Empirically Determined Decision Levels
Development and Use In An *In Vitro* Bioassay Program

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This paper discusses empirically determined decision levels in an *in vitro* bioassay program. Specifically, the paper will provide an overview of ANSI N13.30 and ANSI N42.2 decision level concepts, the process used to determine empirical *in vitro* decision levels, and the use of the decision level in reporting personnel results.

BACKGROUND

Lloyd Currie's work[1] on radioassay results reporting provides the statistical basis for ANSI N13.30 and ANSI N42.2. ANSI N13.30 **Performance Criteria for Radiobioassay** has been published for nearly ten years, having been revised numerous times. It still exists as a draft standard. ANSI N42.2 **Measurement Quality Assurance for Radioassay Laboratories** has also recently (2/9/94) been issued as a final revision. Both of these documents provide important guidance to internal dosimetrists, and others. It is just recently, nearly thirty years after publication, that Currie's concepts are becoming widely used by analysts.

Key to Currie's work is the decision level concept. The decision level, as defined in ANSI N42.2, is that quantity of analyte at or above which an *a priori* decision is made that a positive quantity of the analyte is present. An *a priori* decision is one made prior to the measurement (as compared to *a posteriori*, or after the measurement). For both ANSI N13.30 and N42.2, the probability of a Type I (false positive) error is 5%.

ANSI N13.30 discusses appropriate blanks for a sample, person, or phantom. An appropriate blank is, ideally, identical in physicochemically and radiologically significant ways to the sample or person of interest. But what is an appropriate blank for urine samples? Urine samples are subject to natural variability. Variability can arise from diet, water supply, sampling differences, etc. How does an internal dosimetrist take all this into account when running an *in vitro* program? We propose the use of an empirically determined decision level.

RADIOLOGICAL WORKERS, NON-RADIOLOGICAL WORKERS, and EMPIRICAL DECISION LEVELS

The purpose of an *in vitro* bioassay program is to provide an assessment of whether radiological workers have been exposed to operational internal radioactivity, and if so, what the resulting dose is. The keyword is operational. The internal dosimetrist is not interested in non-operational radioactivity. With the analytical system sensitivities available today, we are able to measure very low levels of radioactivity, including levels that exist in samples of interest as naturally occurring

1. **Analytical Chemistry**, Volume 40, Number 3, March 1968

radioactivity, and which are indistinguishable from operational radioactivity.

The expectation we have is that, overall, results of routine urine samples for radiological workers are no different than those of non-radiological workers. This is because the radiological engineering and work controls used for the performance of radiological work preclude the potential of internal exposure. Stating this statistically, we do not expect the mean of baseline results for people who have never handled the nuclide of interest to be any different from the mean of routine samples for radiological workers. This can be tested by testing for a difference between the means of the distributions using the following equation:

$$T = \frac{(\overline{X}_1 - \overline{X}_2) - (\mu_1 - \mu_2)}{\sqrt{\frac{S_1^2}{n_1} + \frac{S_2^2}{n_2}}}$$

where:

T is the calculated test statistic,
$\overline{X}_1$ is the mean of data available for distribution 1,
$\overline{X}_2$ is the mean of data available for distribution 2,
μ_1 is the true mean of distribution 1,
μ_2 is the true mean of distribution 2,
S_1^2 is the variance of distribution 1,
S_2^2 is the variance of distribution 2,
n_1 is the number of results in distribution 1,and
n_2 is the number of results in distribution 2.

The null hypothesis is that $\mu_1 - \mu_2 = 0$; that is, there is no difference in the two means. To determine the test statistic (t) determine $t_{\upsilon,1-\alpha/2,}$ (using a Student t Distribution table) where $\alpha = .05$ and υ is determined by:

$$\upsilon = \frac{(a_1 + a_2)^2}{\frac{a_1^2}{(n_1-1)} + \frac{a_2^2}{(n_2-1)}}$$

where, $a_1 = S_1^2/n_1$ and $a_2 = S_2^2/n_2$. If $T < t_{\upsilon,1-\alpha/2}$, there is no statistically significant difference in the means of the two distributions, and our null hypothesis is upheld. Alternatively, if $T > t_{\upsilon,1-\alpha/2}$, there is a statistically significant difference in the means of the two distributions.

The expected equality of these two sample distributions is the basis for the development of what we have termed the empirical decision level.

Section A.7.3 of ANSI N42.2, **Interpretation of Individual Measurement Results**, states:

> "For the purpose of having a laboratory interpret whether an individual sample measurement is different from its representative appropriate blank, it is recommended that the laboratory compare the net count or count rate of the

measurement with a decision level calculated using the sample specific "appropriate blank". The "appropriate blank" should include measurement interferences from impurities that are not typically known *a priorily* or included in the *a priori* decision limit. This "true" decision level is different from the nominal *a priori* decision level in that it truly represents the appropriate blank at the time of measurement. For some measurement processes, the determination of the "true" appropriate blank for each sample may be impractical."

We consider that for **radiological worker** *in vitro* bioassay, the distribution of **non-radiological** worker results for the corresponding analytical process can be treated as the "appropriate blank". The decision level for radiological worker *in vitro* bioassay can be estimated by analyzing a population of non-radiological workers' *in vitro* samples. Non-radiological worker samples contain interferences from impurities and various levels of naturally occurring radioactivity. These same interferences are present in the radiological worker population.

As stated earlier, Currie considers the decision level as that quantity of analyte at or above which an *a priori* decision is made that a positive quantity of the analyte is present. If we are willing to accept a 5% chance of a Type I (false positive) error, then the "true" decision level can be estimated by the 95th percentile of the distribution of results for non-radiological workers. By setting the empirical decision level at the 95th percentile of the distribution of results for non-radiological workers, we accept a false positive rate of approximately 5%. It is the 95th percentile of the distribution of results for non-radiological workers that is compared to individual results for radiological workers. Results below the decision level indicate that the subject is indistinguishable from the unexposed population from a bioassay standpoint, and followup is therefore not warranted.

As recommended in ANSI N13.30, Appendix A, equation A.9, the decision level can be calculated by:

$$L_c = 2.33\, s_{b,}$$

where s_b = standard deviation of the blank counts.

In addition, the decision level can be estimated from the count result of the 95th percentile result from the counts of the unexposed population. Comparing the calculated decision level and the 95th percentile result serves as a good crosscheck of the population selected (i.e., it is large enough, follows a normal distribution, and does not have significant anomalies).

A critical point to make regarding empirical decision levels is that an empirical decision level value is specific to a nuclide, and dependent on the assay procedure and the performing laboratory. Any change to the analytical process or change in vendor laboratory would be sufficient reason to reestablish the decision level.

METHODOLOGY FOR DETERMINING EMPIRICAL DECISION LEVEL(EL_c)

The methodology for determining an empirical decision level for *in vitro* samples is:

1.) Identify personnel at your facility who have not been exposed to the nuclide of interest.

2.) Collect a statistically meaningful number of samples (40-50) from the personnel identified above. Samples should be collected over a period of time and submitted periodically to the analytical laboratory. This will tend to maximize the distribution of results, and reflect the reality of how you collect and submit operational samples. All of this variability should be included in the determination of the "reasonable blank".

3.) Handle and analyze the samples using the same process that your operational samples will be subject to.

4.) Results of the decision level samples should be reported as the result $\pm 2\sigma$ error.

5.) Once the population of non-radiological worker data has been collected, the data should be ranked in order of results. Statistical outliers, if any, should be discarded using standard statistical methodologies.

6.) The empirical decision level is determined by calculating the standard deviation of the non-radiological worker population of results, and multiplying this by 2.33.

DATA - URANIUM-234

Forty samples were collected over a period of about six months, and submitted to a vendor laboratory for uranium isotopic analysis. Table 1 provides the data returned by the vendor, ranked in ascending order. Table 2 provides summary data, calculational results for standard deviation, and the empirical decision level (EL_c).

Although results for U-233/234, U-235, and U-238 were requested for an isotopic uranium analysis, it is necessary only to establish a decision level for U-234. The percent activity for natural uranium (~2.55% enrichment) is 49.6% from U-234, and 48.2% from U-238, essentially equal amounts. For work involving higher enrichments of uranium, U-234 quickly becomes the only significant activity source because of the difference in specific activity (6240 pCi/μg for U-234 and only 0.336 pCi/μg for U-238).

Table 1: U-234 Data

pCi/L	2σ Error
-.0230	.017
-.0110	.015
-.0110	.021
-.0100	.010
-.0077	.005
-.0075	.015
-.0072	.010
-.0058	.011
-.0058	.011
-.0055	.011
-.0054	.010
-.0054	.011
-.0047	.009
-.0046	.009
-.0033	.019
-.0029	.011
-.0028	.011
.0000	.022
.0000	.016
.0000	.011
.0025	.015
.0025	.015
.0027	.010
.0029	.011
.0029	.017
.0030	.012
.0031	.018
.0055	.011
.0078	.020
.0080	.016
.0082	.016
.0088	.017
.0089	.014
.0090	.023
.0093	.014
.0095	.014

Table 1: U-234 Data

pCi/L	2σ Error
.0095	.023
.0095	.014
.0097	.014
.0097	.019
.0110	.016
.0110	.016
.0120	.023
.0130	.026
.0131	.010
.0150	.015
.0150	.018
.0160	.019
.0170	.017
.0180	.016
.0180	.017
.0190	.016
.0190	.027
.0190	.019
.0190	.022
.0200	.015
.0220	.020
.0240	.021
.0280	.024
.0300	.020
.0300	.021
.0340	.026

Table 2: U-234 Summary

Entity	Value (*pCi/L)
U-234 Ave.	.0063*
S_b	.0113
U-234 Min	-.0230*
U-234 Max	.0340*
$EL_c = 0.03^*$	

DISCUSSION

The empirical decision level determined by calculation was 2.33 * 0.0113, or ~0.03 pCi/L. This should represent the 95th percentile of results; that is about the fifth highest result in a sample of 100, or about the third in a sample of 62. As can be seen from the sample of blanks, 3 of 62 were greater than or equal to EL_c. Results are consistent with the expectation that the calculated decision level should yield 5% false positives. The data indicates that by studying the standard deviation of the blank, as recommended by ANSI N13.30, and applying the theoretical calculation for L_c, the results meet the expectation. About 5 out of 100 samples would be determined to be positive.

IMPLEMENTATION OF DECISION LEVEL REPORTING

ANSI N.42.2 provides recommendations on the interpretation of radioassay results. Specifically, as mentioned in section A.7.3, the service laboratory should compare the sample count or count rate to the decision level count or count rate using an appropriate blank. The empirical decision level, determined as above, becomes the screening level for reporting results as "positive" or negative. Again, it is emphasized that a ~5% false positive rate is built into the process.

Once the empirical decision level for a specific analytical process and laboratory has been established, contractual reporting requirements should be made with the service laboratory. Section A.8 of ANSI N42.2 provides recommendations regarding results reporting by service laboratories. Our reporting is consistent with ANSI N42.2; however, we have specific recommendations regarding when and how this information should be reported.

Transmittal of Information for Radiation Health Records

This information should be delivered 1 sample per report page for proper filing. The reports should be printed on service laboratory letterhead. Our recommendation for the formal report to be stored in the radiation health record includes the following information recommended in ANSI N42.2:

- sample identification code
- reference date/time (specifically the sample date/time)
- identification of the specific measurement procedure (and we would add key instrumentation information like make, model, serial numbers, etc.)
- identification of radionuclides specified for analysis
- the result reported as:
    - < Decision Level (Value & Units), if less than decision level, or
    - Result $\pm 2\sigma$ error, if greater than decision level.

We do not recommend the actual analytical result being stored in the radiation health record unless the result exceeds the empirical decision level. Storing data less than empirical decision level only serves to confuse the record system over time. Restating what we said earlier in this paper, results below the decision level indicate that the subject is indistinguishable from the unexposed population from a bioassay standpoint, and followup is therefore not warranted.

Follow-up of Operational Personnel Positive Results

As mentioned earlier, by design, approximately 5% of the occupational personnel samples submitted for analysis are expected to yield a result above the decision level. Upon notification of an operational personnel positive result we ensure the result reported has been recounted. The second half of the sample is analyzed by the vendor on an expedited basis (result due within 3 business days), and two additional samples are obtained from the involved individual. If the analysis result of the second half of the initial sample is less than decision level (the expectation) then the first of the two resamples is submitted to the vendor for analysis on an expedited basis. If the resample is less than decision level, followup is complete, and no intake is considered to have occurred.

In the event the reanalysis is also positive, both resamples would be submitted for expedited analysis. In the unlikely event that one or both of the resamples were positive, additional sampling would continue until two consecutive negative samples were obtained. A dose investigation would be started.

Continued Study of Non-radiological Worker and Radiological Worker Populations

A follow-up rate significantly above 5% would require an investigation into the cause, including the possibility of a radiological situation requiring remediation or anomalies in statistics obtained by counting the unexposed population. Follow-up rates much less than 5% may indicate that the empirical decision level requires adjustment downward. Either could be caused simply by a change in the analytical process that needs to be reviewed with the vendor and corrected. The statistical test offered at the beginning of the paper can be helpful in analyzing the data to assess the equivalency of the exposed and unexposed populations. In order to do this analysis, it is necessary to have the analytical laboratory report the actual analytical result and its 2 σ error. We recommend a separate report which is provided at the same time as the results reports for radiation health records. This raw data is maintained in a file separate from the radiation health records, should its recovery be required.

Summary

Empirical decision levels provide a simple but powerful method of screening radiological worker *in vitro* sample results. The methodology is easily transferrable to *in vivo* and other analyses.