# LABORATORY PERFORMANCE DURING PROFICIENCY TESTS FOR THE ANALYSIS OF ACTIVE SUBSTANCES IN PESTICIDE FORMULATIONS DURING 2021-2023

A. Santilio*, R. Cammarata**, Silvana Girolimetti*

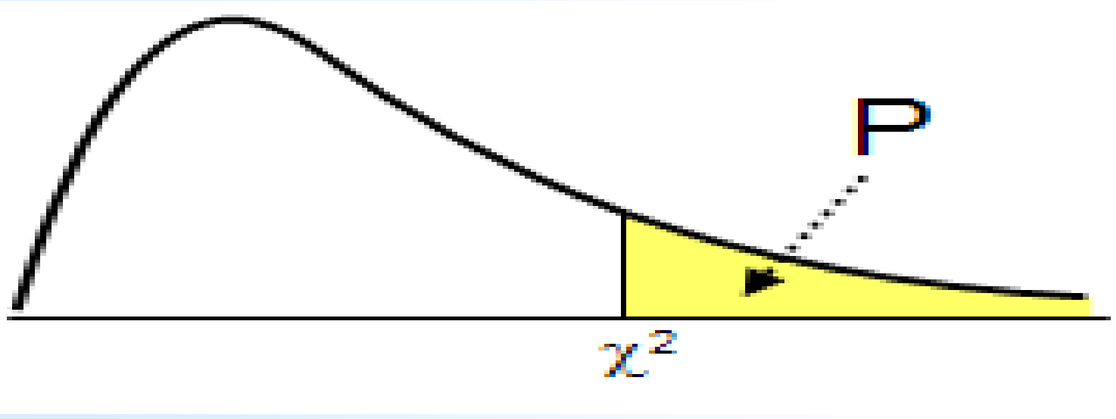
*Italian National Institute of Health, Department of Environment and Health—ECASS Unit
**Italian National Insitute of Health—Central Directorate of Human and Economic Resources
V.le Regina Elena, 299—00161 Rome (IT)
Email: angela.santilio@iss.it; roberto.cammarata@iss.it

10th Eurachem Workshop on Proficiency Testing in Analytical Chemistry, Microbiology and laboratory Medicine - Windsor (UK) 25-28/09/2023

European Regulation (CE) n.1107/2009 gives provisions for the authorisation, placing on market, use and control of Plant Protection Products (PPPs). The rules established through the evaluation of risk assessments warrant a high level of protection for both human and animal health and for the environment. These rules assure the functioning of the EU market and improvement of agricultural production. The EU regulation n.2017/625, replacing Regulation (EC) n.882/2004, expands and improves provisions for the official controls and other official activities, performed to ensure the application of food and feed law, rules on animal health and welfare, plant health and plant protection products. In this framework, official laboratories are appointed by the competent authorities to perform analysis on PPP samples. They shall possess appropriate expertise, well-performing equipment and qualified staff, as well as being accredited, according to ISO/IEC 17025, for the tests required under this framework. Independent demonstration of competence through the successful participation in PT is a requirement for accreditation, therefore it is important to organize PTs for the active substance content for the laboratories involved in this activities.

| Active substance | Formulation Type | Declared Concentration (%, w/w) | Homogeneity Significativity level (P) | Stability Deviation first analysis; Deviation to declared content |
|---|---|---|---|---|
| Bentazone | Water Soluble Granule | 87.0 | 0.07 | 1.4%; 3.3% |
| Cyprodinil | Emulsifiable Concentrate | 30.0 | 0.81 | 0.39%; 2% |
| Deltametrin | Dustable powder | 0.02 | 0.65 | -0.79%; -6.5% |
| Fehnexamid | Suspension concentrate | 42.8 | 0.19 | 4.83%; -2.45% |
| Imazamox | Soluble concentrate | 7.62 | 0.18 | -3.94%; -5.25% |
| Spinosad | Soluble Concentrate | 44.2 | 0.29 | 2.9%; 2.5% |
| Tau-fluvalinate | Emulsion | 21.4 | 0.03 | 1.9%; 2.9% |
| Trifloxystrobin | Water dispersible granule | 50.0 | 0.35 | -1.9%; -2.6% |
| Trinexapac-ethyl | Capsule suspension | 11.3 | 0.57 | -5.50%; -4.21% |

During the period 2021-2023, Italian National Institute of Health organised, in collaboration with the Ministry of Health, PTs for about 25 national and other EU laboratories involved in the monitoring programmes fo PPPs even if in 2021 there was a decrease of the participants, due to the COVID pandemic. Table shows the formulation type and active substances and declared concentration (%w/w).
**Homogeneity tests**— ten bottles were randomly chosen and analysed in duplicate, in two different days. Considering that the σPT is unknown, the statistically significant differences between PT items used were evaluated with the analysis of variance T-test at α=0.05, if the data series are more than two the Fisher Test is necessary. The T-test shows a significativity level (P) higher than 0.05 for each active substance. It is possible to say the samples are not different one each other: they are homogeneous.
**Stability tests**— The stability test was performed using two bottles, randomly chosen, which were analysed in duplicate in two occasions and each occasion twice:
– Day 1: before the shipment of the samples;
– Day 2: at the deadline for reporting results.
Stability test was judged acceptable as the percentage difference of concentration for each active substance was found less than 10%.




$$Z_i = 0{,}6745 * (X_i - \text{Median}) / \text{MAD}$$

**$-3{,}5 \le Z_i \le 3{,}5$** Z values falling outside the range were marked as **outliers.**

**$-3{,}5 \le Z_i \le -1{,}96$ & $1{,}96 \le Z_i \le 3{,}5$**

Z values falling in these range were marked as **questionable**

The **statistical evaluation** started by applying the Jarque-Bera test to verify the hypothesis of normality of the data distributions. To apply this test asymmetry and curtosi of the data distributions were calculated and used in the Jarque-Bera formula:

Jarque-Bera: $[(\mathbf{GdL}^{\wedge(\text{Asimmetria};2)}/6)+(\mathbf{GdL-1})^{\wedge(\text{curtosi};2)}/(\mathbf{GdL+1})]$

Jarque-Bera = **2,651977**

$\chi^2 = (\mathbf{0,95;2}) = \mathbf{5,991465}$ (Critical value)

**JB < $\chi^2$** Normal distribution JB acceptance region

The results were compared with critical values from the χ2 distribution at a 95th percentage probability level.
After this verification, the performance of each participant for each sample was calcuated with the following formula:

$$\mathbf{Z_i = 0.6745*(X_i — Median)/MAD}$$

Where:
Xi = data
MAD = the median absolute deviation
0.6745 = constant come from MAD calculation.

**Robust Mean**
The purpose of using a robust estimator for the mean was to cope with the possibility of outlying data points without having to remove them from the sample.
The robust mean estimator used was the median.
**Robust Estimate of Standard Deviation**
The robust estimate of the standard deviation used was the $MAD_E$ value. To obtain the $MAD_E$, calculate Median Absolute Deviation (MAD) from the sample median:
$MAD = median\ (|X_i - median\ (Xi)|_{i=1,2...n})$
Calculate $MAD_E$:
$MAD_E = K \times MAD$
For a data series with Normal distribution $K \approx 1{,}483$

| Lab # | MODIFIED Z SCORE (2021) | | |
|---|---|---|---|
| | IMAZAMOX | FENEXAMID | TRINEXAPAC-ETHYL |
| 1 | 0.73 | -0.27 | 0.20 |
| 2 | -0.85 | -0.13 | 0.01 |
| 3 | 0.00 | 0.67 | -0.38 |
| 4 | 0.70 | -0.00 | 0.39 |
| 5 | 1.62 | -0.13 | 0.16 |
| 6 | -0.31 | 1.01 | -1.34 |
| 7 | -- | -- | -1.15 |
| 8 | 1.55 | 0.33 | -- |
| 9 | 0.02 | -0.27 | 0.57 |
| 10 | 1.55 | 0.13 | 1.88 |
| 11 | -0.81 | -2.43 | -1.05 |
| 12 | -0.09 | --- | -1.98 |
| 13 | 0.12 | 0.27 | 0.59 |
| 14 | -0.82 | -0.78 | -0.57 |
| 15 | -0.10 | -3.10 | 1.24 |
| 16 | 0.09 | 0.29 | 0.41 |
| 17 | -0.65 | -0.94 | -0.57 |

| Lab # | MODIFIED Z SCORE (2022) | | |
|---|---|---|---|
| | BENTAZONE | SPINOSAD | TAU-FLUVALINATE |
| 1 | -0.429 | -1.781 | 0.487 |
| 2 | -0.308 | -0.027 | 0.499 |
| 3 | -- | 0.681 | 0.545 |
| 4 | -0.509 | -1.140 | -0.334 |
| 5 | -0.477 | -1.862 | 0.393 |
| 6 | 1.746 | **3.514** | 2.833 |
| 7 | 0.497 | 0.007 | --- |
| 8 | -2.366 | 0.084 | -1.924 |
| 9 | 0.344 | -0.121 | -1.595 |
| 10 | -0.469 | 3.177 | -2.387 |
| 11 | 0.819 | 0.142 | -0.862 |
| 12 | 2.954 | 0.007 | 1.015 |
| 13 | --- | --- | --- |
| 14 | -2.458 | -1.781 | -0.199 |
| 15 | 0.530 | -0.890 | 0.679 |
| 16 | 0.163 | -0.034 | -0.006 |
| 17 | 0.968 | 0.128 | -0.657 |
| 18 | --- | --- | --- |
| 19 | --- | --- | --- |
| 20 | 2.269 | 1.457 | -1.490 |
| 21 | -1.315 | --- | --- |
| 22 | -1.126 | --- | 3.026 |

| Lab # | MODIFIED Z SCORE (2023) | | |
|---|---|---|---|
| | CIPRODINIL | DELTAMETRIN | TRIFLOXYSTROBIN |
| 1 | 1,656 | --- | -0,716 |
| 2 | 0,652 | -0,405 | 0,142 |
| 3 | 0,150 | 0,675 | 0,366 |
| 4 | 1,356 | --- | 0,533 |
| 5 | -0,817 | -1,147 | 1,541 |
| 6 | 3,298 | 0,675 | 2,531 |
| 7 | 0,675 | -0.202 | 0,450 |
| 8 | -1,102 | --- | -0,333 |
| 9 | -0,180 | 2.765 | 0,283 |
| 10 | -0,375 | 0.675 | -1,357 |
| 11 | -0,247 | --- | -1,232 |
| 12 | -1,349 | -0,675 | 1,282 |
| 13 | -0,225 | -0.202 | -0,633 |
| 14 | -1,626 | 0,202 | -1,949 |
| 15 | -0,150 | -0.337 | -2,298 |
| 16 | 0,150 | -1.21 | 1,832 |
| 17 | 0,854 | 0,540 | -0,608 |
| 18 | 1,274 | -1.349 | 0,200 |
| 19 | 0,600 | 1,282 | 1,949 |

**CONCLUSION**
The outcome of the ITPTs can be considered satisfactory.
The participation of the Italian and European laboratories was good.
The performance of the laboratories expressed in terms of modified z-score were satisfactory by almost all participants for all substances, except for one of them who got higher than -3.5 z-score value for the active substances Spinosad.
Based on the results it can be concluded that the PT was successfully organized also based on the number of participants.