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INSTYTUT CHEMII I TECHNIKI JĄDROWEJ

POLISH CERTIFIED REFERENCE MATERIAL

FOR MULTIELEMENT TRACE ANALYSIS

POLISH VIRGINIA TOBACCO LEAVES (INCT-PVTL-6)

GENERAL INFORMATION

INTENDED USE:	CHECKING THE ACCURACY OF ANALYTICAL WORK OF THE LABORATORIES ENGAGED IN THE DETERMINATION OF TRACE ELEMENTS IN BIOLOGICAL SAMPLES. CALIBRATION OF APPARATUS AND METHODS
ELEMENTS FOR WHICH CERTIFIED CONCENTRATIONS WERE ESTABLISHED:	Ag, Al, As, B, Ba, Br, Ca, Cd, Ce, Co, Cu, Er, Eu, Hf, Hg, K, La, Li, Mg, Mn, Mo, Nd, Ni, P, Pb, Rb, S, Sb, Sc, Sm, Sr, Ta, Tb, Th, V, Zn
ELEMENTS FOR WHICH NON-CERTIFIED "INFORMATION" VALUES ARE AVAILABLE:	Bi, Cl, Cr, Cs, Fe, Na, Pr, Sn, Ti, Tl, U, Y, Yb

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ORIGIN, PREPARATION AND TESTING

Polish Virginia Tobacco Leaves material, grown in Poland, was prepared by the Institute of Nuclear Chemistry and Technology (INCT). Dried leaves were procured from the POLSKI TYTOŃ Company (Poland). The grinding and sieving process of the material was performed at the company: Przetwórstwo Rolno-Spożywcze "KANIA" Sp. z o.o. (Częstochowa, Poland). Tobacco leaves were cut and ground using a grinding machine, followed by sieving through sieves 100 μm. The part of the material, which passed through the sieve was collected and stored in a polyethylene (PE) bag, whereas the retained fraction was ground and sieved again. Approximately 48 kg of ground and sieved tobacco leaves powder was finally obtained. Examination by optical microscopy revealed that Martin's diameter of over 90% of particles was below 60 μm. The whole lot of the material was then transferred to a 110 dm^3 PE drum and homogenized by rotating in three directions for 16 hours. After this time the preliminary homogeneity testing was carried out by determining Ca, Fe, K and Sr content by X-ray fluorescence (XRF) in several 5 g subsamples taken from the drum. The results did not reveal any signs of inhomogeneity. The next step was distribution of Polish Virginia Tobacco Leaves (INCT-PVTL-6) into 150 cm^3 polypropylene (PP) bottles with a screw cap in 50 g portions (future CRM) and in 10 g portions into 60 cm^3 polyethylene tetraphtalate (PET) bottles (intercomparison samples). In order to ensure long-term stability of the new CRM, all containers with INCT-PVTL-6 were sterilized by electron beam radiation (energy 13 MeV) from a linear accelerator LAEA-13. The dose amounted to approximately 30 kGy. Final homogeneity checking of the material for the sample size 100 mg was accomplished by means of instrumental neutron activation analysis (INAA) and inductively coupled plasma-mass spectrometry (ICP-MS). Statistical evaluation of results (using Fisher's and t-Student's tests) obtained by INAA for Ba, Co, Cs, Eu, Fe, Hf, Rb and Sc proved good homogeneity. Analogous conclusion followed from the analysis of variance (ANOVA) of the results for Ba, Cd, Co, Eu, La, Li, Mg, Mn, Sr and V determined by ICP-MS. Hence, INCT-PVTL-6 can be considered as homogeneous at least for sample mass ≥100 mg.

Long-term stability of the material was checked by comparing results obtained for one bottle (randomly chosen) stored under controlled conditions: in the air-conditioned room at 20°C (normal storage). Samples of the CRM (ca. 100 mg) were taken from the bottle after 0, 6, 12, 18 and 22 months of storage and concentration of six selected elements (Ce, Co, Fe, Rb, Sc and Zn) was determined by INAA method. The stability of INCT-PVTL-6 will be monitored during the whole storage period. On the basis of previous experiments the standard uncertainty due to short-term stability was found to be negligible.

Determination of moisture content

In order to express the concentrations of elements on a dry-mass basis, moisture content should be determined on a separate subsample (not taken for analysis) by drying at 75°C for 30 hours.

CERTIFICATION

Assigning of certified values was done on the basis of world-wide interlaboratory comparison in which 87 laboratories from 18 countries participated providing 1250 laboratory averages (5581 individual determinations) for 79 elements. Their cooperation is gratefully acknowledged. The participants along with the INCT-PVTL-6, analyzed also the CRM provided by the organizers the identity of which was unknown to them. This enabled to create two databases. The first called "original" contained all results for all elements in Polish Virginia Tobacco Leaves supplied by participating laboratories. The second database called hereafter "alternative" was created from "original" database for these elements for which certified values in the CRM (sent to participants and analyzed by them along with INCT-PVTL-6) were available. The alternative database was created as follows. For every laboratory its results obtained for a given certified element in the CRM were examined whether the confidence limits of the laboratory results at a significance level $\alpha = 0.05$ overlap with the confidence limit of the certified value in the CRM. If not, the results of this laboratory for the considered element were removed from the original population of results for INCT-PVTL-6. Each of the two databases was subjected to statistical evaluation according to the method published previously [1] with some later modifications [2-4]. This approach is based on the outlier's rejection procedure, which uses concurrently four statistical criteria, namely those of Dixon (D), Grubbs (G), Skewness (S) and Kurtosis (K) at $\alpha = 0.05$, followed by calculation of overall means of results (remaining after the outlier rejection), standard deviations, standard errors, confidence intervals etc.

The criteria used to decide whether the overall mean can be given the status of the certified value were as follows:

1. The ratio of the one-sided confidence interval and the overall mean:

$$\frac{SD \cdot t_{0.05}}{\overline{X} \cdot \sqrt{N}} \begin{cases} \leq 20\% \text{ (trace elements)} \\ \leq 10\% \text{ (major elements)} \end{cases}$$

or relative standard deviation:

$$\frac{SD}{\overline{X}} \begin{cases} \leq 25\% \text{ (trace elements)} \\ \leq 15\% \text{ (major elements)} \end{cases}$$

For the purpose of this work, elements with concentration exceeding 5000 mg kg^{-1} (ppm) were considered to be the major elements.

2. The overall mean was calculated on the basis of at least four "accepted" laboratory means ($N \geq 4$) obtained by more than one analytical technique. If results from only one analytical technique are available, the number of "accepted" laboratory averages used for the calculation of the overall mean can not be smaller than five ($N \geq 5$).
3. If the conditions (1) and (2) are fulfilled but the number of outliers exceeds 50%, the additional procedure is activated which repeats the process of outlier rejection from the beginning, checking simultaneously the changes of the mean and standard deviation accompanying successive rejections. The process of rejecting of outliers is then stopped when the successive change in both the mean and standard deviation becomes lower or equal to 15%. The condition (1) is then rechecked.
4. If the above criteria are met but there are indications that after outlier rejection performed on the whole population, the remaining populations of results obtained by various analytical techniques differ significantly, the assignment of certified value is suspended.

"Information" values were assigned to those elements for which the results while not fulfilling simultaneously the conditions (1)-(4) still fulfilled the following condition:

$$\frac{SD \cdot t_{0.05}}{\overline{X} \cdot \sqrt{N}} \begin{cases} \leq 50\% \text{ (trace elements)} \\ \leq 30\% \text{ (major elements)} \end{cases}$$

calculated on the basis of at least three "accepted" laboratory averages, and are quoted as numbers only, i.e. without confidence intervals. The elements for which the data did not fulfill the above criterion were considered to be outside of any classification. Evaluation of data from the original and alternative databases by the method described above gave nearly in all cases similar values of the overall mean and the confidence interval. For some elements (As, Cd and U) these data were additionally confirmed by the results obtained by the radiochemical NAA ratio primary measurement reference procedure (definitive method) [5-8]. The values for Al, As, Ba, Br, Ca, Cd, Ce, Cl, Co, Cr, Cs, Cu, Eu, Hg, K, La, Mg, Mn, Na, Ni, Pb, Rb, S, Sc, Sm, Sr Tb, Th, Tl, V, Yb and Zn were finally determined (certified or information values assigned) using the alternative database. The values for other elements were derived from the original database. The traceability of this CRM to the SI units is ensured by the use of RNAA ratio primary measurement reference procedure when applicable, the use of analytical methods calibrated against pure metals or oxides with full uncertainty budget and QA/QC reference material analyzed together with the candidate reference material.

According to recent trends and recommendations to CRM producers, in addition to analytical uncertainties also the uncertainty due to the long-term stability, moisture determination and inhomogeneity are taken into account. Consequently, the combined standard uncertainty of the certified value u_c consists of four contributions:

$$u_c = \sqrt{u^2_{interlab} + u^2_{lstab} + u^2_{inhom} + u^2_m}$$

where $u_{interlab}$ is estimated as standard deviation of the overall mean, u_{lstab} the standard uncertainty estimated from the long-term stability studies, u_{inhom} the standard uncertainty estimated from homogeneity studies and u_m the standard uncertainty due to moisture determination. The expanded uncertainty (U), corresponding to 95% confidence level, is obtained by multiplying u_c by a coverage factor $k = t_{0.05}$ (t-Student's parameter for $\alpha = 0.05$ and $n - 1$ degrees of freedom, where n is number of laboratory averages).

CERTIFIED VALUES FOR INCT-PVTL-6 ($\bar{X} \pm U$)

Major and Minor Elements

Element	Concentration wt%
Ca	2.297 ± 0.078
K	2.640 ± 0.090
Mg	0.241 ± 0.009
P	0.242 ± 0.015
S	0.378 ± 0.059

Trace Elements

Element	Concentration mg kg^{-1}	Element	Concentration ng g^{-1}
Al	252 ± 49	Ag	19.1 ± 3.8
B	33.4 ± 1.9	As	138 ± 10
Ba	41.6 ± 1.9	Ce	743 ± 51
Br*	19.5 ± 1.0	Co	154 ± 7
Cd	2.23 ± 0.12	Er*	18.5 ± 3.2
Cu	5.12 ± 0.20	Eu	14.0 ± 2.6
Li	3.35 ± 0.67	Hf*	161 ± 8
Mn	136 ± 5	Hg	23.2 ± 1.6
Ni	1.49 ± 0.14	La	540 ± 27
Rb	5.97 ± 0.28	Mo	396 ± 29
Sr	133 ± 6	Nd	322 ± 24
Zn	43.6 ± 1.4	Pb	972 ± 147
		Sb	37.2 ± 3.9
		Sc*	59.5 ± 3.4
		Sm	58.0 ± 4.3
		Ta*	10.9 ± 1.2
		Tb	8.1 ± 1.0
		Th	88.8 ± 6.8
		V	405 ± 56

* certified on the basis of results by single analytical method

INFORMATION VALUES FOR INCT-PVTL-6

Bi	140 ng g^{-1}	Fe	258 mg kg^{-1}	Ti	12.3 mg kg^{-1}
Cl	0.457 wt%	Na	62.4 mg kg^{-1}	Tl	22.8 ng g^{-1}
Cr	911 ng g^{-1}	Pr	82.9 ng g^{-1}	U	22.0 ng g^{-1}
Cs	26.6 ng g^{-1}	Sn	31.1 ng g^{-1}	Y	218 ng g^{-1}
				Yb	28.3 ng g^{-1}

The shelf life of INCT-PVTL-6: 31 December 2020

The preparation and certification of INCT-PVTL-6 certified reference material was performed by the staff of the Laboratory of Nuclear Analytical Methods, Institute of Nuclear Chemistry and Technology, under the direction of Dr. Zbigniew Samczyński. More information on the material and the certification procedure is available in the published report [9].

Warszawa, March 31, 2010

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INSTYTUT CHEMII I TECHNIKI JĄDROWEJ

POLISH CERTIFIED REFERENCE MATERIAL
FOR MULTIELEMENT TRACE ANALYSIS

POLISH VIRGINIA TOBACCO LEAVES (INCT-PVTL-6)

1. **The stability of Polish Virginia Tobacco Leaves (INCT-PVTL-6) has been monitored during storage. On the basis of the obtained results, the shelf life is extended until 31 December 2024.**
2. The material is stored in an air conditioned room, in which the temperature does not exceed 22°C. Its stability is monitored at appropriate time intervals.
3. The users should store the material at room temperature [(20±5)°C] in tightly closed original containers.
4. For chemical analysis and simultaneous determination of water content, samples of the material of adequate size should be taken. Immediately after sampling, it is necessary to cover firmly the container. The unused part of samples must not be placed back in the container.

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