of the Ministry of the Interior to implement the automated exchange of DNA and dactyloscopic data (CRO DNA/DKT)"

The project is currently in preparation phase and it is expected to be circulated in 1st quarter of 2016. Overall project consists of two components: Twinning and Supply. The purpose of the Supply component is to procure and install automated robotic equipment for DNA analyses and workstations for input of dactyloscopic data, and to educate the staff on usage of the procured equipment. The purpose of the Twinning component is to develop procedures for the automated exchange of DNA and dactyloscopic data and to train forensic science experts in order to establish preconditions for implementation of Prüm Decisions.

Transition Facility "Disaster victim identification in natural and accidental disasters and terrorism acts" (CRO DVI)

This Twinning project (HR 14 IPA JH 04 16) is currently in circulation and it is expected to start in 4th quarter of 2016. The purpose of the project is to improve efficiency of Forensic Science Centre (FSC) "Ivan Vučetić" in order to provide fast and effective response in case of mass disaster incidents (MDI) through acquisition of knowledge and skills as well as experience exchange within the international environment.

TAIEX "Statistical evaluation of forensic DNA evidence using LRmix program"

This study visit to University of Copenhagen, Department of Forensic medicine, Dennmark, was conducted in March 2015. The purpose of the study visit was to collect basic knowledge of forensic DNA mixture interpretation using free statistical software program LRmix, which has consequently improved knowledge of FSC "Ivan Vučetić" experts on statistic calculation and interpretation of forensic DNA mixtures in DNA laboratory of FSC "Ivan Vučetić".

IPA 2011 "Strengthening capacities of the Ministry of the Interior in combating cybercrime" (HR/11/IB/JH/01, Twinning partners: Spanish/Austrian consortium)

This Twinning project started in June 2015 and finished in January 2016. The following results were achieved: Standard Operating Procedures for collection, examination, analysis and reporting on electronic evidence according to ISO 17025 standard and ENFSI procedures developed, Training programme and training manual, including training of trainers programme and corresponding materials, on forensic cybercrime investigation for forensic science experts prepared, Capacity of forensic science experts on providing cybercrime forensic services strengthened, Comprehensive Assessment Report with recommendations for improvement of the system for fighting cybercrime prepared and Standard Operating Procedures for cybercrime investigations drafted, Proposal of the new organisational model of the Criminal Police Directorate and of Police Administrations - PA Zagrebačka, PA Splitsko-dalmatinska, PA Primorsko-goranska, PA Osječko-baranjska, PA Istarska prepared and presented to the General Police Directorate management, Capacity of the Criminal Police Directorate for fighting cybercrime strengthened and Capacity of the Police Academy to provide training on fighting cybercrime enhanced.

3.3 Results:

Result 1: Validation study for NGS instrument conducted and protocols according to ISO 17025 standard procedures developed

Indicators of achievement:

- Validation study of the forensic NGS platform for forensic STR¹ and SNP typing conducted, protocol developed and validation report prepared
- Workshop with the purpose to present validation report for performed validations on forensic NGS platform for forensic STR and SNP typing with at least 5 participants conducted
- Validation study of the research NGS platform for mtDNA sequencing conducted, protocol developed and validation report prepared
- Workshop with the purpose to present validation report for performed validations on research NGS platform for mtDNA sequencing with at least 5 participants conducted
- On-the-job training for at least 3 DNA experts related to conducting independent validation for NGS instrument and developing protocols organized and conducted
- On-the-job training for at least 1 DNA expert related to biostatistic and bioinformatic data analysis and interpretation of NGS results organized and conducted

Result 2: Standard Operation Procedures for DNA analysis on NGS instrument prepared

Indicators of achievement:

• Standard Operation Procedures for:

- o forensic STR and SNP typing
- o mtDNA sequencing
- o data analysis and interpretation of forensic STR and SNP typing results
- o data analysis and interpretation of mtDNA sequencing results

drafted

 Pilot implementation of DNA analysis on NGS instrument according to the drafted Standard Operating Procedures in FSC "Ivan Vučetić" conducted and report with recommendations prepared

¹ All references to STR typing in indicators of achievement of the Twinning project refer to X-STR, Y-STR and autosomal STRs.

- Pilot implementation of data analysis and interpretation of NGS results according to the drafted Standard Operating Procedures in FSC "Ivan Vučetić" conducted and report with recommendations prepared
- Final Standard Operation Procedures based on the recommendations resulting from pilot implementation of drafted Standard Operation Procedures and round table discussion with at least 5 participants prepared
- Workshop for at least 10 participants with the purpose to present Standard Operation Procedures organized and conducted

Result 3: Capacity of forensic DNA experts in the area of NGS methods strengthened

Indicators of achievement:

- Training need analysis (TNA) related to NGS methods conducted and TNA report prepared
- Long-term training programme² and training manual prepared
- On-the-job training in the BC for at least 6 forensic DNA experts conducted
- Internship for 4 participants in the MS (in duration of 8 weeks for 2 forensic DNA experts and 4 weeks for 2 forensic DNA experts) organized and conducted and internship report prepared

NOTE: Activities of the project will be implemented using equipment of the FSC "Ivan Vučetić", which is based on sequencing-by-synthesis technology (Illumina MiSeq desktop sequencer upgraded with forensic genomic system (FGx)).

3.4 Activities:

Member State(s) is kindly requested to develop activities in the submitted proposal which are needed in order to achieve results stipulated in the fiche.

Minimum two visibility events will be organized in the course of the implementation of the project; Kick-off meeting at the start of the implementation and the Final meeting at the end of the implementation of the project activities.

3.5 Means/ Input from the MS Partner Administration:

MS Project Leader may participate in the project also as the short-term expert (STE) and in this case the MS Project Leader should satisfy requirements stipulated in the fiche for both the Project Leader and the relevant STE profile.

3.5.1 Profile and tasks of the Project Leader

Profile of the Project Leader

Requirements:

- University level education or equivalent professional experience of 10 years in forensics
- Minimum 5 years of experience in forensics
- Working level of English language

² It is envisaged that activity will include training modules on following topics: forensic STR / SNP typing and mtDNA sequencing using sequencing-by-synthesis technology along with data analysis and interpretation of sequencing results.

- Proven contractual relation to public administration or mandated body, as defined under Twinning Manual 5.4.5
- Computer literacy
- Experience in project management

Assets:

- Experience in international police cooperation
- Experience in the field of DNA forensics

Tasks of the Project Leader:

- Overall coordination and co-managing the implementation of the project in cooperation with the BC Project Leader
- Ensuring sound implementation of the envisaged activities and, if necessary, proposing remedial actions
- Coordinating the MS experts' work and availability
- Project reporting
- Ensuring backstopping and financial management of the project in the MS
- Supervising and coordinating implementation of the project
- Providing efficient leadership of the project
- Participation in Steering Committee meetings
- Organization of internships

3.5.2 Profile and tasks of the RTA

Profile of the Resident Twinning Adviser

Requirements:

- University level education or equivalent professional experience of 10 years in forensics
- Minimum 3 years of experience in forensics
- Working level of English language
- Proven contractual relation to public administration or mandated body, as defined under Twinning Manual 5.4.5
- Computer literacy
- Experience in project management

Assets:

- Experience in the field of DNA forensics
- Experience in implementing NGS methods
- Experience in international police cooperation
- Experience in conducting trainings

Tasks of the Resident Twining Adviser:

- Support and coordination of all activities in the BC
- Day to day management of the project in the beneficiary institution
- Providing technical advice on EU policies and best practices, and assisting Croatian administration in the context of project work plan
- Coordination of the short-term experts activities
- Monitoring project implementation of the project, budget spending and proposing corrective actions, if required

- Executing administrative issues (e.g. assisting in reporting)
- Organization of visibility events (kick-off and final event)
- Organization of PIU and Steering Committee meetings
- Networking with stakeholders of the project in Croatia and in MS

The duration of the RTA secondment is 12 months.

3.5.3 Profile and tasks of the short-term experts

For each of the proposed experts in the submitted proposal the Member State(s) is kindly requested to indicate the expert's profile.

Profile of the Short-term expert 1 (STE 1)

Requirements:

- University level education or equivalent professional experience of 8 years in the field of molecular biology
- Minimum 3 years of experience in DNA sequencing
- Working level of English language
- Proven contractual relation to public administration or mandated body, as defined under Twinning Manual 5.4.5
- Computer literacy

Assets:

- Experience in the field of forensics
- Experience in forensic STR and/or SNP typing by NGS
- Experience in NGS of mtDNA
- Experience in implementing sequencing-by-synthesis technology
- Experience in preparation of procedures for implementing NGS methods
- Experience in conducting trainings

Tasks of the Short-term expert 1 (STE 1):

- Close cooperation with the Croatian experts in undertaking all activities
- Providing professional advice to Croatian experts during the project implementation period
- Undertaking all activities and achieving the mandatory results related to DNA sequencing such as:
 - o Conducting validation study and preparing validation report
 - o Organizing and conducting workshop, on-the-job training and round table discussion
 - Preparing Standard Operation Procedures
 - o Supervising pilot implementation according to the drafted Standard Operating Procedures
 - o Conducting TNA and preparing TNA report
 - o Preparing long-term training programme and training manual

Profile of the Short-term expert 2 (STE 2)

Requirements:

• University level education or equivalent professional experience of 8 years in the field of molecular biology

- Minimum 3 years of experience in bioinformatics and/or biostatistics
- Working level of English language
- Proven contractual relation to public administration or mandated body, as defined under Twinning Manual 5.4.5
- Computer literacy

Assets:

- Experience in forensic biostatistics
- Experience in data analysis and/or interpretation of NGS results
- Experience in conducting trainings

Tasks of the Short-term expert 2 (STE 2):

- Close cooperation with the Croatian experts in undertaking all activities
- Providing professional advice to Croatian experts during the project implementation period
- Undertaking all activities and achieving the mandatory results related to bioinformatics and/or biostatistics such as:
 - o Conducting validation study and preparing validation report
 - o Organizing and conducting workshop, on-the-job training and round table discussion
 - o Preparing Standard Operation Procedures
 - o Supervising pilot implementation according to the drafted Standard Operating Procedures
 - o Conducting TNA and preparing TNA report
 - o Preparing long-term training programme and training manual

Note:

The pool of experts should include:

- At least one short-term expert who in addition to the respective profile requirements has experience in forensic STR and/or SNP typing using NGS technology.
- At least one short-term expert who in addition to the respective profile requirements has experience in NGS of mtDNA.
- At least one short-term expert who in addition to the respective profile requirements has experience in data analysis and interpretation of NGS results.

4. Institutional Framework

The beneficiary institution of the project is Ministry of Interior of the Republic of Croatia. Forensic Science Centre "Ivan Vučetić" is a part of the General Police Directorate of the MoI. It provides forensic expertise in the following areas: documents, DNA, drugs, fibers, finger prints, firearms, fire and explosion, handwriting, marks, paint, road accident analysis, cybercrime and when needed crime scene investigations and forensic services to the Ministry of Defense and the Customs. Forensic experts' reports are sent to the ordering authority and become part of important documentation in the course of criminal investigation and judicial proceeding. Forensic science experts are often called upon the main hearing in court in order to corroborate the evidences according to their expertise.

The Department of Biology and Fibers at FSC performs biological expertise, mainly DNA analysis of any kind of biological traces and referent samples for comparison. Biology section employs 24 persons, out of whom 18 employees are forensic science experts of 3 different ranks, 5 employees are laboratory technicians and one person is an administrative officer.

Implementation of NGS methods into forensic DNA laboratory would generate the maximum amount of information even from challenging evidence samples, or known reference samples, by improving

the work of the criminal police and effectiveness of combating crime, also answering the questions being asked by law enforcement, while retaining compatibility with existing databanks.

The results of the project will not lead to a change of the institutional framework.

The beneficiary institution will dedicate all necessary human and financial resources in order to guarantee an effective implementation of the respective project. In particular, the beneficiary institution will insure the availability of the following provisions:

- Adequately equipped office space for the RTA and the RTA assistant for the entire duration of their secondment (in particular a desk, a telephone line, PC with e-mail account and internet access, possibility to use fax & copy services).
- Adequate conditions for the STEs to perform their work while on mission to the BC.
- Training and conference venues as well as presentation and interpretation equipment.
- Costs for travel by BC participants from their capitals to a MS or between MS (study visits).
- Its active involvement in preparation of the PIU and Steering Committee meetings and participation of its members on the same.
- The availability of the BC human resources (BC experts) during the implementation of the activities.

5. Budget

Implementing Next Generation Sequencing (NGS) technology in DNA forensic science laboratory (CRO NGS)	Transition Facility Contribution	National Co-financing	TOTAL
Twinning Contract	(90 %) 576.173 EUR	(10 %) 64.019 EUR	640.192,00 EUR

The total amounts of the Transition Facility Contribution and National Co-financing stipulated in the above table represent the total maximum amounts and therefore, they may be reduced at the level of the Twinning contract, while the relevant ratio (percentages) should be maintained and fixed.

The co-financing requirement foreseen under Transition Facility will be considered fulfilled according to the provision of the relevant Financing Decision.

Interpretation costs will be reimbursed from the budget only for the purpose of workshops and seminars, up to 7% of the Contract amount can be used for translation and interpretation purposes.

6. Implementation Arrangements

6.1 Implementing Agency responsible for tendering, contracting and accounting:

Central Finance and Contracting Agency (CFCA) Ulica grada Vukovara 284, Objekt C 10000 Zagreb, Croatia Ms Nataša Mikuš Žigman, Director

Phone: +385 1 6042 400 Fax: +385 1 6042 598

E-mail: procurement@safu.hr

Twinning Administrative Office

Central Finance and Contracting Agency Ulica grada Vukovara 284, Objekt C 10000 Zagreb, Croatia

Ms Nirvana Sokolovski, Twinning NCP

Phone: +385 1 6042 400 Fax: +385 1 6042 598 E-mail: twinning@safu.hr

6.2 Main counterpart in the BC:

<u>Deputy Senior Programme Officer (SPO):</u>

Mr Krešimir Perović, Acting Head of Independent Sector for Schengen Coordination and

European Union Projects

Ministry of Interior

Ulica grada Vukovara 33

10000 Zagreb, Croatia

Phone: +385 1 61 22 561 Fax: +385 1 61 22 461

E-mail: kperovic@mup.hr

Project Leader counterpart:

Mr Dražen Mayka, Assistant Director of Forensic Science Centre Ministry of Interior Ilica 335 10000 Zagreb, Croatia

RTA counterpart:

Ms Marina Korolija, DNA expert Ministry of Interior Ilica 335 10000 Zagreb, Croatia

6.3 Contracts:

It is envisaged that the Project will be implemented through one Twinning Contract, with an maximum amount of 640.192,00 EUR.

7. Implementation Schedule (indicative)

7.1 Launching of the call for proposals: 1Q 2016³

7.2 Start of project activities: 4Q 2016

7.3 Project completion: 4Q 2017

7.4 Duration of the execution period (number of months): 15 months; the execution period will end 3 months after the implementation period of the Action (work plan) which will take 12 months.

³ Member States submitting proposals for Twinning projects implemented in Croatia, as well as the beneficiary institutions, will be requested to finalise drafting of the contracts in maximum four months regardless of the period of the year during which the drafting will take place.

8. Sustainability

This Twinning project is designed to provide all necessary preconditions to implement NGS methods in forensic science casework.

NGS instrument validation study according to the ISO 17025 standard will be conducted, protocols developed and corresponding report prepared. Additionally, DNA experts will be trained in order to enable them to conduct independent validations of NGS instrument, bioinformatic and biostatistic data analysis and interpretation of NGS results thus ensuring lasting value of the relevant project result.

Moreover, through this Twinning project Standard Operating Procedures for NGS methods will be drafted, tested, discussed and finalized in line with suggestions resulting from pilot use and round table discussions. These written documents, together with increased capacities of forensic DNA experts (achieved through on-the-job trainings and internships), will be a permanent asset to forensic science work in Croatia even after the end of the Twinning project implementation.

The Ministry of Interior shall provide support for effective project implementation, which will guarantee the adequate sustainability of project results. Therefore, it is expected that the project will have positive long term impact on implementing NGS methods in routine forensic casework.

9. Crosscutting issues

Based on the fundamental principles of promoting equality and combating discrimination, as provided in Croatia's legislation and practice, participation in the project will be guaranteed on the basis of equal access regardless of sex, racial or ethnic origin, religion or belief, disability, age or sexual orientation.

10. Conditionality and sequencing

n/a

ANNEXES TO PROJECT FICHE

- 1. Logical framework matrix in standard format
- 2. Detailed implementation chart
- 3. Contracting and disbursement schedule by quarter for full duration of programme (including disbursement period)

Annex 1. Logical framework matrix in standard format

Implementing Next Generation Sequenci laboratory (CRO NGS)	ng (NGS) technology in DNA forensic science	Programme name and number: Transition Facility IPA/2013/24986 (Annex of C(2013) 8057 final); Institution Building Envelope	
Forensic Science Center "Ivan Vučetić Interior, Croatia	" at General Police Directorate, Ministry of	Contracting period expires: 3 years from the day on which the Commission notifies the Republic of Croatia that all of its internal procedures necessary for the adoption of this Decision have been fulfilled	Disbursement period expires: 4 years following the expiration of the contracting deadline
		Total budget: 640.192,00 EUR	Transition Facility financing: 576.173 EUR (90%) National co-financing: 64.019 EUR (10%) ⁴
Overall objective	Objectively Verifiable Indicators	Sources of Verification	
Capacities of the Ministry of Interior enhanced to effectively combat crime within the EU and international environment, in line with the related EU policies and strategies.	 Enhanced effectiveness in combating crime Decreased number of unsolved cases 	 Annual Peer Base Missions report Relevant EC reports Relevant national reports MoI annual and statistical reports FSC «Ivan Vučetić» annual and statistics reports 	
Project purpose	Objectively Verifiable Indicators	Sources of Verification	Assumptions
	Validation study for NGS instrument conducted and protocols according to ISO 17025 standard procedures developed		 Full commitment of the MoI Organizational, technical and infrastructure capacities necessary for implementation of the project in place Human resources for the implementation of the project in place

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⁴ The total amounts of the Transition Facility Programme Contribution and National Co-financing stipulated in the above table represent the total maximum amounts and therefore, they may be reduced at the level of the Twinning contract, while the relevant ratio (percentages) should be maintained as fixed. The co-financing requirement foreseen under Transition Facility will be considered fulfilled according to the provision of the relevant Financing Decision.

		 List of participants on workshops List of participants on round tables List of participants on internships Internships reports 	
Results	Objectively Verifiable Indicators	Sources of Verification	Assumptions
Result 1: Validation study for NGS instrument conducted and protocols according to ISO 17025 standard procedures developed	 Objectively Verifiable Indicators Validation study of the forensic NGS platform for forensic STR and SNP typing conducted, protocol developed and validation report prepared Workshop with the purpose to present validation report for performed validations on forensic NGS platform for forensic STR and SNP typing with at least 5 participants conducted Validation study of the research NGS platform for mtDNA sequencing conducted, protocol developed and validation report prepared Workshop with the purpose to present validation report for performed validations on research NGS platform for mtDNA sequencing with at least 5 participants conducted On-the-job training for at least 3 DNA experts related to conducting independent validation for NGS instrument and developing protocols organized and conducted On-the-job training for at least 1 DNA expert related to biostatistic and bioinformatic data analysis and interpretation of NGS results organized and conducted Standard Operation Procedures for: forensic STR and SNP typing data analysis and interpretation of forensic STR and SNP typing results data analysis and interpretation of mtDNA sequencing results 	Sources of Verification Twinning project reports Documentation produced under the project (e.g. reports with recommendations)	Full commitment of the MoI Organizational, technical and infrastructure capacities necessary for implementation of the project in place Human resources for the implementation of the project in place

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Result 3: Capacity of forensic DNA experts in the area of NGS methods strengthened	 Pilot implementation of DNA analysis on NGS instrument according to the drafted Standard Operating Procedures in FSC "Ivan Vučetić" conducted and report with recommendations prepared Pilot implementation of data analysis and interpretation of NGS results according to the drafted Standard Operating Procedures in FSC "Ivan Vučetić" conducted and report with recommendations prepared Final Standard Operation Procedures based on the recommendations resulting from pilot implementation of drafted Standard Operation Procedures and round table discussion with at least 5 participants prepared Workshop for at least 10 participants with the purpose to present Standard Operation Procedures organized and conducted Training need analysis (TNA) related to NGS methods conducted and TNA report prepared Long-term training programme and training manual prepared On-the-job training in the BC for at least 6 forensic DNA experts conducted Internship for 4 participants in the MS (in duration of 8 weeks for 2 forensic DNA experts and 4 weeks for 2 forensic DNA experts) organized and conducted and internship report prepared 		
Activities	Means	Specification of costs	Assumptions
Member State(s) is kindly requested to	Analysis, Preparation of documentation,	Twinning project:	In line with the assumptions specified for
develop activities in the submitted		640.192,00 EUR	results.
proposal which are needed in order to			
achieve the results stipulated in the	job training, Internship.		
fiche.			
			Preconditions:
			n/a

Annex 2. Detailed implementation chart

Implementing Next Generation	2016					2017											20	18						
Sequencing (NGS) technology in DNA																								
forensic science laboratory (CRO NGS)																								
Month	M	Α	M	J	J	Α	S	О	N	D	J	F	M	Α	M	J	J	Α	S	O	N	D	J	F
Twinning	Т	Т	Т	Т	С	С	С	С	С	A/ I	I	I	I	Ι	I	I	I	Ι	Ι	I	I	R	R	R

T – Call for proposals and evaluation
C – Contracting
A/I – Arrival of the RTA/ Start of the implementation of activities
I – Implementation of activities

R – Report

Annex 3. Contracting and disbursement schedule by quarter for full duration of programme (including disbursement period)

Implementing Next Generation Sequencing (NGS) technology in DNA forensic science laboratory (CRO NGS)	EUR (provisional)										
	2017										
	I	II	III	IV							
Twinning	640.192,00										
TOTAL (EUR):	640.192,00										

Implementing Next	2017				2018			
Generation Sequencing (NGS) technology in DNA forensic science								
laboratory (CRO NGS)	I	II	III	IV	I	II	III	IV
Twinning	499.662,05				140.529,95			
TOTAL (EUR):	499.662,05				640.192,00			