

26th ENGL STEERING COMMITTEE MEETING

25-26 March 2014, Ispra, Italy

MEETING REPORT

1. Introductory activities

1.1 Welcome, quorum, apologies, new members

The Chairman welcomed the participants, announced the presence of a new member from Croatia. Apologies were received from UK, SE, RO, NO, IE and GR.

1.2 Approval of the agenda

The agenda (see Annex 1) was approved without modifications

1.3 Approval of the report SC25 meeting

The report of the last meeting was adopted without changes.

1.4 Review of Dynamic Action List (DAL SC25)

The Secretary reviewed the DAL of the 25th Steering Committee meeting. No comments were raised.

2. Progress reports ENGL working groups

2.1 WG MPR (Method Performance Requirements): final draft of the MPR document for approval, prior to sharing with industry and general ENGL

The working group co-chair reminded that the last version had been already presented at the ENGL plenary in December 2103. The WG has since then decided to modify slightly the acceptance criteria for PCR efficiency by being less stringent for the single slopes but applying the criterion to all individual runs. The SC agreed that the document could now be submitted to Europabio and to all ENGL members for comments. It should be finalised for approval by the ENGL plenary in June 2014. The SC congratulated the WG for its good work.

$2.2~\mathrm{WG}$ SPP (Sample Preparation Procedure): presentation of final draft for approval before adoption at plenary ENGL

The document has been already circulated for comments and only editorial changes were further suggested. The SC agreed and appreciated that document will now be finalised by the chair of the WG and thereafter published.

2.3 AG SMV (Advisory Group on Selection of Methods for Validation): approval of methods for validation

The WG leader explained that the WG finally had its first face-to-face meeting in February 2014, where it established selection criteria that should be taken into account when selecting a method for validation. Higher priority should be given to methods closing an analytical gap and/or offering an added value in comparison to previously adopted and already available methods. In any case, there should be evidence that the selected method complies with the ENGL methods acceptance criteria. The AG-SMV also decided to revise the initial list of method established (see Report of AG SMV of Feb 2014 at https://englnet.jrc.ec.europa.eu/WG-AG-SMV/Shared%20Documents/ReportAGMSV_02_2014_final.pdf).

The following analytical gaps were newly identified by the advisory group:

- CMV-CP (Cucumber Mosaic Virus coat protein) element detection
- Detection of Cauliflower Mosaic Virus (CaMV) natural contamination
- Detection of Agrobacterium tumefaciens natural contamination
- GM papaya construct-specific method
- Detection of the T-35S element from pCAMBIA vector
- high-throughput strategies
- several taxon-specific methods (see Report of AG SMV of February 2014)

Some new methods targeting the identified analytical gaps were reviewed by the AG-SMV. The members agreed that the method targeting the T35S element from pCAMBIA (Fraiture *et al.* 2013) seems to meet the established criteria and decided to consider it for validation.

The Chair of the WG, supported of the chair of the ENGL, invited all ENGL members to submit methods they believe covering any of the analytical gaps listed above or show, in their view, an added value over already available methods. To support this, the EU-RL will place a web-form on the ENGL website for the submission of proposals. This form will be in line with the selection criteria established by the AG-SMV. It will request, *inter alia*, to explain how the method meets the specified criteria, and which analytical gap it covers and/or which added value it has. Any submission must include the sequences of the primers, probes, and target elements.

The chair of the AG-SMV informed the SC that the results on the CaMV strains coverage by the three detection methods tested by the ISP (BE) (Chaouachi *et al.* 2008 and manuscripts in preparation for the CAP and CRT methods) will be published soon.

The SC acknowledged the progress made by the AG-SMV and agreed to the proposed priority and supported the invitation to all ENGL members to submit methods for consideration by the AG. The idea to put a user friendly web-form template on the ENGL website was appreciated.

The Chairman of the ENGL announced that the cry1Ab/Ac, P-FMV and P-nos methods already validated by the German Federal Office will be included into the GMOMETHODS database.

2.4 WG DIR (Detection Interpretation Reporting); progress report

The WG leader announced that two members of the subgroups will complete each of the parts already drafted and the WG intends to provide a consolidated version in end of May in order to allow sending it out to the entire ENGL for comments and to finalise it for adoption at the ENGL plenary on the 4th-5th of June. The Chairman of the ENGL stated that in case that this ambitious timing cannot be fully met, there should be at least an in-depth discussion at the ENGL plenary, allowing the WG to finalise the report and to approve it by written procedure after the ENGL plenary.

2.5 WG-IGSE (Identification of stacked GM events (WG-IGSE): presentation of final draft for approval before adoption by ENGL, and discussion on follow-up

The Chairman of the ENGL remarked that the WG has been able to deliver the final document in a very short time. The WG leader reviewed the WG mandate and history (set up in September 2013, first meeting on the 12th-13th of November 2013, 2nd meeting on the 13th-16th of January 2014) and reminded the draft report that already has been submitted to the ENGL and ENGL-SC members.

The conclusion is that the methodologies available allow distinguishing stacked events from mixtures of non-stacked events only by analysing single cells or seeds. He questioned what the next steps would be, e.g. agreement by the ENGL-SC and approval of the final report at the next ENGL plenary.

The latter point was discussed and it was remarked that the ENGL may be reluctant to adopt the document as a technical guidance, which indeed, it is not. The Chairman suggested considering it as a review paper and asked the WG to monitor the advancements in technology, for the time being without the duty of producing further work, hoping that this would allow, over time to identify new possible solutions.

He also stated that the report will be communicated to DG SANCO, who had asked the ENGL to address the issue, and any further action should take account of DG SANCO's comments.

3. New activities/issues

3.1 WG on seeds (WG-ST): definition of WG mandate

The Chairman presented a draft mandate for the future working group on seed testing, as prepared by the secretariat. According to this mandate, the WG should prepare a technical document on testing seed samples for the adventitious presence of GM seeds, taking due account of and describing the current practices of GMO seed testing in the Member States. As the practical LOD in seed lots can be lowered by means of sub-sampling and increased efforts of testing, the WG is asked to estimate the correlation between effort (cost) and theoretical LOD for the most relevant crops, i.e. those for which contamination with GMO may occur. The purpose is to determine what is realistically possible and to identify the parameters that are significantly influencing the practical LOD, and to propose a reasonable compromise. The mandate was adopted as presented in annex (see Annex 2).

The Chairman invited the SC members to propose suitable experts for the WG and suggested to involve experts from laboratories with practical experience of testing seed for GMO presence. He agreed that also non-ENGL members could be invited. The secretariat was asked to send a call for expression of interest to become a member of this working group to the ENGL members. It was also agreed that at least two meetings should planned for drafting the document; the draft report should be presented at the Steering Committee meeting in September 2014.

3.2 WG on DNA extraction methods

The Chairman explained that the EU-RL GMFF is aware that for the day-to-day work of the GMO control laboratories, DNA extraction is as demanding if not more than the PCR experiment as such. In order to promote exchange of good practice and practical experience between the ENGL members, the secretariat therefore proposed setting-up a new WG on DNA extraction from difficult matrices. In the discussion of this proposal, the SC remarked the extensive literature already existing on the subject but expressed interest in improving the ENGL internal sharing of information and advice on methods used for particular matrixes.

It was proposed to re-launch, for this purpose, the ENGL website (ENGL intranet) as a discussion and exchange platform. Members suggested stimulating such an exchange of know-how also on other topics and asked to provide on the ENGL-net a table into which the ENGL members should

introduce a description of their projects and expertise. The Chairman commented that an efficient strategy or platform for sharing particular experience could then be based on identifying common problems from that table.

3.3 Potential link between ENGL activities and allergen detection

The Chairman suggested discussing this point at the next ENGL plenary. However, he explained that in his view, the particular expertise of the ENGL could also be useful for many other areas than GMO analysis, such as allergens. He proposed to enter into a discussion about other areas in which the experience of the ENGL could be exploited. As many ENGL member laboratories apply their expertise in DNA extraction and analysis also to other areas, it was agreed to design a (simple, short) questionnaire for mapping non-GM activities of the ENGL-laboratories. This proposal was agreed and the issue will also be taken-up at the next ENGL plenary meeting.

As a follow-up to this discussion, participants suggested having presentations at the meetings on new DNA detection technology, on metabonomics profiling, and on genomics. While this suggestion was agreed to in principle, the decision on topics and speakers was postponed to the prior identification of the non-GMO expertise and interests of the ENGL members.

3.4 Sharing info on research projects carried out by ENGL members

It was decided that this information can be shared on ENGLnet (e.g. using a table where users can type the name of their institution and mention the projects, see above).

4. General ENGL issues

4.1 Participation to ENGL meetings and general transparency

The Chair expressed his view that the ENGL could be made more visible and transparent by publishing the WG mandates, the final reports of the WGs, and the reports and documents approved by, and used for the ENGL and ENGL Steering Committee meetings.

Concerning participation of non-ENGL members (e.g. biotech industry, NGOs and other stakeholders), no specific rules exist and should therefore be developed. For the time being the requests of participation to the ENGL meetings, or to some sessions of the meetings, are dealt with on a case-by-case basis.

The SC agreed to his proposals and invited the secretariat to prepare a proposal, e.g. a set of rules of proposal, implementing the existing ENGL consortium agreement.

4.2 Preparation of the 21th ENGL plenary meeting (4-5 June 2014)

The Chairman suggested doing parts of the meeting by breaking up the plenary into smaller breakout groups in order to allow for a more lively discussion between the participants. These groups could receive different topics for discussion or all the same. The idea was discussed and agreed to and the SC suggested "DNA extraction from difficult matrices" as one topic for the break-out groups. The chair invited the SC members to reflect and to propose other topics.

5. Scientific / technical issues (part 1)

5.1 Update on EURL-GMFF PSPP (pre spotted plates project)

The EU-RL GMFF summarised the work done so far on the project and specifically on the organisation of a pilot study. The objective of the pilot study is to investigate the economical and practical advantages of the use of PSPs in routine testing laboratories.

Twenty laboratories have been selected for the pilot study; they will receive eight screening-specific and eight event-specific plates with reagents. The participants will be asked to analyse seven of their own known GM-positive and two of their own known GM-negative samples with the plates and to compare their "normal" laboratory internal procedures against the PSP-based workflow.

They should address not only the relative reliability and quality of the testing but in particular they should compare the time, cost and practicality of the two approaches. Participants will be asked to report the results obtained with the PSPs and with their standard procedure, to estimate the relative cost per sample for the analysis and to assess the overall time spent for processing a sample.

The EU-RL GMFF will also ask for a general evaluation of the plates, e.g. participants will be asked to state if they represent an improvement in the laboratory capacity, ease of use and reliability and if they could be chosen as a routine testing tool – or not.

The laboratories should also indicate the target cost they expect for this type of support (PSP) that would the related workflow economically viable. The results are expected within two months and the final report in July-August 2014.

5.2 Revised guidance document on Chinese rice, for approval

The revised guideline was prepared by the EU-RL GMFF and includes specific criteria for the interpretation of results in case of dubious signals produced by the Cry1Ab method. The guideline has been also streamlined to the needs of the users and adapted to the modifications of Decision 2013/287/EU. The guideline will be published on the EU-RL GMFF website in May 2014.

5.3 Validation of the CoSYPS (N. Roosens, BE)

The Belgian member informed that an article on the validation of ten SYBR Green methods supporting the CoSYPS platform was published and is available as open access paper.

6. Scientific / technical issues (part 2)

6.1 Update on enlargement project (JRC)

The Enlargement project has been approved for another year. The activities planned in 2014 were presented. A regional and an international workshop will be organized, as well as two training workshops. The first training workshop, arranged for the 20th-22th of May 2014 will focus on the implementation of quality systems/ISO17025 accreditation with a total of 20-25 participants from all regions. Candidatures for additional expert trainers are welcome.

The second training workshop planned on 24th-26th September 2014 in Ispra will focus on practical aspects of regulatory GMO testing and implementation of analytical workflows. Volunteers' trainers were requested.

A second regional workshop for MENA countries will be organised in Rome on 25th-26th June for Middle East and North African countries. Invitations for speakers were launched.

In the context of JRC enlargement & integration activities, an international workshop on harmonization of GMO detection and analysis will be arranged on the 2nd or 3rd week of October 2014; it will be organised in Skopie, Macedonia with approx. 40 participants from the region.

The second international workshop on GMO Analysis and Networking will be organised on the 3rd-4th December 2014 in Ispra, back-to-back to the second ENGL-plenary in 2014. Delegates will be invited from all regions including representatives from regional networks and individual countries. It will be organised in conjunction with the ENGL plenary to facilitate participation of ENGL members.

6.2 Update on GM wheat (Sec)

The Secretary informed that a second and more sensitive event-specific method is ready to be published and informed that the EU-RL GMFF is preparing plasmids to be used as a control sample for MON71800. The Chairman asked if laboratories are implementing the detection strategy developed by the EU-RL GMFF and some members confirmed its use.

6.3 Update on papaya

Recent findings on the analysis of genome sequences of papaya received from the German Federal Office and the Belgian Walloon Agricultural Research Centre were presented by the JRC. A bioinformatics assembly of the sequences was made since the sequences publicly available from the genetically modified rainbow variety could not be used as a reference genome.

The presence of many genetic GM elements in the new papaya genome is suggested, including coat protein coding sequences, some apparently non-functional; 35S and T-nos elements, and the nptII and beta-lactamase genes. The organisation of three fully characterised insertion fragments confirms the results of the detection made by the German Federal Office.

The Chairman asked if laboratories were still testing for GM papayas Participants confirmed that controls were still implemented but without positive findings. It was recalled, however, that some papaya supplements (pills) have been found positives for 35S, T-nos and nptII elements.

7. Scientific / technical issues (part 3)

7.1 Detection of GM material in food when the plant species of such GM material is not declared as an ingredient

According to the feed law, botanical impurities are accepted below the 5% level. It was explained that for complex matrixes, screening assays point to the presence of undeclared ingredients and that it is unpractical to quantify GM contamination of the missing ingredient on the total overall amount (mass/mass).

It was asked how this issue is handled in the different countries. The Chairman reminded that the ENGL's mandate is limited to the scientific technical level and does not include discussion of the interpretation of the regulations – that is the responsibility of the competent authorities, not of the testing laboratories.

SANCO representative specified that for food samples the ingredient must be declared while a botanical impurity needs to be reported only if it is an allergen; the laboratories could only provide information on the relative presence of GM in respect to the corresponding species and the CA have the responsibility to take decision on such issues.

7.2 Status of official methods when scientific evidence suggests risk of underperformance

A member of the ENGL SC asked if, and under which conditions, it would be appropriate to use reference gene assays different from those used in the validation of GM event specific assays if there is, for example, new scientific evidence of an improved performance.

The EU-RL GMFF responded that, in principle, the methods should be implemented exactly as they have been validated.

However, under the perspective of accreditation, other peer-reviewed (taxon specific) assays could also be implemented if correctly verified in the laboratory and recognised by the accreditation body. In this context it would be important that the concept of modularity is applied and that using the assay is supported by (peer reviewed) scientific studies.

The EU-RL GMFF, for example, has carried out bridging studies on a number of stack events that confirmed that different reference genes could be interchangeably used without rendering a validated detection method invalid. This is, for example the case of using the maize *hmg* reference gene in place of the maize *adh*1 reference gene originally used in the official validation of the event-specific method for MON863 and NK603

The Italian NRL confirmed this possibility and referenced a comparison study where in-house validated taxon specific methods in combination with officially validated event specific methods were considered to be in line with the accreditation requirements.

8. AOB

A brief presentation on the progress of the GMOval project was given. Within the project, the EU-RL GMFF is responsible for organising the ring trials for the validation of four qualitative methods. The validation studies will be conducted taking into account the implementation of the POD (probability of detection) approach.

The Chairman proposed to reflect on the consortium agreement and on the rules for publishing and approving documents. The intention is to produce guidelines for internal procedures without modifying the consortium agreement. Feedback and comments were requested. On the basis of the feedback the Secretariat will propose a document covering also consultations using the internet discussion platform.

Clarifications on the follow-up on the report of the WG IGSE were requested; the Chairman informed that the document probably will be published on the web, after consultation with SANCO. In any case, the ENGL members are invited to propose research activities to support the advancement in this area.

The Secretariat presented the draft agenda for the 21st ENGL plenary meeting; additional proposals were discussed and included in the agenda.

9. DAL SC26 and next meetings

The Secretary reviewed the DAL of the 26th Steering Committee meeting (see Annex 3).

The 21st ENGL plenary meeting will take place on the 4th -5th June 2014 in Barza, Ispra.

The 27th ENGL Steering Committee meeting will take place on 15th -16th September in Ispra.

The Chairman expressed the intention for having the SC more involved in aspects of the organisation, governance and functioning of the network.

He thanked the participants and closed the meeting.

List of Annexes:

- 1) Agenda
- 2) Mandate of the WG-ST (see 3.1)
- 3) DAL (Dynamic Action List) SC 26

Annex 1: agenda





26th ENGL STEERING COMMITTEE MEETING

25-26 March 2014, Ispra, Italy

DRAFT Agenda (Version 21/03/2014)

	12:30	Buffet lunch					
AP	Time	Topic	Documents in ENGLnet				
$\overline{}$	Day 1	•					
1.1	14:00	 Welcome, quorum, apologies, new members 					
1.2		 Approval of the agenda 	Agenda				
1.3		 Approval of the report of the SC25 meeting 	Report SC25				
1.4		 Review of Dynamic Action List (DAL SC25) 	DAL SC25				
	15:00	Coffee Break					
2	15:30	Progress reports ENGL working groups					
2.1		WG MPR (Method Performance Requirements): final draft of Final WG draft					
		the MPR document for approval, prior to sharing with					
		industry and general ENGL					
2.2		 WG SPP (Sample Preparation Procedure): presentation of 	Final draft				
		final draft for approval before adoption at plenary ENGL					
2.3		 AG SMV (Advisory Group on Selection of Methods for 	Report				
		Validation): approval of methods for validation	_				
2.4		 WG DIR (Detection Interpretation Reporting): progress report 	DIR update				
2.5		 WG-IGSE (Identification of Identification of stacked GM 					
		events (WG-IGSE): presentation of final draft for approval	Final draft				
		before adoption by ENGL, and discussion on follow-up					
	17:30	End of day 1					
	19:30	Social dinner Hotel Europa, Ispra					
3	Day 2	New activities					
3.1	09:30	 WG on seeds: definition of WG mandate (Sec) 	Background & draft TOR				
3.2		 WG on DNA extraction methods (Sec) 					
3.3		Potential link between ENGL activities and allergen detection					
		(M. De Loose, BE)					
3.4		Sharing info on research projects carried out by ENGL					
		members (P. Phillip, FR)					
4		General ENGL issues					
4.1		 Participation to ENGL meetings and general transparency 					
Ι.		(Sec)					
4.2		 Preparation of the 21th ENGL plenary meeting (4-5 June 	Draft agenda 21 th ENGL				
_		2014)	Plenary				
	10:45	Coffe Break					
5	11:15	Scientific / technical issues					
5.1		Update on EURL-GMFF PSPP (pre spotted plates project)	Presentation				
		(Sec)					
5.2		Revised guidance document on Chinese rice, for approval	Final draft				
5.3	10.00	Validation of the CoSYPS (N. Roosens, BE)					
	12:30	Buffet lunch					
6	14:00	Scientific / technical issues					
6.1		Update on Enlargement Project activities (M. Querci, JRC)					
6.2		Update on GM wheat (Sec)					
6.3		Update on papaya (Sec and SC members)					
	15:15	Coffee Break					

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7	15:45	Scientific / technical issues	
7.1		 Detection of GM material in food when the plant species of such GM material is not declared as an ingredient 	
7.2		 Status of official methods when scientific evidence suggests risk of underperformance 	
8	16:30	AOB (date SC27; date ENGL 22nd plenary)	
9	16:45	DAL SC26 and End of Meeting (17:00)	DAL SC26

Annex 2: Mandate of ENGL WG-ST



EUROPEAN COMMISSION JOINT RESEARCH CENTRE

Institute for Health and Consumer Protection Molecular Biology and Genomics Unit



Mandate for an ENGL Working Group Seed Testing

The WGST shall prepare an ENGL/EURL advice to the Commission concerning the testing of seed samples for the adventitious presence of GM seeds.

In preparing this advice the WGST shall take due account of, and describe the current practice of GMO seed testing in the Member States of the European Union.

As the practical detection limit for GM seed in non-GM seed lots can be lowered by means of sub-sampling and increased efforts of testing, the WGST is asked to estimate the relation between effort (cost) and theoretical detection limit for the most relevant crops.

Background

In a number of Member States of the EU there is an active breeding industry that put seeds on the market in the EU and beyond. As some of these might also apply genetic engineering technologies for producing GMOs for the EU or non-EU markets, adventitious presence of GM seeds in conventional seed lots is difficult to be excluded.

At the same time seed is imported into the EU from countries where GMOs of the same species are planted and adventitious presence of GM seeds in imported conventional seed lots is difficult to be fully excluded.

Testing seed lots for the adventitious presence of GM seeds is practiced in EU member states. No threshold is set for labeling of seed lots if GM seeds are found to be present. In most EU countries sampling seems to be based on the ISTA seed testing rules for purity (to be verified by the WGST) but hitherto no EU wide practical guidance exists neither for seed sampling nor for the analytical methods to be applied.

Timeline

The working group is expected to produce a final draft for approval by the ENGL Steering Committee to be held on 15-16 September 2014. At least two meetings are foreseen.

Annex 3: DAL (Dynamic Action List) SC 26

AND ENGLISHEDING COMMITTEE ACTION LIGHT				
26th ENGL STEERING COMMITTEE ACTION LIST				
ACTIONS	Resp. ▼	Timelines ▼	Status	Comments
ENGL CONSORTIUM AGREEMENT				
Circulate report of 26th ENGL SC	SEC	Apr-14	Open	
Organise 21th ENGL Plenary in Ispra	SEC	Dec-14	Open	4-5 June
Organise 27th ENGL SC	SEC	Sep-14	Open	15-16 september
ENGL WORKING GROUPS				
WG Detection Interpretation Reporting (DIR)				
Final draft of the doc ready for the ENGL plenary 21st	SEC	May-14	Open	
WG Method Performance Requirements (MPR)		·		
Final draft to the ENGL and Europabio	SEC	Apr-14	Open	
Prepare final draft for adoption by ENGL 21st	SEC	May-14	Open	
WG Sample Preparation Procedure (SPP)		.,		
Publish the final doc	SEC	Apr-04	Open	verify with Gilbert
Advisory Group on "selection of methods for validation"				
Proposal for new template to submit methods	WG chair	Apr-14	Open	
Webform of the termplate	SEC	May-14	Open	where to put the form
Organise a meeting of the group	SEC	Oct-14	Open	to select methods for 2015
WG on Identification of stacked events (IGSE)				
Consult with SANCO	SEC	Apr-04	Open	
Submit the final draft to the ENGL	SEC	Apr-14	Open	FOR COMMENTS
OTHERS				
			Open	
Prepare a table on activities of the ENGL members	SEC	01/04/2014	Open	Blank table on ENGLnet
WG seeds: send mandate and EoI to ENGL	SEC	Mar-14	Open	
WG seeds: nomination of members and organise 1st meeting	SEC	25/04/2104	Open	
Questionnaire on non-GMO detection activities	SEC	May-04	Open	mapping activities
Publish the revised guidance on Chinese rice	SEC	Mar-14	Open	
Gather information on issues with CRMs	SEC	May-14	Open	go back to emails
Ask for control samples for wheat MON71800	EURL		Open	The EURL will distribute a PCS
·			Open	
Procedures to be annexed to the ENGL CA	SEC			proposal for the SC27
Publish the GM wheat event-specific method	SEC	Apr-14	Open	If NRLs need the draft report, ok.