Ring Test Certified Standard Solutions

(2018)

Introduction

An increasing number of analytical laboratories are buying certified standard solutions from external companies for quantitative purposes, since it leads to important savings in terms of time and laboratory work. However, there is not enough evidence of the quality of these solutions. The present Ring Test is aimed at verifying the comparability of four certified standards solutions containing 28 pesticides present in the priority list of the European Union Multi-Annual Control Programme (EU-MACP).

Test items

This Ring Test is based on the analysis of four custom certified standard solutions that contain a number of LC amenable pesticides included in the scope of the EU-MACP. The solutions were prepared by four external specialised firms, that are kept confidential, with a certified concentration of 50 mg/L. They were diluted to 1 mg/L in acetonitrile by the EURL for Pesticide Residues in Fruit and Vegetables. The samples were stored at -20°C prior to shipment.

Participants received diluted aliquots of the four standard solutions. These aliquots were sent in glass vials sealed with parafilm, containing 1 mL of each solution in acetonitrile. The expected concentration of each pesticide in the vials was 1 mg/L. For a detailed description of the pesticides in each solution and their concentrations, see Appendix 1.

Steps followed

The procedure to follow by each one of the seven participant laboratories was:

1. The samples were delivered by courier to the participant laboratories on week 16 (April 16th, 2018) in polystyrene boxes containing dry ice. A message was sent out by e-mail the day of the shipment.

2. The standard solutions were diluted 20 times (that way the concentration values were expected to be 0.050 mg/L). This dilution was performed following the laboratory´s own procedures using the appropriate solvents for each analytical technique.

3. Each solution was analysed six times in pure solvent by LC chromatography, alternating with the laboratory's own standard solution following the sequence: Lab std. mix, A, B, C, D, Lab std. mix, A, B, C, D…

4. The areas of the compound signals for each injection were reported. Additionally, concentrations were calculated using the laboratory's own standard solutions and reported as well.

5. Participant laboratories submitted their results before the end of July 2018. For this purpose, each participant received an Excel document to be filled in with instrumental data (equipment used, polarity, mobile phase and solvent used) and results obtained.

Data treatment

The first step was to discard the differing results, which were considered to be caused by errors committed in the laboratories. With that purpose, two criteria were followed:

1. To maintain at least five results per pesticide. Therefore, no more than two results were discarded in each case.
2. The deleted result had to be at least 20% different from the average result of the pesticide.

After discarding the differing results, the average concentration and coefficient of variation (CV) for each pesticide were calculated (see Table 1).

Table 1. Average concentration and coefficient of variation of each pesticide in the solutions								
	Solution A		Solution B		Solution C		Solution D	
	Conc. (mg/L)	CV (%)	Conc. (mg/L)	CV (%)	Conc. (mg/L)	CV (%)	Conc. (mg/L)	CV (%)
2,4-D	0.046	9	0.046	6	0.045	5	0.052	7
Abamectin	0.053	14	0.059	12	0.056	37	0.059	17
Acephate	0.048	9	0.048	8	0.051	7	0.052	8
Acetamiprid	0.048	10	0.048	9	0.050	10	0.054	8
Ametoctradin	0.057	4	0.029	10	0.035	6	0.058	6
Bromuconazole	0.048	7	0.047	5	0.051	8	0.049	6
Bupirimate	0.047	10	0.046	9	0.046	9	0.057	6
Carbaryl	0.048	7	0.052	7	0.051	7	0.055	6
Carbosulfan	0.043	41	0.035	44	0.038	42	0.052	16
Cyromazine	0.048	31	0.050	13	0.047	26	0.057	7
Demeton-S-methylsulfoxide	0.056	8	0.051	10	0.062	7	0.061	7
Diuron	0.049	5	0.050	9	0.052	3	0.056	4
Emamectin benzoate	0.029	49	0.024	57	0.036	53	0.020	55
Fenamiphos – sulfoxide	0.052	9	0.049	10	0.036	9	0.057	8
Fenthion	0.049	8	0.047	9	0.047	9	0.049	6
Fipronil	0.046	10	0.048	8	0.046	8	0.055	9
Haloxyfop	0.049	9	0.051	10	0.048	9	0.054	9
Kresoxim methyl	0.051	9	0.051	6	0.048	8	0.055	9
Malathion	0.049	7	0.047	10	0.046	6	0.057	7
Methiocarb sulfone	0.049	26	0.046	31	0.057	52	0.036	26
Omethoate	0.050	9	0.055	19	0.053	17	0.043	18
Procymidone	0.050	25	0.051	27	0.046	24	0.058	24
Prothioconazole-desthio	0.048	4	0.049	9	0.047	4	0.062	5
Spinosad	0.047	20	0.036	33	0.041	21	0.054	6
Thiobencarb	0.048	10	0.051	8	0.046	7	0.052	7
Triadimefon	0.050	6	0.052	7	0.050	6	0.061	7

Especial case: Benomyl and carbendazim

Benomyl is a very unstable pesticide that decomposes into carbendazim. By including both of these compounds in our request of the four solutions, we intended to evaluate the reaction of the companies.

Company A provided both pesticides in its solution, but in separated vials so that one did not interfere with the other one's quantification. For its part, company B included only benomyl in their solution. It would have been correct, if their reasoning had been that benomyl and its metabolite should not be mixed together. Instead, they alleged low solubility of carbendazim in the solvent used. Company C included only carbendazim and alleged solubility problems of benomyl. On the other hand, Company D had no objections to including both pesticides together in the same solution, even though this would make it impossible to quantitate correctly none of them.

Assessment of the solutions

Only those pesticides that showed consistent results in all cases were used for the general assessment of the solutions. With that purpose, a maximum inter-laboratory CV of 10% was stablished to ensure that most participants reported very similar concentrations. Eighteen out of the twenty-eight pesticides met this criterion and could be used for the assessment of the solutions.

To identify the acceptable results for this study, a maximum deviation of 10% with regard to the certified concentration was stated. Therefore, all concentrations between 45 and 55 mg/L were considered as correct.

As can be seen in table 1, ten pesticides had an acceptable concentration in the 4 solutions: 2,4-D, acephate, acetamiprid, bromuconazole, carbaryl, fenthion, fipronil, haloxyfop, kresoxim-methyl and thiobencarb. On the other hand, 5 pesticides showed an unacceptable concentration only in one solution, while being correct in the other three solutions. The solution which presented a concentration level deviated from the rest of the solutions was always solution D, with concentrations higher than the maximum limit of 55 mg/L. These pesticides were bupirimate, diuron, malathion, prothioconazole-desthio and triadimefon. The other 3 pesticides (fenamiphos-sulfoxide, demeton-S-methylsulfoxide and ametoctradin) had an unacceptable concentration in two or more solutions. Both fenamiphos-sulfoxide and ametoctradin showed a remarkably low concentration in at least one solution (for example, 0.029 mg/L reported for ametoctradin in solution B).

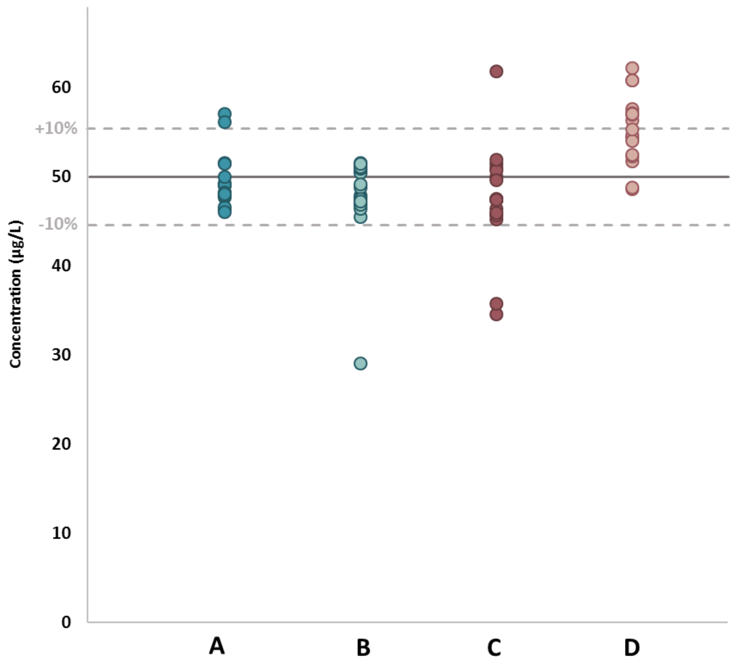


Figure 1. Concentrations found for the 18 pesticides in the four certified solutions.

Individual studies

For the following 8 pesticides, there was not a consensus in the results reported by the participants. These pesticides did not meet the previous criteria of inter-laboratory CV lower than 10% for at least 5 participants. Therefore, a maximum CV of 15% and a minimum number of results of 4 were accepted.

Abamectin, emamectin benzoate and spinosad

Abamectin showed inter-laboratory coefficients of variation greater than 40% in the four solutions (figure 2, left). This pesticide is a mixture of two components: avermectin B1a and avermectin B1b, which differ in one lateral chain. Different ways of quantifying the total concentration of abamectin, together with a different proportion of both avermectins in the solutions used by each participant to quantify this compound may have influenced the reported concentrations.

However, participants 1and 4 reported concentrations that were much lower than the rest of participants in all solutions. If their results are not considered, coefficients of variation are now much lower in solutions A and B and thus, the average concentrations are more reliable (figure 2, right).

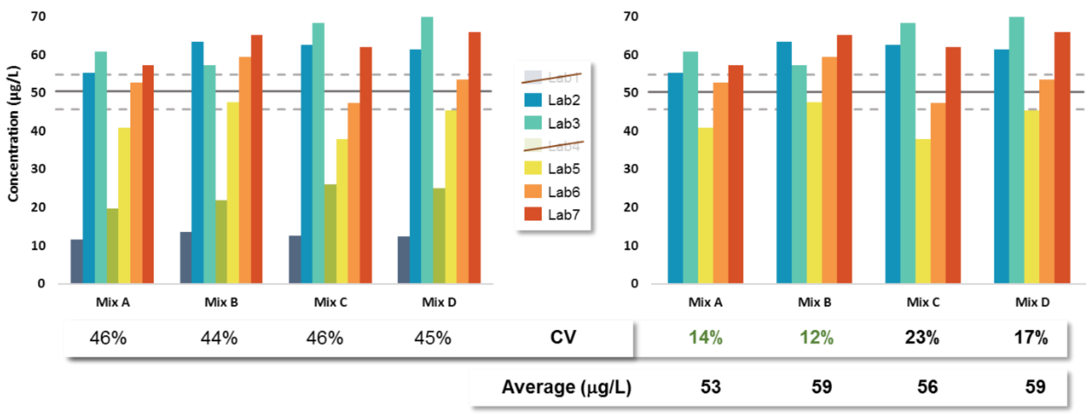


Figure 2. Concentrations reported for abamectin in the four solutions. Left: considering the seven participants. Right: discarding the results of participants 1 and 4.

The standard of emamectin benzoate also contains a mixture of 2 components: emamectin B1a and emamectin B1b. The structural difference is one lateral chain. If the low results reported by participants 1 and 4 are not used for the calculations, low CVs are obtained in solutions A, C and D (figure 3). It is noteworthy that the average concentration was in most solutions much lower than the certified value.

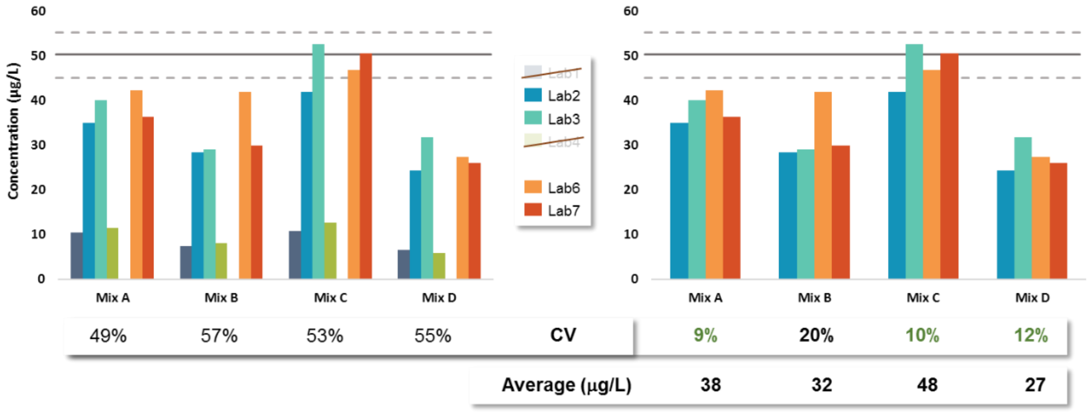


Figure 3. Concentrations reported for emamectin benzoate in the four solutions. Left: considering the six participants that reported results. Right: discarding the results of participants 1 and 4.

The same reasoning was applied to spinosad (figure 4). This pesticide consists of spinosyn A and spinosyn D, which has an extra methyl group. Participants 1 and 4 reported very low concentrations. If they are not used, CVs below 10% are obtained in two of the solutions (Solution A and D). Moreover, the average concentrations in these solutions are within the accepted range.

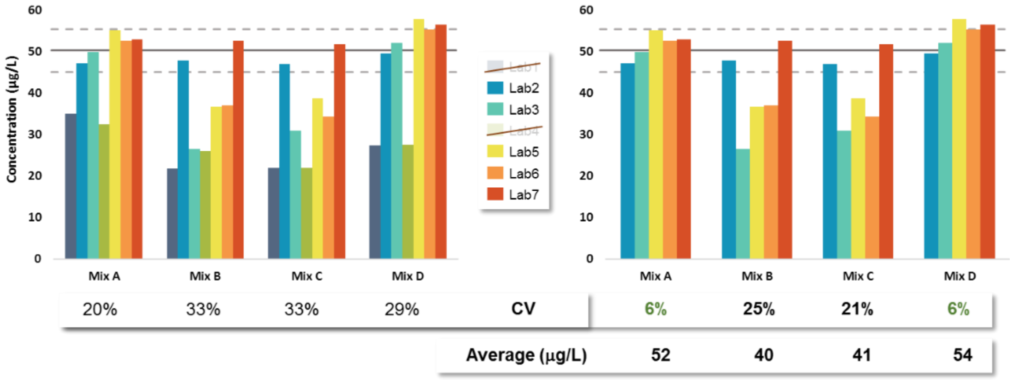


Figure 4. Concentrations reported for spinosad in the four solutions. Left: considering the seven participants. Right: discarding the results of participants 1 and 4.

<u>Carbosulfan</u>

Carbosulfan is a compound prone to degradation during the analysis and, moreover, this pesticide was not in the scope of participants 2 and 4. However, most of the variability was due to the results of participant 1, which were much lower than the rest. After discarding them, acceptable concentrations and CVs were achieved in solution A and C (figure 5).

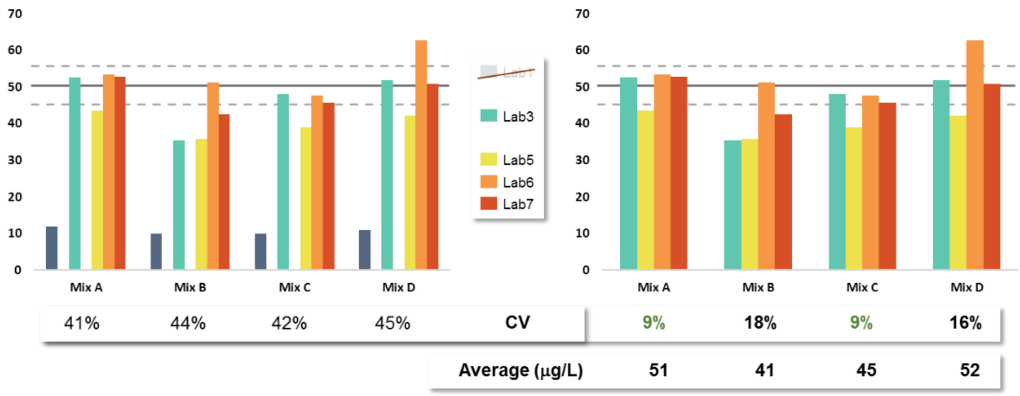


Figure 5. Concentrations reported for carbosulfan in the four solutions. Left: considering the five participants that reported results. Right: discarding the results of participant 1.

Cyromazine

Cyromazine is a difficult compound as a consequence of its high polarity. The differing results for this pesticide were mainly reported by participants 3 and 5 (figure 6). Lab 3 reported a poor peak shape for this compound, which may have caused their high results. However, the rest of participants reported consistent results in almost all cases (except solution A), with acceptable concentration values in most solutions.

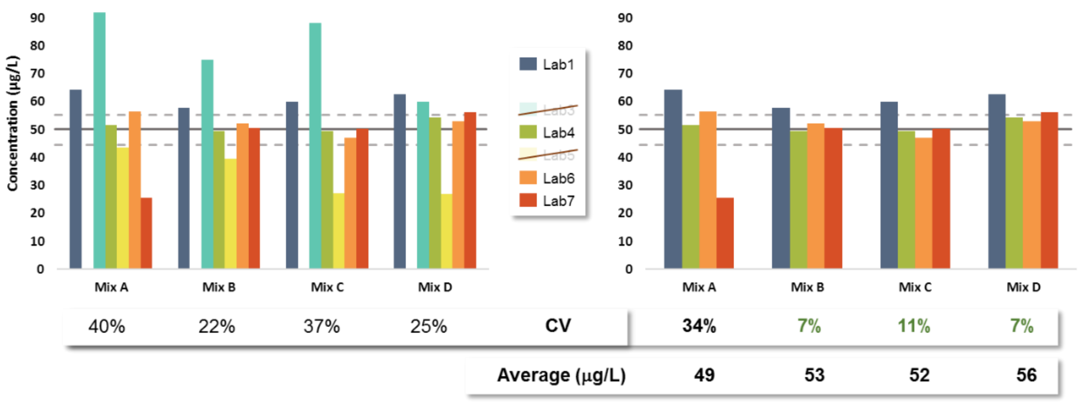
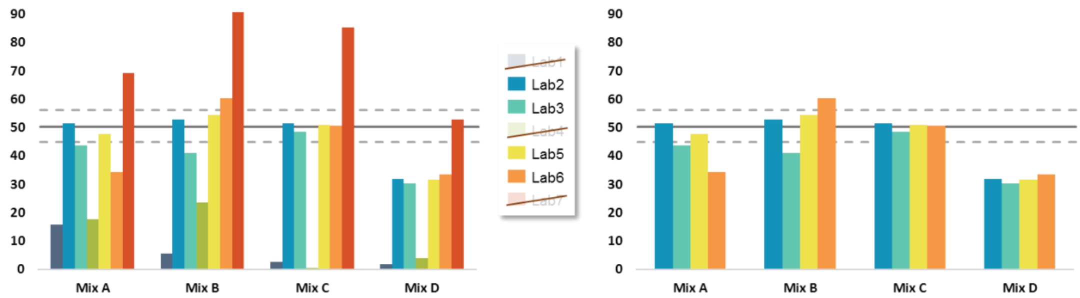


Figure 6. Concentrations reported for cyromazine in the four solutions. Left: considering the six participants that reported results. Right: discarding the results of participants 3 and 5.

Methiocarb-sulfone

In the case of methiocarb-sulfone, it was necessary to discard the results of three participants in order to obtain acceptable coefficients of variation (figure 7). The results reported by participants 1 and 4 were extremely low, whereas those reported by participant 7 were remarkably high. The resulting concentrations were also very different among the different solutions, ranging from 0.032 to 0.052 mg/L.

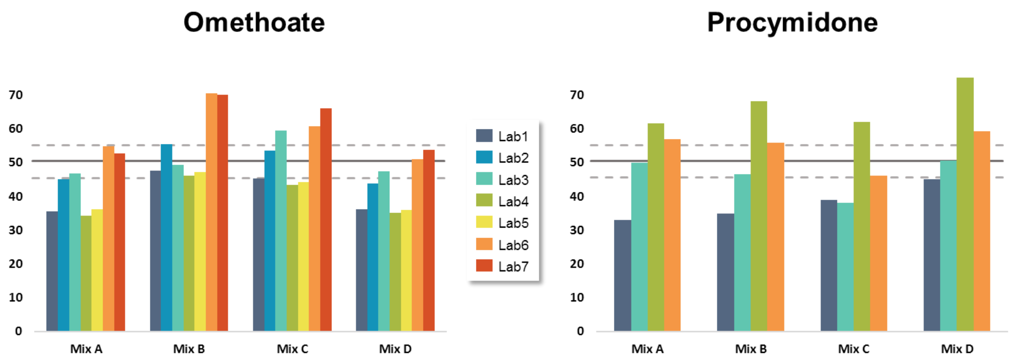


48%	58%	72%	67%	CV	17%	16%	3%	4%
				Average (µg/L)	44	52	51	32

Figure 7. Concentrations reported for methiocarb-sulfone in the four solutions. Left: considering the seven participants. Right: discarding the results of participants 1,4 and 7.

Omethoate and procymidone

In the case of omethoate (figure 8, left) and procymidone (figure 8, right), any participant reported concentrations that were significantly different from the rest. Therefore, any results could be discarded and the average concentrations of the solutions are not as reliable as the rest.



19%	19%	17%	18%	CV	25%	27%	24%	24%
44	55	53	43	Average (µg/L)	50	51	46	58

Figure 7. Concentrations reported for omethoate (left) and procymidone (right) in the four solutions.

Conclusions

If these data are used to assess the solutions, it can be concluded that for solutions A, B and C, only a few pesticides did not show a concentration within the accepted range of 45-55 mg/L. However, in solution D, almost half of the pesticides had a concentration higher than 55 mg/L (with deviation greater than 10% with regard to the certified value), so this solution was clearly unsatisfactory.

Considering the 18 pesticides and the 4 solutions, 80% of the average concentrations were correct for the pesticides. However, considering that something could have gone wrong in solution D and this solution is not used for the calculation, the percentage of correct results goes up to 88%.

However, in some cases, the companies do not have enough knowledge of the behavior of some pesticides in solution, as in the case of benomyl and carbendazim.

APPENDIX I. Original composition of solutions

Solution A. Five separated solutions were received from this company, being the solvent in all cases acetonitrile:

Solution 1:

Pesticide	Concentration (mg/L)
2,4-D	50.25
Abamectin	50.27
Acephate	50.17
Acetamiprid	50.19
Ametoctradin	50.19
Bromuconazole	50.24
Bupirimate	50.33
Carbaryl	50.30
Carbosulfan	50.21
Demeton-S-methylsulfoxide	50.00
Diuron	50.28
Emamectin benzoate	50.30
Fenamiphos - sulfoxide	50.41
Fenthion	50.23
Fenthion-oxon	50.24
Fipronil	50.22
Haloxyfop	50.23
Kresoxim methyl	50.20
Malathion	50.23
Methiocarb sulfone	50.19
Omethoate	50.39
Procymidone	50.32
Prothioconazole-desthio	50.16
Spinosad	50.47
Thiobencarb	50.29
Triadimefon	50.36

Solution 2:

Benomyl	100.01

Solution 3:

Carbendazim	100.00

Solution 4:

Cyromazine	100.00

Solution 5:

Formetanate	100.00

Solution B. One solution was received from this company containing all the analytes, being the solvent acetonitrile:

Pesticide	**Concentration (mg/L)**
2,4-D	50.20
Abamectin	50.20
Acephate	50.00
Acetamiprid	50.10
Ametoctradin	50.00
Benomyl	50.20
Bromuconazole	50.20
Bupirimate	50.10
Carbaryl	50.20
Carbosulfan	50.00
Cyromazine	50.20
Demeton-S-methylsulfoxide	50.00
Diuron	50.20
Emamectin benzoate	50.10
Fenamiphos - sulfoxide	50.20
Fenthion	50.10
Fipronil	50.10
Formetanate	50.00
Haloxyfop	50.10
Kresoxim methyl	50.20
Malathion	50.00
Methiocarb sulfone	50.00
Omethoate	50.20
Procymidone	50.20
Prothioconazole-desthio	50.00
Spinosad	50.10
Thiobencarb	50.00
Triadimefon	50.20

Solution C. Two solutions were received from this company, being the solvent acetonitrile in solution 1 and acetonitrile:methanol (50:50, v/v) for solution 2:

Solution 1:

Pesticide	**Concentration (mg/L)**
2,4-D	50.00
Bupirimate	50.10
Carbaryl	50.40
Carbosulfan	49.20
Fenamiphos - sulfoxide	49.70
Fenthion	49.00
Fipronil	50.00
Haloxyfop	50.10
Kresoxim methyl	49.80
Malathion	49.50
Methiocarb sulfone	50.70
Omethoate	49.80
Procymidone	49.80
Thiobencarb	50.10
Triadimefon	49.30

Solution 2:

Abamectin	50.30
Acephate	50.70
Acetamiprid	50.20
Ametoctradin	50.50
Bromuconazole	50.20
Carbendazim	49.70
Cyromazine	50.20
Demeton-S-methylsulfoxide	50.30
Diuron	49.80
Emamectin benzoate	49.70
Fenthion-oxon	49.50
Formetanate	50.20
Prothioconazole-desthio	49.80
Spinosad	49.60

Solution D. One solution was received from this company containing all the analytes, being the solvent acetone:

Pesticide	**Concentration**
2,4-D	50.00 ± 2.68
Abamectin	50.00 ± 2.68
Acephate	50.00 ± 2.68
Acetamiprid	50.00 ± 2.68
Ametoctradin	50.00 ± 2.68
Benomyl	50.00 ± 2.68
Bromuconazole	50.00 ± 2.68
Bupirimate	50.00 ± 2.68
Carbaryl	50.00 ± 2.68
Carbendazim	50.00 ± 2.68
Carbosulfan	50.00 ± 2.68
Cyromazine	50.00 ± 2.68
Demeton-S-methylsulfoxide	50.00 ± 2.68
Diuron	50.00 ± 2.68
Emamectin benzoate	50.00 ± 2.68
Fenamiphos - sulfoxide	50.00 ± 2.68
Fenthion	50.00 ± 2.68
Fenthion-oxon	50.00 ± 2.68
Fipronil	50.00 ± 2.68
Formetanate	50.00 ± 2.68
Haloxyfop	50.00 ± 2.68
Kresoxim methyl	50.00 ± 2.68
Malathion	50.00 ± 2.68
Methiocarb sulfone	50.00 ± 2.68
Omethoate	50.00 ± 2.68
Procymidone	50.00 ± 2.68
Prothioconazole-desthio	50.00 ± 2.68
Spinosad	50.00 ± 2.68
Thiobencarb	50.00 ± 2.68
Triadimefon	50.00 ± 2.68