CIPAC

COLLABORATIVE INTERNATIONAL PESTICIDES ANALYTICAL COUNCIL LIMITED

Commission Internationale des Méthodes d'Analyse des Pesticides (CIMAP)

Minutes of the 60th Annual meeting

The 60th meeting was held on Wednesday 15th June 2016 in Iino Hall Conference Center, Tokyo, Japan.

Those attending

- Items 1 to 7 on 15th June: members, correspondents, observers and expert witnesses.
- Items 8 to 12 on 15th June: members, correspondents and observers (representatives of industry and commercial laboratories, by special invitation only)

1. Welcome and introductory remarks by the chairman

The Chairman, Mr R Hänel, opened the 60th CIPAC meeting, and welcomed all the participants. As Mr T De Rijk the assistant secretary was not present on the meeting Mr L Benke agreed to act as temporary assistant secretary during the meeting.

2. Apologies

Apologies were received from:
Mr W. Dobrat, Mr J. Foltýn, Mrs L. Janeš, Mrs A. Kashouli-Kouppari, Mrs S. Marais, Mr A. Martijn, Mr M. Müller, Mr T. De Rijk, Mr F. Sánchez-Rasero, Mrs A. Santilio, Mrs J. Schlosserova and Mrs A.Venant.

3. Adoption of the agenda

The following amendments were made to the Agenda:
The name of Mr Keita Tsunemi and Mr Qibai Jiang were miss-printed in the previously circulated agenda.
Item 4.7 will be presented by Ms Bu Haiyan.
There were no other observations, the agenda was adopted.

4. Reports of expert witnesses on collaborative trials:

4.1 Chlorantraniliprole by Mrs. Mary-Ellen McNally (5034, 5035)

Ms McNally presented the results of a full scale collaborative study on the determination of chlorantraniliprole in technical material, FS, WG and SC formulations using HPLC-UV with a reversed phase C18 column, UV-detection at 275 nm, and internal standard calibration. The recommended HPLC column was an ACE C18 3µm-particle size, 4.6x150 mm column. As a result of ESPAC small scale study in 2015, the proposed column can be substituted with a 2.6 µm particle size, 4.6 x 100 mm Kinetex® C18 column or a 2.7 µm particle size, 4.6 x 100 mm Restek Raptor® C18 column. These three HPLC columns were validated for the method under good laboratory practices, for linearity, accuracy, precision and specificity. There were some comments from the participating laboratories on the high number of calibration points and high back pressure on the column during the chromatographic run.

Twenty-four laboratories received samples for this collaborative trial and twenty-one laboratories submitted the results.
The statistical evaluation was carried out according to the guidelines in the CIPAC document "Guideline for CIPAC collaborative studies Procedure for Assessment of Performance of Analytical Methods". The data were tested for outliers firstly using Cochran's test on the within laboratory variance and then using Grubbs test on laboratory means to test the between laboratory variance.
Using the proposed internal standard calibration method for chlorantraniliprole, the technical material and all of the formulations met the Horwitz criteria when the one Grubbs outlier was eliminated for the 35 WG formulation.
Based on these results, Ms McNally recommended that the chlorantraniliprole method to be accepted as provisional CIPAC method.
The following comments were received from the meeting:

- A high column back pressure was notified at the specified flow rate by one participant, who asked to mention in the trial protocol that stainless steel capillary connections should be used during the measurements instead of PEEK tubing.
- There were some comments on sending the collaborative trial results, the participants did not receive the report before the CIPAC meeting
- The Secretary explained that results were received late by CIPAC, it was not enough time to distribute them among the participants
- Sending the samples of the trial encountered also some problems

4.2 Clothianidin by Mr Michael Haustein (5051, 5052)

Mr Haustein presented the results of a study carried out for extension of CIPAC method 738 to WP formulations by two independent laboratories on five batches of a WP formulation.

Measurement of each individual batch was carried out at two different days in two different laboratories, double determination and double injection each. For the analysis a Zorbax Eclipse XDB-C18, 150 mm x 4.6 mm, 5 µm particle size column was used, a mobile phase of with acetonitrile/water/phosphoric acid, 150/850/1 (% v/v/v) as eluent, with a flow rate of 1 ml/min. The injection volume was 5 µl and the detector wavelength 269 nm.

Based on the relative standard deviation results (RSDr) obtained for the five individual WP batches, the CIPAC method was proposed suitable for the extension to WP formulation types. The repeatability results (ranging from 0.5 % to 0.7 % relative) were below the modified Horwitz criterion and were comparable to those of the original full scale trial for a WG formulation with equal a.i. content.

- There were no questions or comments to the presentation

4.3 Cyenopyrafen by Mr Keita Tsunemi (5038, 5039)

Mr Tsunemi presented the results of a small scale trial on the determination of cyenopyrafen in TC and SC formulations using HPLC-UV with a reversed phase ODS column, UV-detection at 280 nm and external standard calibration.
The study was carried out by four independent laboratories.
During the trial the participants conducted duplicate determinations on two different days for each sample, injected each sample solution in duplicate, calculated the mean value and checked the linearity before the determination. Two TC and three SC formulations were used.
Four data sets were obtained from four participant laboratories. The statistical evaluations were carried out according to ISO 5725. No stragglers and outliers were observed.
For all samples, the values of RSD_R (reproducibility relative standard deviation) were smaller than those calculated by Horwitz's equation. The proposed method was considered to be appropriate for the determination of cyenopyrafen in technical and suspension concentrate.

Mr. Tsunemi proposed to proceed with a large scale collaborative trial.

- There were no questions or comments to the presentation.

4.4 d-tetramethrin by Mr Quibai Jiang (5040, 5041)

Mr Jiang presented the results of a small scale trial on the determination of the active ingredient content and isomer ratio of the diastereomers of d-tetramethrin in technical material. d-tetramethrin is a mixture of the isomers (1*R-trans*, *R*), (1*R-trans*, *S*), (1*R-cis*, *R*) and (1*R-cis*, *S*) of tetramethrin in an approximate ratio of 4:4:1:1. In practice the *trans* isomer range is 75-85 % and the *cis* isomer range is 15-25 %.
d-tetramethrin was determined by gas chromatography with internal standardization and the isomer ratio of the diastereomers was determined by normal phase high performance liquid chromatography using a CHIRALPAK® AY-H, 250 × 4.6 mm (i.d.), 5 µm column, hexane/ethanol/diethylamine, 930/70/1 (v/v) mobile phase, detection wavelength 230 nm.
Three TC samples were sent to three independent laboratories. During the trial the participants conducted duplicate determinations on two different days with duplicate injections for each sample.
The statistical evaluation was carried out according to the CIPAC guidelines. The results were tested for outliers firstly using Cochran's test on the within laboratory variance and then using Grubbs test on laboratory means to test the between laboratory variance. The tests were carried out at the alpha level of 0.01 for outliers and 0.05 for stragglers.
After the initial evaluation the calculated RSD_R fulfilled the Horwitz criteria for two technical materials. One straggler occurred, using Cochrans' test (for TC-A material, in case of laboratory 2).
On the basis of the results, Mr Jiang proposed to proceed to a large scale collaborative study.
The following comments were received from the meeting:

- For reasons of safety would it be possible to use heptane in the mobile phase instead of hexane? Were there any tests carried out with heptane? The answer was that this option was not checked.
- In the presentation it was mentioned that the isomer ratio is roughly 4:4:1:1, but on the slide presented this ratio could not been observed. The answer was that the figure on slide 7 was for the technical material and not for the pure standard.
- Does the technical material have an ISO common name? The answer was that they are going to apply for an ISO common name.

4.5 Fenazaquin by Mr Rene Cochran (5036, 5037)

Mr Cochran presented the results of a full scale trial on the determination of fenazaquin in TC and SC formulations using HPLC-UV with a reversed phase column, UV-detection at 260 nm, and external standard calibration. The measurements were carried out in duplicate weighings and two injections on two different days. Two TC samples and two SC samples were investigated by the 14 participating laboratories; the results were evaluated according to ISO 5725 guidelines.
According to Cochran's and Dixon's tests, among the received results various stragglers and outliers were observed.
If no data were excluded (elimination of stragglers and outliers by Dixon's and Cochran's tests) and all the results provided by each one of the fourteen participants was used for the statistical evaluation, the Horwitz criterion was satisfied in all four cases: Fenazaquin TC1, Fenazaquin TC2, Fenazaquin 200 SC1 and Magister 200 SC2.
Mr. Cochran considered that the proposed methods can be considered to be suitable for the fenazaquin determination in technical and suspension concentrate and proposed the method to be accepted as a provisional CIPAC method.
The following comments were received from the meeting:

- There were some comments on sending the collaborative trial results, the participants did not receive the report before the CIPAC meeting (Luis, and others)
- As a general remark one participant draw the attention of the laboratories on the importance of the right analytical view regarding the interpretation and spread of the results. In case of TC-2 one laboratory obtained 1003.7g/kg on day1 and for the same sample 964.4 g/kg on day2. In such cases it is highly recommended to check the measurement for possible errors.

4.6 Flupyradifurone by Mr Michael Haustein (5049, 5050)

Mr Haustein presented the results of a small scale study carried out by DAPA with 6 laboratories on one TC and six different formulation samples (AL; EC; EW; FS; SL; WG) to demonstrate that the method is suitable for the determination of flupyradifurone in technical and in main formulation types. The homogenized sample containing flupyradifurone was dissolved in solvent mixture acetonitrile / purified water followed by active ingredient determination using gradient reversed phase high performance liquid chromatography, UV detection at 280 nm with an external standard calibration using a Phenomenex Kinetex C18, 50 mm x 4.6 mm, 2.6 µm particle size column.

The linearity of the method was checked in two concentration ranges 0.005-0.1mg/ml and 0.1-0.75 mg/ml. Both calibrations were linear. For all samples, the values of RSD_R were smaller than those calculated by Horwitz's equation. The proposed method was considered to be appropriate for the determination of flupyradifurone in technical and main formulation types.

The following comments were received from the meeting:

- Was it necessary to sonicate the samples for 15 minutes, this can warm up the samples? The answer was that in some cases shorter times may be sufficient.

4.7 Mancozeb by Ms Bu Haiyan (5047, 5048)

Ms Haiyan presented the results of a small scale collaborative study with 5 laboratories on an HPLC-UV method for analysing mancozeb in technical and WP formulations. The concentrations of mancozeb in the sample solutions were determined by external calibration method. The mobile phase and the diluted sample solution should be kept under alkaline conditions, pH 9.5-10.0, to avoid decomposition of the mancozeb. The linearity, selectivity, recovery and repeatability of the proposed method were evaluated. Comparison of the results using existing CIPAC method and the proposed HPLC method gave good results, differences less than 1%. The statistical evaluation of data was carried out according to ISO 5725 guidelines. Three results were identified as outlier (Grubbs test and Cochran variance homogeneity test). It was assumed that incomplete dissolution of the sample was responsible for these outlier and stragglers.
The RSD_R as determined from the collaborative study is not larger than RSD_R (calc.). Based on the results of this study, it was proposed to perform a CIPAC collaborative study to determine mancozeb in TC and WP by the HPLC method.

The following comments were received from the meeting:

- Was there any reason to use for column temperature 29 °C? The answer was that at 40 °C the mancozeb decomposes.
- It would be better to give the exact pH value instead of pH 9.5 - 10
- One participant considered that the pH range relates to the column type, which should be resistant at high pH values
- One participant raised the problem of purity of the used standard and the applicability of the method for other dithiocarbamates. The answer was that the standard was purified in-house and the method can be used also for other dithiocarbamates.
- One participant asked why sodium sulphite is used, and why Solution A should not be

used after 24 hours. The answer was that the solution decomposes in time. It was proposed to use the remark: freshly prepared solution should be used.
- What compound is detected during the HPLC measurement? There was no unequivocal answer.
- There were some comments on the selectivity of the method. It is not selective.
- What is the difference between the two lines on slide 18? One line was the existing CIPAC method the other is the proposed CIPAC method.

5. Reports of expert witnesses on other matters

5.1 Extension of 715/TC/M/ to pyriproxyfen/alpha cypermethrin Royal Guard LN (CIPAC/4886/m) by Mr Atmakuru Ramesh (5043, 5044)

Mr Ramesh presented an extension of the scope of two methods for the determination of pyriproxyfen and alpha-cypermethrin in Royal Guard LN. The content of pyriproxyfen and alpha-cypermethrin in long lasting net has been determined with reference to the method CIPAC/4887/R validated for pyriproxyfen and permethrin. The method of determination is a minor extension of the method CIPAC /4887/R.
High performance liquid chromatography with UV detector at 254 nm, a C18 HPLC column 250 mm x 4.6 mm, 5 µm and di-cyclohexyl phthalate internal standard was used for the analysis. The specificity, precision and accuracy of the method were checked. Based on the results obtained Mr Atmakuru proposed the CIPAC/4887/R extension method for LLIN, when active substances, pyriproxyfen and alpha-cypermethrin are incorporated in HDPE polymer, as there are only minor differences in the high performance liquid chromatography method using PDA detector.
The alpha-cypermetrin content was determined by method 454/LN, with the modification of using di-cyclohexyl phthalate internal standard instead of dioctyl phthalate, to use only one extraction process for both active substances.
The following comments were received from the meeting:
- It was questioned if citric acid has also been added to the calibration solutions and if the concentration range of the calibrations covered the concentration of the samples. The answer was that the calibration solutions were prepared according to the method except that acetic acid was added.
- 2 different method extension pyriproxyfen and alpha-cypermethrin

No other comments

5.2 Extension (applicability) of MT 46.3 to matrix release formulation by Mr Takashi Sasaki (5045, 5046)

Mr Sasaki presented a small scale collaborative trial on the extension of the CIPAC method MT 46.3 to MR formulation. This method is an extension to MR of the existing CIPAC method MT 46.3.1 Liquid formulations, MT 46.3.2 Solid formulations, MT 46.3.3 Solid formulations stored under pressure and MT 46.3.4 LN formulation for the accelerated storage procedure. The trial involved the testing of three different MRs (MR-Green, MR-Red, and MR-White).
The parameters tested show that:
- The mean pyriproxyfen content of pyriproxyfen MR after accelerated storage does not significantly differ from the content before storage and complies with the limit of the WHO specifications.

- The relative standard deviation (RSD) of the pyriproxyfen content in the 3 pieces of pyriproxyfen MR analysed individually after accelerated storage is not significantly different from the RSD before storage and unquestionably low.

- The pyriproxyfen retention rate after accelerated storage does not significantly differs from the retention rate before storage and complies with the limit of the WHO specifications.

These results confirm the applicability of the extension of the CIPAC method MT 46.3 (accelerated

storage procedure) to MR. Mr Sasaki proposed that this method extension would be adopted as a full CIPAC method.

The following comments were received from the meeting:

- Why Al foil is used in the trial, is it really needed?
- How are the results affected by the use of Al foil?
  The answer was that it was easier to handle the sample if it was wrapped in the Al foil, as the formulation is very solid and difficult to cut. The active ingredient would be adsorbed less on Al foil than on the plastic bag.
- In CIPAC MT 46.3 the sample is placed in a glass bottle, why Al is used? The answer was that they did not want to cut the product.
- The objective was to check the storage stability in an inert container. The commercial package is still not available.
- How they checked that the extraction was complete?
- All the checked samples were too good if it is possible also a weaker formulation is needed for the tests.
- Several extensions of the methods are published in different handbooks (MT 46.1; 46.2; 46.3 etc.) it is difficult to handle them.

5.3 Wash resistance index of long lasting insecticide net (Duranet Plus) containing alpha-cypermethrin 0.58 % w/w and piperonyl butoxide 0.2 % w/w by Mr Atmakuru Ramesh (5053)

Mr Ramesh presented a study for the determination of the wash resistance index of Duranet Plus containing alpha-cypermethrin 0.58 % w/w and piperonyl butoxide 0.2 % w/w. Pieces of the net were subjected to successive washing-rinsing-heating cycles and the total active ingredient content was determined in washed samples after a number of washings. The wash resistance index was determined by the decrease of the total active ingredient content after several cycles using the appropriate analytical method following CIPAC MT 195.

Alpha-cypermethrin content in the net sample was determined by GC-FID using CIPAC 454/LN/M/3.2 as reference method.

Piperonyl butoxide content in the long lasting insecticide net was determined by GC-FID using CIPAC 33/LN/M/3.0 as reference method.

The wash resistance index was calculated after the 4 washing-rinsing-heating cycles.

The retention indexes calculated from data before wash and after 4^{th} wash were 99.54% and 93.56%.

The following comments were received from the meeting:

- In case of alpha-cypermethrin three point calibration was used in case of piperonyl butoxide five point calibration was used. Answer: linearity of the calibration was excellent also with three points.

No other comments or questions.

6. Revision/update of CIPAC guidelines

The Chairman remarked that the process of revising or updating of our CIPAC guidelines goes on. The meeting is requested to send in comments on all guidelines available on the website.

7. Replacement of obsolete methods, comments to existing methods

7.1 Comments to existing methods

Errata: retention order of the peaks in the published CIPAC/454 method is not correct. The correct order will be published on the website as an errata. (alpha-cypermethrin cisI approx. 27 min, alpha-cypermethrin *cis* II : approx. 29 min)

Comments on HORRAT are welcomed up to the end of July.

7.2 Revision of MT methods

No contribution was made to this point.

The Chairman, Mr Hänel, thanked all presenters, all participants of the collaborative studies and all meeting participants for their extensive comments.
Mr Hänel declared the open meeting closed.

8. Minutes of the 59th meeting (5031/P)

The minutes were circulated to the participants by e-mail and are available on the website. No comments were received, as a conclusion the minutes are accepted as a true record of the last year meeting.

9. Secretary's report (5032/P)

Mr Bura presented the Secretary's report for the period from the 59th CIPAC meeting held in Athens, Greece. The report had been previously circulated to members by e-mail. No comments were received.

10. Discussion of individual compounds

635 Silthiofam

At the 59th meeting, 2015 in Athens the method was accepted as provisional. No further comments were received.

The method can be promoted to a **full CIPAC method**.

715 Pyriproxyfen, retention properties

The method for the determination of retention properties of pyriproxyfen matrix release formulations (CIPAC/4999) was accepted as a tentative CIPAC MT method at the 59th meeting, 2015 in Athens with the request of additional validation data. The status of the method remains tentative as an additional 4 laboratories data will be presented in the next CIPAC TC meeting. It was proposed to use an extraction at room temperature for 24 hours or the same with a footnote of the other option.
This should be clarified with Sumitomo before goes to full method.
The extension of the scope (CIPAC/4997) of CIPAC method 715/TC/M/3 for the determination of the pyriproxyfen content of a matrix release formulation (MR) (incorporated type) was accepted as **full** CIPAC method

794 Chlorantraniliprole

The meeting discussed the comments received during the open meeting. A high column back pressure was notified at the specified flow rate by one participant, who asked to mention in the trial protocol that stainless steel capillary connections should be used during the measurements instead of PEEK tubing. I was agreed that the problem of tubing of the HPLC instrument is a technical hardware problem and it is not closely related to the method. The method presented is a robust method. The key issue is the resolution between the a.s. and the impurity, the flow rate can be reduced because of the pressure if the resolution is maintained.
The method was accepted as **provisional CIPAC method**.

738 Clothianidin method extension to WP formulation

The method was accepted as **provisional CIPAC method**.

983 Cyenopyrafen

It was proposed to go for full scale collaborative study with some amendments in the text of the method description concerning the sample preparation for the SC formulation.

989 d-tetramethrin

The problem with the isomer ratio was clarified. The Company should apply for ISO common name.
It was proposed to go for full scale collaborative study with the modification of using heptane instead of hexane.

693 Fenazaquin

The method was accepted as **provisional CIPAC method**.

987 Flupyradifurone

Proposed for full scale collaborative trial, with the note of clarifying before the start of the trial whether 15 min of sonication is needed, or can be reduced.

34 Mancozeb

It was proposed for full scale collaborative trial with some notes: the recommended temperature possibly be 30 °C, clarification of the pH value of the eluent and its usability with the column, the importance of using the proper analytical standard. It was proposed to change the wording to "freshly prepared" instead of "not after 24 h". The Company should also provide an identity test.

5.1 Extension of 715/TC/M/ to pyriproxyfen/alpha cypermethrin Royal Guard LN (CIPAC/4886/m)

It was confirmed that the existing extension of the scope (CIPAC/4887) of CIPAC method 715/TC/M/2 for the determination of the pyriproxyfen content of a the long lasting insecticidal mosquito net (incorporated type) (LN) containing permethrin and pyriproxyfen is applicable for the long lasting insecticidal mosquito net containing pyriproxyfen and and alpha-cypermethrin.

The extension of the scope (CIPAC/5043) of CIPAC method 454/LN/M/3.2 for the determination of the alpha-cypermethrin content of the long lasting insecticidal mosquito net (incorporated type) (LN) containing alpha-cypermethrin and pyriproxyfen, with the modification of having di-cyclohexyl phtalathe as internal standard, instead of dioctyl phtalathe, was accepted as a tentative CIPAC method, with the need for the provision of a second data set according to the provisions of the CIPAC guideline.

5.2 Extension of MT 46.3 to matrix release formulation

Several extensions of the methods are published in different handbooks (MT 46.1; 46.2; 46.3 etc.) it is difficult to handle them. It was proposed to revise the old method and to include in it the MR method.
It was decided that Mr Pigeon will make a comparison of the existing variants of the MT 46.3 and the proposed decision will be communicated to the Company.
The method was accepted as a tentative CIPAC MT method.

5.3 Wash resistance index of long lasting insecticide net Duranet Plus

It was accepted that the applicability of the method has been proven.

11. Matters related to FAO and WHO specifications

Mrs Yang presented some general comments regarding CIPAC work. It was mentioned the problem of impurities and relevant impurities.
The Chairman, Mr Hänel pointed out that the methods are available on the web site. Mrs Yang said that a link will be placed on the FAO website to the methods for the relevant impurities on the CIPAC website.
Mr Jean Philippe Bascou mentioned that it would be useful to receive a list of obsolete methods. The Chairman answered that CIPAC will not publish such a list with the obsolete methods, but of methods no longer supported.

12. Any other business

It was proposed to indicate on the collaborative trials information sheet that the results of the trial should be sent to the participants at least two weeks before the CIPAC meeting.

13. Closure

The Chairman thanked the organising team and the participants for their contribution to the success of the meeting and closed the meeting.

Lajos Benke	László Bura
temp. assist. secretary (July 2016)	secretary