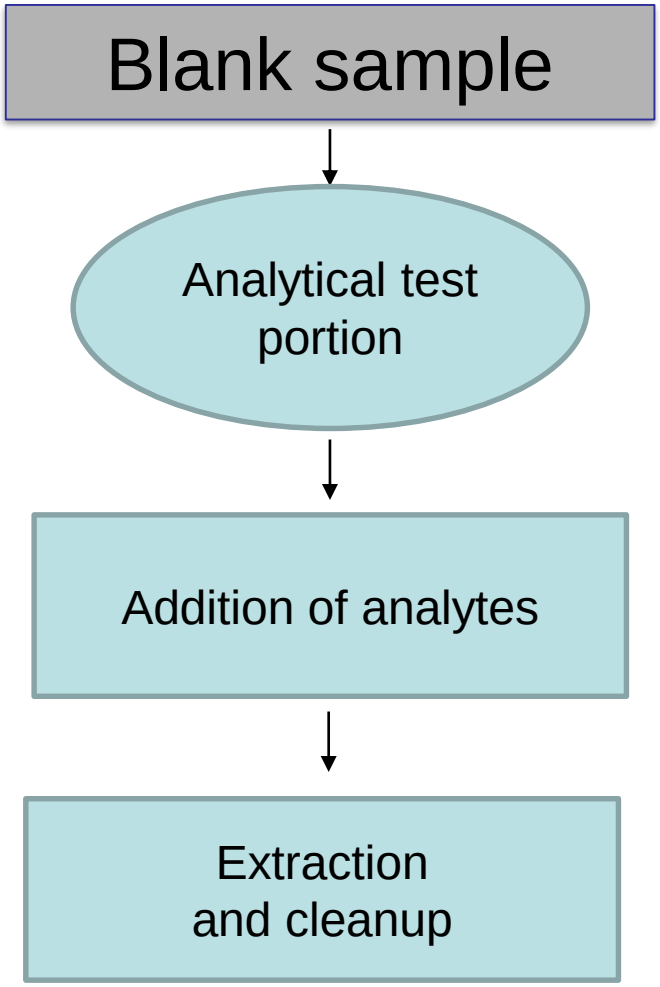
REVISION OF SANTE DOCUMENT

PROCEDURAL VS. MATRIX MATCHED CALIBRATION

Recovery experiment



Influence of calibration?

Calibration types

SANTE document Appendix E Table 1

		Reduces bias due to				
Option	**Procedure**	losses during the extraction	cleanup losses	injection errors	matrix effects	cross reference
1. Matrix-matched calibration	calibration standards prepared in extract of blank sample of the same matrix	no	no	no	yes	C21-C23
2. Procedural calibration	calibration standards prepared in sub-portions of blank sample of the same matrix, analyte added before extraction	yes [1]	yes	no	yes	C28
3. Use of internal standard (IS) (other than the isotopic analogue of the analyte)	a. Internal standard added to the calibration standards, and to each sample before extraction (procedural internal standard)	possibly [1,2]	possibly [2]	possibly [2]	possibly [2]	C32-C34
	b. Internal standard added to the raw extract before cleanup (procedural internal standard)	no	possibly [2]	possibly [2]	possibly [2]	C32-C34
	c. Internal standard added to the calibration standards, and to the final extract of each sample (injection internal standard)	no	no	possibly [3]	possibly [2]	C32-C34
4. Use of isotopically labeled internal standard (ILIS) [4]	a. isotope analogue added to the calibration standards, and to each sample before extraction	yes [1]	yes	yes	yes	C35-C37
	b. Isotope analogue added to the raw extract before cleanup	no	yes	yes	yes	C35-C37
	c. isotope analogue added to the calibration standards, and to the final extract of each sample	no	no	yes	yes	C35-C37
5. Standard addition method	a. *Sample standard addition*: analyte standard added to test-portions of each sample before extraction	yes [1]	yes	no	yes	C24
	b. *Extract standard addition*: analyte standards added to aliquots of the final extract of each sample	no	no	no	yes	C25

Calibration types

- Solvent-based
- Matrix-based
- Matrix-matched
- Procedural
- ILIS
  - to test portion
  - to extract

- Standard addition
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Results lead to apparent recoveries in all cases,
Typically, matrix-matched results is the best estimation of absolute recovery!

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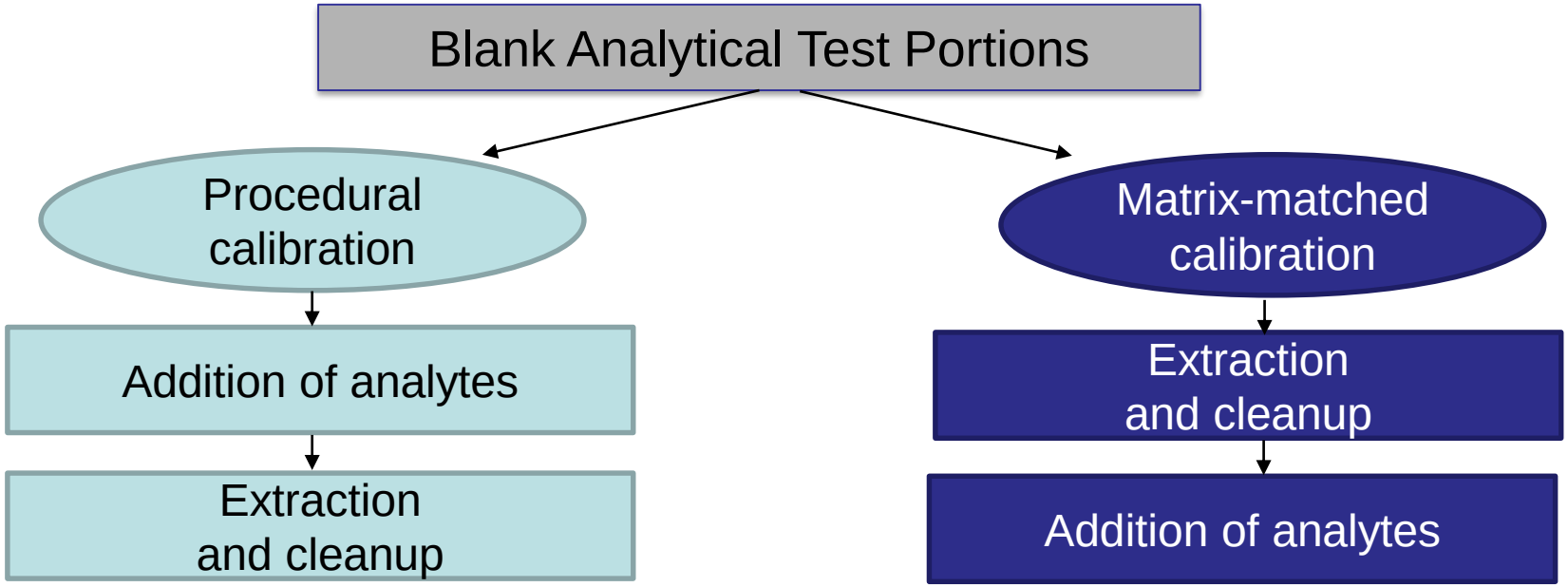

compensates for
- ✓ losses
- ✓ matrix effect

Validation criteria:
- mean recovery: 70-120%
- repeatability ≤ 20%
- Experiments for absolute recovery necessary during initial validation

compensates for
- ✓ matrix effect

Validation criteria:
- mean recovery: 70-120% (in exceptional cases 30-140%)
- repeatability ≤ 20%
- No extra experiments for absolute recovery necessary

Validation results for fish - procedural vs. matrix-matched calibration

Table 1: Procedural calibration

Analyte	n	Apparent Recovery [%]	RSD [%]
Penthiopyrad	6	109	8
Phosalon	6	108	7
Phosmet	6	114	5
Phoxim	6	102	14
Pinoxaden metabolite M4	6	110	13

Table 2: Matrix-matched calibration

Analyte	n	Recovery [%]	RSD [%]
Penthiopyrad	6	107	10
Phosalon	6	105	2
Phosmet	6	83	11
Phoxim	3	35	46
Pinoxaden metabolite M4	6	107	6

Comparison - procedural vs. matrix-matched calibration

Table 1: Matrix matched calbration

Analyte	Mean concentration [µg/kg]	SD [µg/kg]	Recovery rate [%]	RSDr [%]	Spike level [µg/kg]
Aldrin	0.48	0.10	65.0	25.4	0.6
Bifenthrin	0.76	0.13	126.9	16.8	0.6
Bromocyclen	0.43	0.15	71.4	33.9	0.6
Chlordane, cis-	0.56	0.08	93.3	14.9	0.6
Chlordane, trans-	0.58	0.09	95.8	16.2	0.6
Cyfluthrin (I-IV)	0.70	0.21	115.8	30.6	0.6
Cyhalothrin, lambda- (I-II)	0.55	0.12	92.2	20.8	0.6
Cypermethrin (I-IV)	0.66	0.16	109.2	23.9	0.6
DDD, 4,4-	0.71	0.11	118.3	16.0	0.6
DDE, 4,4-	0.58	0.09	95.8	15.6	0.6
DDT, 2,4-	0.61	0.09	102.2	15.0	0.6
DDT, 4,4-	0.68	0.10	113.9	15.3	0.6
Deltamethrin	0.72	0.14	119.4	19.7	0.6
Diallate, cis- (74%)	0.38	0.18	85.2	47.6	0.45
Diallate, trans- (26%)	0.12	0.05	72.9	46.3	1.6
Dieldrin	0.55	0.08	92.2	14.8	0.6
Endosulfan, alpha-	0.58	0.07	96.1	12.1	0.6
Endosulfan, beta-	0.64	0.07	105.8	11.8	0.6
Endosulfan-sulfate	0.70	0.11	116.7	15.8	0.6
Endrin	0.60	0.07	99.2	11.6	0.6
Fenvalerate, R- (75%)	0.68	0.11	151.9	15.6	0.45
Fenvalerate, S- (25%)	-	-	-	-	-

Table 2: Procedural calibration

Analyte (percentage composition of isomers)	Mean concentration [µg/kg]	SD [µg/kg]	Recovery rate [%]	RSDr [%]
Aldrin	0.48	0.09	96.7	18.6
Bifenthrin	1.15	0.05	113.3	4.3
Bromocyclen	0.48	0.09	96.7	18.6
Chlordane, cis-	0.57	0.09	113.3	16.6
Chlordane, trans-	0.57	0.09	113.3	16.6
Cyfluthrin (I-IV)	5.95	1.20	119.0	20.1
Cyhalothrin, lambda- (I-II)	10.62	1.08	106.2	10.2
Cypermethrin (I-IV)	5.98	0.99	119.6	16.5
DDD, 4,4-	1.17	0.07	116.7	6.4
DDE, 4,4-	0.52	0.09	103.3	17.4
DDT, 2,4-	0.53	0.09	106.7	17.7
DDT, 4,4-	0.55	0.10	110.0	17.4
Deltamethrin	1.18	0.07	118.0	6.3
Diallate, cis- (74%)	1.15	0.05	115.0	4.3
Diallate, trans- (26%)	0.28	0.04	109.0	13.2
Dieldrin	0.55	0.10	110.0	17.4
Endosulfan, alpha-	0.55	0.08	110.0	13.9
Endosulfan, beta-	0.50	0.06	100.0	11.5
Endosulfan-sulfate	0.58	0.09	116.7	15.4
Endrin	0.55	0.10	110.0	17.4
Fenvalerate, R- (75%)	7.88	0.72	105.1	9.2
Fenvalerate, S- (25%)	2.60	0.26	104.0	9.9