Call for Methods for Saw Palmetto, SAMe, Vitamin E, CoEnzyme Q_{10}, P*anax quinquefolius*, *Panax ginseng*, Eleuthero, Kava Kava, Yohimbe and Cranberry

AOAC INTERNATIONAL, under contract with the FDA and NIH, has embarked on an effort to develop a group of fully validated methods, validated through the AOAC's Official Methods℠ Program, for selected dietary supplements.

AOAC is now looking for analytical methods for the following ingredients in dietary supplements, applicable to the raw botanical as well as the processed product: Saw Palmetto, SAMe, Vitamin E, Coenzyme Q_{10}, Panax quinquefolius Panax ginseng, Eleuthero, Kava Kava, Yohimbe and Cranberry. If you are aware of or are using a method which is applicable to any of these dietary supplement ingredients, please contact Al Pohland or Mai Nguyen.

Please send the reference, the paper itself, or electronic copy of the method, as well as any available validation data and please indicate whether reference standards are available and their source. Expert Review Panels and AOAC staff will use "Single Laboratory Validation Acceptance Criteria" as a guide to the suitability of the method for further full collaborative study. If the method has undergone no validation, please indicate your willingness to undertake single lab validation studies; a guide for undertaking such studies will be provided on request.

The method and validation data will be evaluated by an Expert Review Panel composed of experts in the analysis of botanicals and with experience in the validation of methods. This Panel will select the methods that will be validated in subsequent full collaborative studies.

AOAC is embarked upon a project of great significance for the future; we hope you can assist us in this effort.

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Single Laboratory Validation Acceptance Criteria (Chemistry methods)

Purpose:

To provide guidance to method submitters on the criteria to be used for accepting the method as an SLV method.

To provide uniformity in the review of methods submitted to AOAC

To assist those laboratories engaged in single lab validation in understanding the types of information the AOAC requires.

Acceptance Criteria:

Is the method clearly and understandably written? (Where alternatives are permitted, are the circumstances for each option clearly and explicitly defined?)

Is the scope and applicability clearly stated? Methods submitted to the Dietary Substances Project must be applicable to one or more of the following matrices or commercial products:

a. The parent botanical
b. A negative control or blank similar to the parent botanical or exhaustively extracted botanical
c. Concentrated powders and extracts
d. Tablets/pills and capsules (gel caps - hard and soft)
e. Combination products including foods based

(Describe steps taken to authenticate reference botanicals, negative controls and marketed products, e.g. exhaustive extraction, certificate of analysis by a certified laboratory, use of tracers, etc.)

Has the method been "optimized?" Have efforts been made to decrease run times, increase resolution, improve peak shapes, minimize extraction time and improve extraction efficiency, separations, selectivity, and address stability. Have tests for analyte stability during processing and under typical conditions of storage been addressed where appropriate?

Has provision been made for confirmation of analyte identity? (The method may be sufficiently specific so that confirmation of identity may not be necessary.)

Was ruggedness testing done during the method development phase identifying variables tested. Have these criteria been written into the method? (Note: ruggedness delineates the limits of minor variables; optimization applies to major variables.)

Are the following performance characteristics available?

OVER

a. The recommended analytical range is LOQ to 200% of expected analyte concentration.

b. Does calibration cover the analytical range? At least 5 concentrations should be used for calibration. Are reference and internal standards available and is the source identified? Are reference standards stable as prepared for use? (The stability of the analytical curve should also be established by duplicating it on another day. Note: It has been repeatedly pointed out that a linear calibration curve is not a critical item, but merely a convenience, and R^2 values close to one may be deceptive [cf. *Analyst* **113**, 1469 (1988)]. The calibration curves for immunoassay are negative [0 concentration is a maximum] and exponential.)

c. Accuracy/Recovery: Was recovery determined from spiked blanks or samples with at least 7 independent analyses per concentration level at a minimum of 3 concentration levels covering the analytical range. Independent means at least at different times. If no confirmed (natural) blank is available, the average inherent (naturally containing) level of the analyte should be determined on at least 7 independent replicates. Was accuracy determined using a Standard Reference Material from NIST, if available, or other certified reference material; or if possible, by reference to an internal standard. (It is critical during method development to establish a reference point to which all assays can be referred. When this material is ultimately assayed, all referred assays can be adjusted accordingly).

d. Repeatability Precision (repeatability, standard deviation): Were the number and types of analyses sufficient for obtaining a good estimate of accuracy or bias? Typically for single lab validation one performs **r** replicate analyses of **m** test portions over a period of **d** days for each sample type (matrix) **n**, where **r** is the number of replicates (2,3 …), **m** is the number of test portions in each group, **d** is the number of days, and **n** is the number of different matrices.

r x ***m*** should never be less than 10; ***n*** should be at least 2, preferably more; ***d*** should be at least 2

The calculated HORRAT value should lie between 0.3–1.3. The formula must be given to show that one is calculating the within-lab value.

Recommended recovery and precision limits for single lab validation

Concentration	Repeatability, %	Recovery, %
100	1	98–101
10	1.5	95–102
1	2	92–105
0.1	3	90–108
0.01	4	85–110
10 g/g (10 ppm)	6	80–115
1 g/g (1 ppm)	8	75–120
10 ng/g (10 ppb)	15	70–125

Is method freely available (no restrictions on use and no copyright restrictions)? AOAC will acknowledge the method source.

(For the use of the AOAC staff, the above information should be presented generally following the format found in the "Guidelines for Single Laboratory Validation of Chemical Methods for Dietary Supplements and Botanicals," page 33–34, and outlined below.)

Recommended Format for SLV Study Report

(1) Method identification: title, authors, contact information, published references

(2) Applicability :scope, abstract clearly identifying the analyte(s), matrix(ices), analytical range, safety

(3) Principle: may be included in abstract

(4) Definitions (units if needed)

(5) Reagents/supplies: include reference standards, calibration standards, etc.

(6) Apparatus: include instruments and equipment used

(7) Sampling: types of samples, amounts, sample handling and storage, stability, preparation of test samples

(8) Method: include calibration and procedure and include or refer to (1)

(9) Calculations summary: detection/quantification limits, recovery, precision, bias, and additional information such as stability and measurement uncertainty ($2RSD_r$)

(10) Other pertinent information; e.g. confirmation of analyte identity and ruggedness (may be placed in #8)

(11) Conclusions