

VALIDATION OF A QUICK TEST TO DETECT NOSEMA SP.

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- Nosema disease is caused by microsporidian *Nosema sp.*
- Affects the digestive tract of and produces nonspecific symptoms
- Can be confused with other disorders
- For this reason it is essential to resort to laboratory diagnosis to detect and quantify the causative agent

Currently uses the technique of modified Cantwell:

- Considerable time to get the results
- No time for adopt therapeutic measures or precocious way management
- Not be counteract the harmful effects of the illness and the economic damage it causes.
- Is late for the implementation of drug treatments for not respecting withdrawal period with the consequent risk of contamination of honey and other products of the hive

The technique of the OIE (World Organization for Animal Health):

- Does possible diagnosis with 10 bees
- High sensitivity, high specificity and speed in its implementation
- Less time for sample collection
- Fewer preservatives and size of bottles.
- Low cost???

The efforts of this work were developed in the framework of the Argentine Northwest Agricultural Schools Network and were geared towards validation and adaptation of OIE technique to position it as a usual diagnostic tool

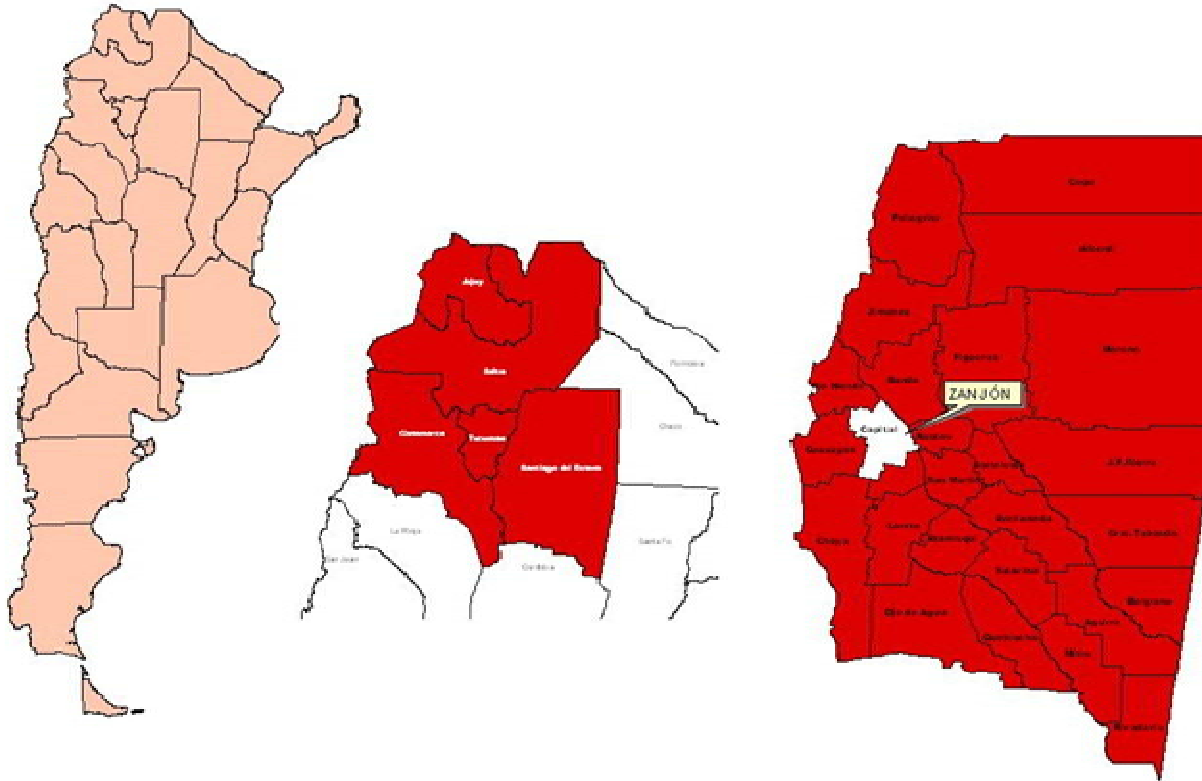
The hypothesis: it is possible to use the technique of the OIE as routine technique?

Development, Materials and Methodology

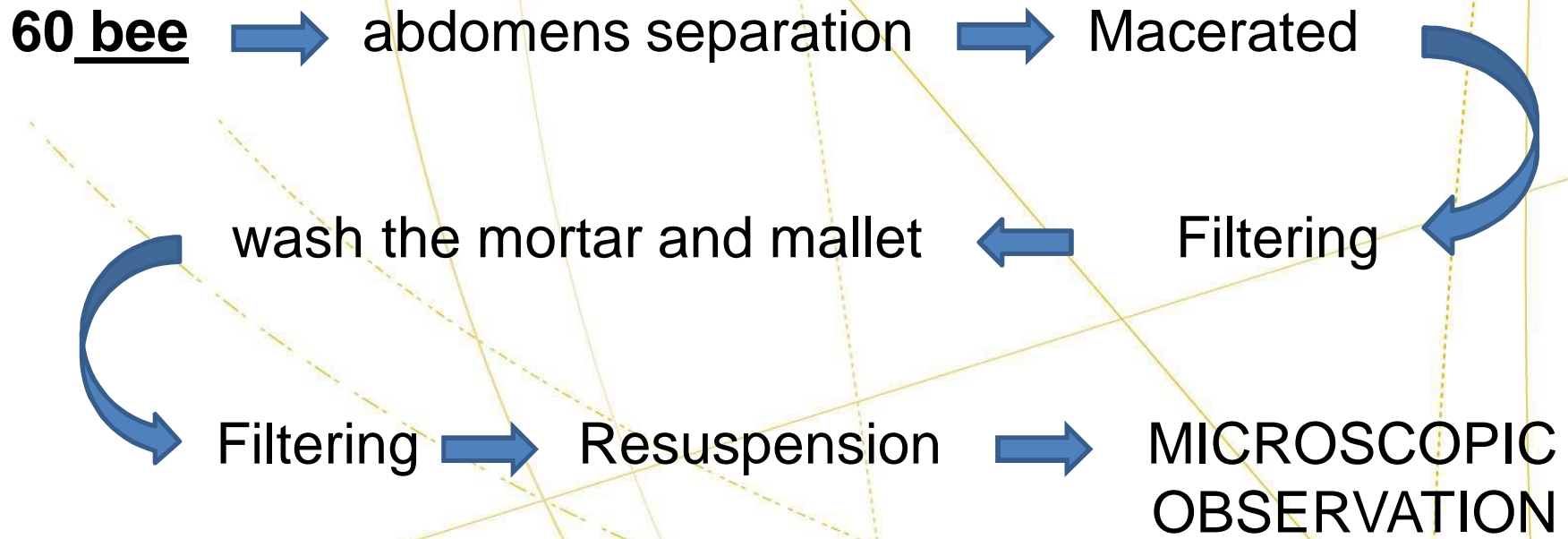
- 100 samples were taken randomly from different colonies in apiaries of the Argentine Northwest Territory
- Composed of 70 or more bees flight returnees, preserved in ethanol
- 60 bees tested with the modified Cantwell method
- 10 bees tested with the method of the OIE
- The results were compared and statistically analyzed using the Wilcoxon test for paired data



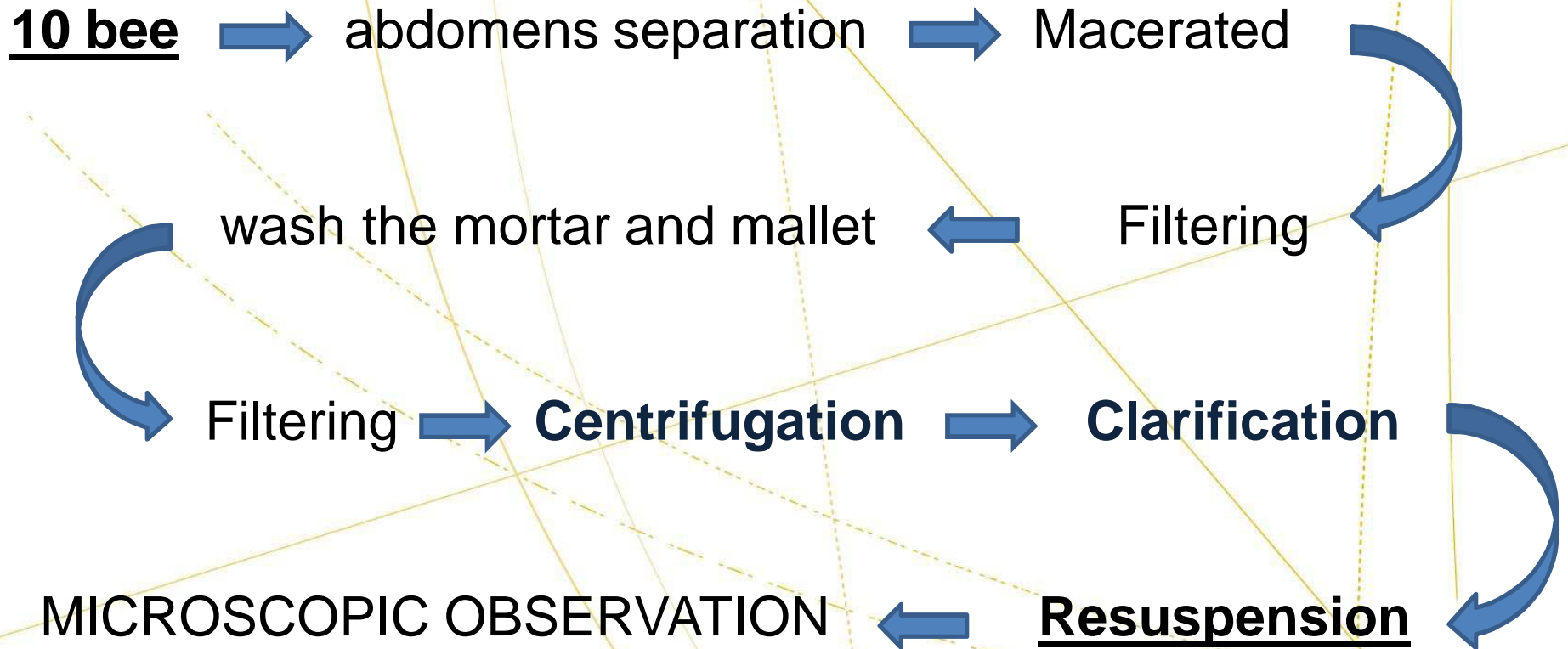
Sampled provinces



Modified Cantwell Method:



Method of the OIE:



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Wilcoxon test (paired samples)

P-value estimated for normal approximation

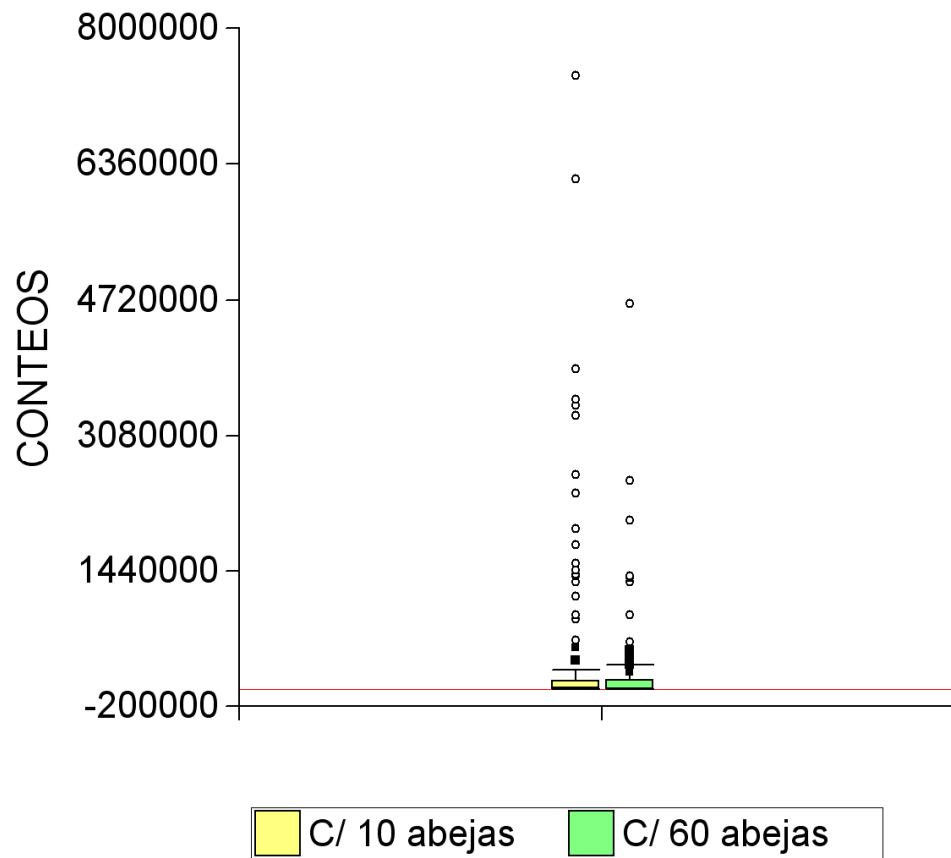
Obs(1)	Obs(2)	N	Suma(R+)	E(R+)	Var(R+)	media (dif)	DE(dif)	Z	p
w/60 bees	w/10 bees	100	428,00	473,00	6771,88	17,21	189,97	-0,55	0,5845

Conclusions

- Pairs were compared with counts obtained with 60 bees procedures and 10 bees and the test does not yield sufficient evidence to reject the null hypothesis with a probability difference $p = 0.5845$
- There is enough statistical evidence to say that the methods are different. The differences found between the counts of both methods are random (due to chance) so that with the evidence found is concluded that the methods work well
- The method of 10 bees showed a greater recovery of spores due to the centrifugation

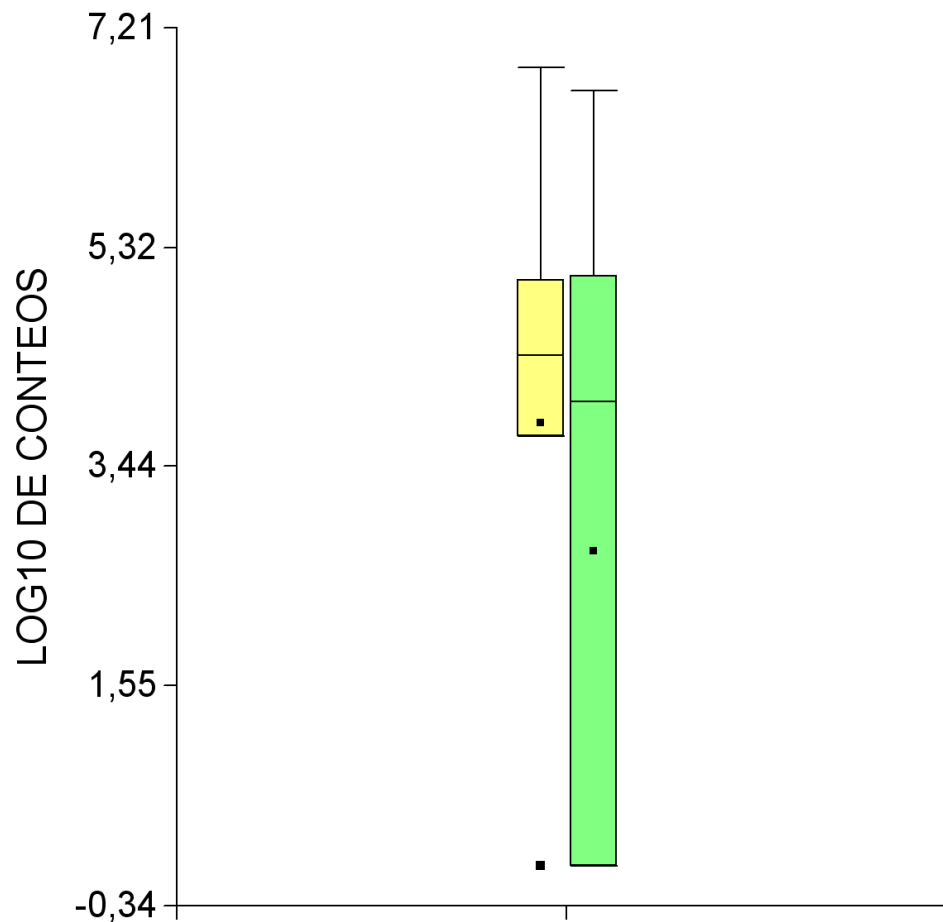
Box plot of counts

DIAGRAMA DE CAJAS DE LOS CONTEOS



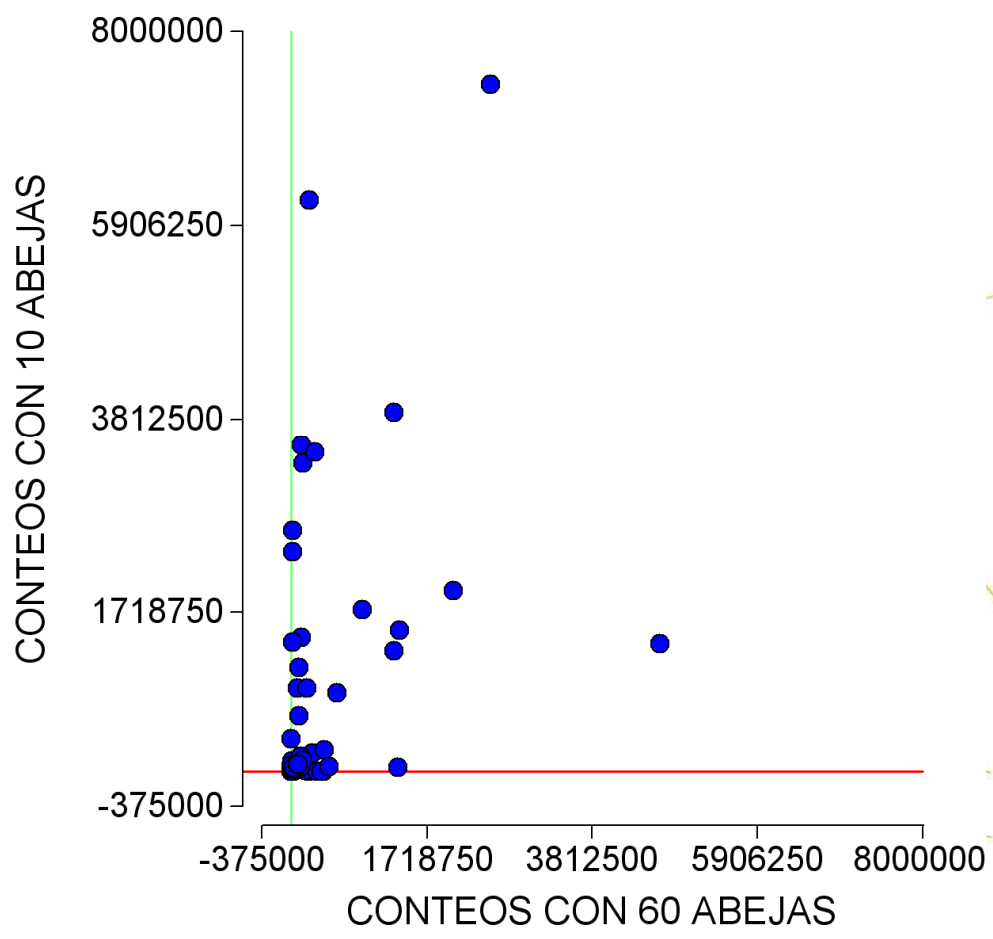
Box plot of the logarithm base 10 of the counts

DIAGRAMA DE CAJA DEL LOG10 DE LOS CONTEOS



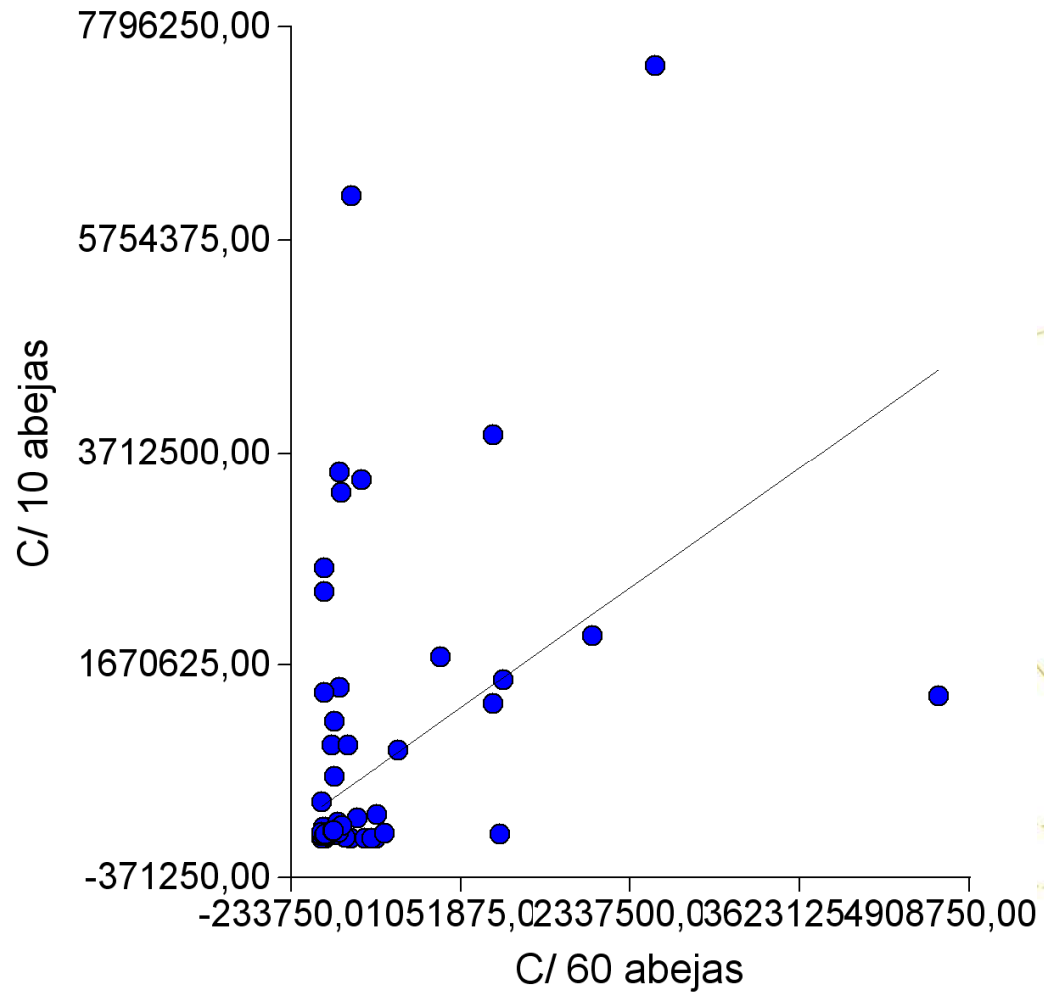
Scatter plot of counts

DIAGRAMA DE DISPERSION DE LOS CONTEOS



Regression line with count values

Línea de regresión con valores de conteo



Regression coefficients and statistics associated

Coef	Est.	E.E.	LI(95%)	LS(95%)	T	p-valor
a	314363,94	117871,31	80452,22	548275,66	2,67	0,0090
b	0,90	0,18	0,54	1,26	4,94	<0,0001

The significance of the model is $p < 0.0001$

The important thing is that the coefficients of model:

OIE count = $a + b * \text{CAT count}$ where, if both methods are equal, to should be 0

b should be equal to 1

The value of a found (other than 0) is explained as the OIE method sensitivity caused by the spinning process of the samples.

The value of b is 0.90 found as point estimates with a confidence interval of 95% between 0.54 and 1.26 which contains the true value of $b = 1$

It is accepted hypothesis that the true value of B is 1

Thank you for your attention!