

18-Jan-21
ASB 113, Standard for Identification Criteria in Forensic Toxicology

#	Section	Type of Comment (E-Editorial, T-Technical)	Comments	Proposed Resolution	Final Resolution
32	All	E / T	The reputation of the ASB (Toxicology) is hinged upon development of scientifically valid documents, and could be significantly impacted by the publishing of those on the contrary. As written, this document contains inaccuracies, is vague and at times internally contradictory, and is developed without consideration for current/future scientific practices, developments, and advancements. It is my belief that, as written, this current standards document is inaccurate, unreliable, and inappropriate for the forensic toxicology community.	Major revisions are required before final approval. This standard document should be overhauled with a careful eye. Re-evaluation of the science behind the statements is required.	REJECT: No specific Proposed Resolution was provided. If other comments are provided below, they will be individually evaluated.
46	All	T	A section should be added that discusses how a lab should handle a situation where their data does not meet and aspect of these criteria (i.e. what happens if you don't meet the criteria?). For example, a drug is detected by LC-HRMS with ppm error at 5.1. Is the analysis awarded no points? Is the analysis awarded only one point for chromatography? Can the analysis be downgraded from High Res to Low Res so salvage points? A second example: There is a mass at 75% height at M-5 from my drug, causing no issues or interference. Since it is present at >50% (as currently written in this standard), is the analysis awarded no points? Is the analysis awarded only one point for chromatography? Can the analysis be downgraded from "full scan" to "HR MS 2.5 pt per ion"? There are many other examples as well.	Add section that addressed these concerns (and others) and confusion.	REJECT: Section 4.2.1 (Validation) of this document indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance. The specific mass spectrometry criteria referenced in the comments are from ASB 098 and not this document.
47	All	T	There is a general lack of correlation between this document and proposed Std 089. Std 098 does not mention "High Resolution Full Scan", however, that is an option here. Here is an example. I analyze a sample using LC-QTOF-MS. I generate chromatographic data, a full scan precursor ion mass spectrum, and an MS/MS product ion mass spectrum. I could get 1 pt for chrom, 2.5 pts for HRMS ion or 3.5 pts for HR full scan, and 4 pts for HR product ion spectrum (not to mention more pts if I use some MRM ⁺ HR type data). The point is, the MS criteria and points for identification are not well aligned at all. And the wording within this document allows for scenarios that the authors are not intending. As written, one could meet identification criteria based on a TOF-MS scan of a precursor ion ALONE (or its non-specific fragment(s)) - I don't think many people would consider that true identification. The system appears to be inherently flawed.	Revise with sections, headers, or more specific language that relates to Std 098.	REJECT: Section 4.4 of ASB 098 does allow for full-scan analysis using a high resolution mass spectrometer. The headers cannot readily align between the two draft standards because ASB 113 differentiates between high resolution mass spectrometry in full scan mode and selective ion monitoring for point assignment, whereas ASB 098 combines both approaches into a single heading to simplify the requirements. The points in the cited example, as interpreted, are correct but two additionally important concepts must be remembered: 1) Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed. "; 2) Section 4.2.1 indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance.
69	All	E / T	My overall concern is how this standard will be implemented and enforced in toxicology laboratories today. This criteria requires laboratories to really assess points at the very least, per reported drug, but really per result, as inevitably issues will be encountered in the absence of perfect chromatography and/or identification.	The standard would benefit from fresh perspectives from current reporting laboratories who could provide insight into how these standards would be implemented and enforced.	REJECT: Section 4.2.1 (Validation) of this document indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance. No specific Proposed Resolution was provided. If other comments are provided below, they will be individually evaluated.
54	General	E	Our group found this document confusing and difficult to follow. We tried to assign points to a few of our own drug confirmations to test section 4.3.2, but we struggled to understand it. Examples provided below.	Of greatest concern is the awarding of sufficient points for identification based on a single test in some cases, which implies identification can be made from a single aliquot. This poses a significant risk for misidentification.	REJECT: Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed." If other comments are provided below, they will be individually evaluated.
14	1	T	Routine volatiles, including acetaldehyde and acetone, are not alcohols. Yet they are commonly analyzed in HS-GC/FID panels with ethanol, methanol, and isopropanol. The statement "This document does not address identification of alcohols..." should be extended to these compounds.	Revise to "This document does not address identification of alcohols" to "volatiles" or "routine volatiles"	ACCEPT: The scope was clarified to add "routine volatiles" to the list of excluded compounds, because compounds such as acetaldehyde and acetone are not classified as alcohols.
66	1	T	does not address identification of alcohols, carbon monoxide, ... This change would now require other low mol weight volatiles to meet this standard. What about things like DFE? Was this purposeful? If using HS-GCMS to identify DFE with scan analysis and library matching you would not have enough points. Requiring more tests above that would be excessive.	Remove changes to this section. Leaving a little vagueness in a document that in general is overly restrictive with not enough exceptions to be practically useful or achievable by most laboratories is preferable.	REJECT: The text was not changed as recommended; however, "routine volatiles" was added to the list of compounds excluded from the scope of this document.

33	3.2	T	This definition is (maybe purposefully?) vague and confusing. Why is there a requirement for a standard to be run concurrently? How does this negatively impact the generation of accurate data? This is a validation consideration and does not belong in a mass spec criteria document.	Remove the definition and reference to it (and see below)	REJECT: We believe this was intended to reference 3.3 in this document (not 3.2 as in 098 standard). The inclusion of a concurrently analyzed reference standard is important to establish the retention time for chromatographic analysis (see Section 4.2.2).
70	3.2	T	"Concurrently analyzed" listed at same time (or close to) under same analytical conditions, but some mass spec based screening techniques will not have concurrently analyzed standards.	Need to be more specific with "concurrently analyzed", and should this apply to screening and/or confirmation?	REJECT: We believe this was intended to reference 3.3 in this document. The definition of "concurrently analyzed" was developed to address the needs of this document (ASB 113) and the Mass Spec Criteria document (ASB 098). This document specifically requires the use of a concurrently analyzed standard to determine the retention time in chromatographic analyses. To receive points assigned to chromatography (whether in a "screen" or "confirmatory" analysis) requires the concurrently analyzed standard.
10	3.3	E/T	This definition is - perhaps purposefully - vague. I could read this as "a positive control needs to be analyzed in the same batch" or "in the same week/month/year" (as long as the analytical conditions are the same. Also, there are two periods.	Clarify and remove the second period.	ACCEPT WITH MODIFICATION: The extra period was removed. The inclusion of a concurrently analyzed reference standard is important to establish the retention time for chromatographic analysis (see Section 4.2.2). Provided that the same analytical conditions are in place, this document defines that it must be "analyzed at or close to the same time" for it to be considered as "concurrently analyzed".
34	3.3	T	The term "diagnostic" should not be used due to its definition in clinical practice and the impact this could have downstream.	Use a word other than "diagnostic". Also, revise how the term is used throughout.	REJECT: We believe this was intended to reference 3.4 in this document. "Diagnostic ion" is commonly accepted terminology in the field of mass spectrometry and is synchronized with ASB 098 on Mass Spectrometry Criteria. The term is also approved by IUPAC in "Definition of Terms Relating to Mass Spectrometry". An alternative terms was not recommended by the commenter.
35	3.3	T	A precursor ion should not be classified as a "diagnostic ion"... this does not make sense within the context of how the word/definition is being used. This is very confusing to a reader. Also, the definition is vague which undermines the purpose of calling the ions "diagnostic". As written, nearly all ions are diagnostic ions which diminishes the purpose of categorizing these as such. The term diagnostic ion should be used only for fragment ions which have high structural significance (again, based on interpretation of the document as written). The current definition and the manner in which it is used in this document would allow the m/z 91 fragment ion of amphetamine to be used for identification purposes (and maybe even 4 points could be gained), when in reality that detection is very non-specific and should be given a low score/points based on what can be determined/concluded from that data alone.	Revise to "A MS/MS or fragment ion that has high structural relevance to the targeted analyte." (Remove MS and molecular ion). This also changes how the term is used throughout so consideration should be given.	REJECT: We believe this was intended to reference 3.4 in this document. The recommended change was not made; however, the definition was modified to read: "A MS or MS/MS ion or fragment ion whose presence and abundance are characteristic of the targeted analyte." Further, WADA Technical Document - TD2015IDCR "MINIMUM CRITERIA FOR CHROMATOGRAPHIC-MASS SPECTROMETRIC CONFIRMATION OF THE IDENTITY OF ANALYTES FOR DOPING CONTROL PURPOSES" also defines diagnostic ion similarly.
59	3.3	T	This definition is more of a validation consideration than a mass spec criteria. The absence of a concurrently analyzed standards does not affect the quality of the mass spectral data obtained.	Consider removing this definition and reference in the document.	REJECT: In this document, the inclusion of a concurrently analyzed reference standard is important to establish the retention time for chromatographic analysis (see Section 4.2.2).
71	3.3	T	Disagree with the use of "diagnostic", although it is used in clinical practice, a more scientific term would be appropriate.	Replace "diagnostic" with a more scientific term.	REJECT: We believe this was intended to reference 3.4 in this document. "Diagnostic ion" is commonly accepted terminology in the field of mass spectrometry and is synchronized with ASB 098 on Mass Spectrometry Criteria. The term is also approved by IUPAC in "Definition of Terms Relating to Mass Spectrometry".
5	3.6	T	Language here doesn't fit 3.1, since interferences are not destined "to be identified or measured".	Change to "Non-targeted substances (i.e., matrix components, (...)" to reflect language in 3.19 and be coherent with 3.1.	ACCEPT: The definition was changed to substitute "non-targeted substances" for "non-targeted analytes".
6	3.8	E	Physicochemical here does not match "physico-chemical" of 3.10.	Use "physico-chemical" for both instances.	ACCEPT WITH MODIFICATION: The hyphenated version was removed. "Physicochemical" was used in both definitions.
7	3.9	E	The first reference of the document is assigned number 3. References should be ordered.	Order references by appearance in the main text, and/or add a reference number to the ASB documents in Section 2.	REJECT: The format and order of references follows the requirements of ASB.
8	3.9	E	Why are reference numbers in ASB 098 superscripted, but in standard text in ASB 113?	Use either superscripted or standard reference numbers for both documents.	REJECT: Per ASB, both formats are acceptable. Different authors drafted the two documents, thus the difference.
74	3.12	T	Why does the comparison with the standard need to be previously injected? What happens if the standard(s) you use to compare are at the end of the batch/run?	Remove "previously"	ACCEPT: The definition was modified as suggested.
75	3.13	T	This seems to indicate that blood, plasma and serum are considered different matrices.	Examples include blood (i.e., whole, plasma, serum), urine, vitreous fluid, hair, and tissue.	REJECT: Blood, plasma, and serum are different matrices.
9	3.15	T	To me, this definition is confusing. I alternate between thinking this is the number of points necessary for something to be considered identified (i.e., 4) and thinking that it is the minimum number of points generated across all analytes of a given validated method (i.e., analyte A only achieves 3 points despite most of the other analytes in the method achieving 5, so minimum identification criteria is 3 for this method). I assume the first one, but the definition is ambiguous.	Change to "Lowest number of points, including a chromatographic separation, that must be achieved within a testing regimen to identify an analyte." (This also reflects language in 4.3.1.1.)	ACCEPT WITH MODIFICATION: The suggested change to the definition was made and further clarification was added.
15	4.1.3; 4.3.1.1	T	Are different blood sources (heart, femoral, cavity, etc.) considered "a given matrix" or "specific matrix"?	Add language if postmortem blood from different sources is considered separate matrices	ACCEPT: The following sentence was added to 4.1.3. for clarification: "For the purposes of identification, data obtained for a given matrix (e.g., blood, vitreous, or kidney) collected from different anatomic sites may be combined."

16	4.1.5	T	<p>A second analysis type, or at least second aliquot, is a widespread historical practice in forensic toxicology. One should not be able to report a result on a single analysis, regardless of the specificity/points achieved.</p> <p>EXAMPLE: an LC-QTOF screen, with product ion library match, would exceed the 4 point identification minimum. Consider a high-throughput laboratory using a broad-scope LC-QTOF screen with a rapid gradient: i.e. 3-5 minutes. This would not permit chromatographic resolution between critical isomer pairs with comparable or identical product ion spectra. Amphetamine and beta-methylphenethylamine is a good example, Another is Norfentanyl and Pipecoloxylidide (PPX, an N-desalkyl metabolite of Ropivacaine, Bupivacaine, and Mepivacaine), as is LSD and LAMPA and MIPLA. Not all laboratories would have evaluated these pairs during method validation specificity experiments. Without a suitable, separate confirmatory gradient to show retention time difference, the laboratory could erroneously report the wrong compound without requiring a confirmation.</p>	Change "should" to "shall".	REJECT: Analysis of a second aliquot is a quality assurance practice as opposed to a minimum expectation for identification. It is primarily to demonstrate sample mix-up did not occur. A laboratory may employ other techniques to ensure sample mix-up does not occur, such as barcode readers. The minimum points to identify an analyte is based on the points awarded to a single technique or combination of techniques and is not impacted by the possibility of samples being swapped. Section 4.2.1 requires that methods be appropriately validated in order to be awarded the specified points. For these reasons, 4.1.5 will not be changed to a "shall".
36	4.1.5	T	Why do these standards document set out such strict "minimum" criteria for identification, but then only say "should" when referring to analysis of two aliquots.	Revise to "shall"	REJECT: Analysis of a second aliquot is a quality assurance practice as opposed to a minimum expectation for identification. It is primarily to demonstrate sample mix-up did not occur. A laboratory may employ other techniques to ensure sample mix-up does not occur, such as barcode readers. The minimum points to identify an analyte is based on the points awarded to a single technique or combination of techniques and is not impacted by the possibility of samples being swapped. Section 4.2.1 requires that methods be appropriately validated in order to be awarded the specified points. For these reasons, 4.1.5 will not be changed to a "shall".
55	4.1.5	T	The underlined portion of this clause needs stronger language so it is clear that drug confirmation from a single aliquot is not permitted. "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or two different matrices from the same subject should be independently analyzed."	Two aliquots of the same or two different matrices from the same subject must be independently analyzed. Perhaps, add this best practice even earlier in the document. Upon initial review of this document, I was concerned that drug identification was permitted from a single aliquot analysis and that is not defensible.	REJECT: Analysis of a second aliquot is a quality assurance practice as opposed to a minimum expectation for identification. It is primarily to demonstrate sample mix-up did not occur. A laboratory may employ other techniques to ensure sample mix-up does not occur, such as barcode readers. The minimum points to identify an analyte is based on the points awarded to a single technique or combination of techniques and is not impacted by the possibility of samples being swapped. Section 4.2.1 requires that methods be appropriately validated in order to be awarded the specified points. For these reasons, 4.1.5 will not be changed to a "shall".
1	4.2.2	T	Lab funding makes concurrently ran reference standards to be impossible	Make it optional.	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte.
2	4.2.2	T	Concurrently analyzed reference standards are not always a feasible option for laboratories. Well-funded laboratories should not dictate the capabilities of all laboratories, nor should national standards be cost prohibitive to meet. National standards should be the minimum criteria, which should not be mistaken for a good practice. If this was a minimum criterion then I am sure the authors would have included it in the first draft as it would have been common knowledge. This requirement should be optional.	Change requirement to: At least one chromatographic or electrophoretic separation technique shall be performed to achieve identification, this should include a concurrently analyzed reference standard/positive control of the analyte of interest. Chromatographic or electrophoretic acceptability criteria (retention time, peak shape, resolution, signal to noise) shall be specified in the validated analytical method and shall be met for analyte identification.	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte.
4	4.2.2	T	Requiring concurrently analyzed reference standard for identification is impossible for every analyte due to laboratory funding. Lab budget does not allow for this.	Change requirement to: At least one chromatographic or electrophoretic separation technique shall be performed to achieve identification, this should include a concurrently analyzed reference standard/positive control of the analyte of interest. Chromatographic or electrophoretic acceptability criteria (retention time, peak shape, resolution, signal to noise) shall be specified in the validated analytical method and shall be met for analyte identification.	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte.

11	4.2.2	T	Following in the footsteps of my comment on 3.3, depending on how the definition is interpreted, the burden placed on the laboratory might be excessive, and absolutely not in line with how certain methods are currently implemented. Non-targeted methods (e.g., GC-MS full scan, LC-QToF) by nature look for thousands of analytes in a sample. Is this standard really requiring us to inject CRMs of thousands of molecules with each batch to achieve identifications? This is what my interpretation of the text leads me to believe, but it is simply impractical. While I agree that an identification should not be "library-based" only, and a CRM should at some point have been analyzed in the same analytical conditions for an ID to be valid, why does it need to be "concurrent" (whatever ASB wants this to mean)? This is not how wide scope methods are currently practiced. Furthermore, to me this is a validation issue. If we have proven that the method/system is stable over time and update in retention times or ion ratios aren't needed, I don't see why there would be an issue with not analyzing a concurrent standard/positive control.	Either remove this expectation, or modify to something along the lines that "points for chromatographic and electrophoretic separations are only awarded when a reference standard/positive control has been analyzed by the lab in the same analytical conditions (same instrument and instrumental parameters)".	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte and therefore would not be awarded the chromatography points.
13	4.2.2	T	including concurrently analyzed reference standard/positive control of the analyte of interest... is restrictive unnecessarily if the analysis is being performed on the same instrument and with the same parameters as defined in 3.3. Provided the instrument and parameters are the same, and the analyte meets established chromatographic quality criteria in the validated analytical method (as stated in 4.2.2), the chromatographic identification should be acceptable without routine concurrent analysis of a reference standard.	In the last sentence, insert "should be" and remove "are only"	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte.
37	4.2.2	T	Why is there no standards document for chromatography? What is the value of 1 point for chromatography if there is no standard for how assays are developed and/or employed?	Revise or develop chromatography standard	REJECT: While there is no specific standard for chromatography, a laboratory is expected to define their requirements (e.g., peak shape, retention time windows, baseline separation, signal-to-noise) as part of the validation of the method (ANSI/ASB 036) and document those requirements within their SOPs (Draft ASB 152).
53	4.2.2	T	Change concurrently running reference standards to be optional. Laboratories with limited funding may not have the resources.	change to "At least one chromatographic or electrophoretic separation technique shall be performed to achieve identification, this should including a concurrently analyzed reference standard/positive control of the analyte of interest. Chromatographic or electrophoretic acceptability criteria (retention time, peak shape, resolution, signal to noise) shall be specified in the validated analytical method and shall be met for analyte identification."	REJECT: This document specifically requires the use of a concurrently analyzed standard or positive control to determine the retention time in chromatographic/electrophoretic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard. Without such, a laboratory would not be able to demonstrate the proper retention time for the analyte.
72	4.2.2	T	There is a lot of criteria around mass spec data, but not chromatography. Will there be a standard for chromatography since a point is assigned?	Add criteria for chromatographic results and/or refer to a chromatography standard (like mass spec)	REJECT: While there is no specific standard for chromatography, a laboratory is expected to define their requirements (e.g., peak shape, retention time windows, baseline separation, signal-to-noise) as part of the validation of the method (ANSI/ASB 036) and document those requirements within their SOPs (Draft ASB 152).
12	4.3	T	This standard seems prone to generate false negatives. Indeed, a drug detected by LC-HRMS with a 5.1 ppm error would be considered "not identified", regardless of the retention time & fragments matching expectations. Unless this analysis can be downgraded to low resolution to salvage some points, a perfectly reasonable identification is thrown in the garbage, with potential serious impact for victims.	Add a section discussing how and allowing for a laboratory can handle "narrowly missed" identifications.	REJECT: To achieve identification, the minimum of four points must be reached. Analysis may need to be repeated or the method may need further validation if an assay "narrowly misses" the requirements set forth in this document or others.
17	4.3.1.1	T	Recommend listing examples of hyphenated techniques	...hyphenated techniques (e.g. GC-NPD, GC-MS, LC-MS/MS, etc.)	ACCEPT: Additional examples were added, as suggested.
18	4.3.1.1	T	If two GC-NPD analyses are utilized with two different column chemistries, that does not meet the point requirement.	Revise language to indicate this.	REJECT: The document already makes clear that a minimum of four points is required for identification. A dual column NPD analysis would be assigned 3 points. This is already demonstrated in Annex B.
60	4.3.1.1	T	Would be beneficial to include other examples of hyphenated techniques	Include other examples of hyphenated techniques (ie, GC-NPD, GC-MS, LC-MS/MS)	ACCEPT: Additional examples were added, as suggested.
56	4.3.1.2	T	This statement conflicts with 4.1.5 because 4.1.5 permits analysis of two different matrices from the same subject for identification, but 4.3.1.2 prohibits the use of identification of a drug based on its confirmation in blood and urine (two different matrices from the same subject).	Perhaps the document should delineate between best practice (analysis of more than a single aliquot for identification) and a rule about the assignment of points only applying to one of the aliquots, as in you can't count points twice. I think these two ideas are causing confusion.	ACCEPT: Section 4.1.5 was modified to delineate the best practice of analyzing an additional aliquot as opposed to the requirement within 4.3.1.2.
67	4.3.1.2	T	This criteria makes no sense. If a laboratory uses chromatography with MS and a contemporaneous standard for the second matrix why on earth would they need to run another immunoassay if the specimens are from the same subject and collected at ~ the same time?	Remove entire requirement	REJECT: The document does not define the analytical scheme that a laboratory chooses to use, but in order to identify an analyte in a given matrix, the minimum of four points is required.
57	4.3.1.3	T	This statement conflicts with 4.1.5 because 4.1.5 permits analysis of two different matrices from the same subject for identification, but 4.3.1.3 prohibits repetition of the same technique on the same matrix for purposes of awarding points for identification.	Similar comment to above, this document needs to make it clear that the best practice is to analyze more than a single aliquot for identification but the assignment of points only applies to one of the aliquots.	ACCEPT: Section 4.1.5 was modified to delineate the best practice of analyzing an additional aliquot as opposed to the requirement within 4.3.1.2.
19	4.3.1.4	T	Add "presumptive" prior to "positive" or "non-negative"	Revise to "presumptive positive"	REJECT: The sentence was modified for clarity, but the suggested change was not incorporated.

20	4.3.1.4	T	In the two situations cited in this section, cannabinoids like THC are described as having "low cross-reactivity" and alprazolam is described as having "good cross-reactivity"; however, there are no cutoffs described for cross-reactivity.	Define low and high cross-reactivity	ACCEPT WITH MODIFICATION: The examples were modified to clarify the role that method validation plays in determining when points are allowed for immunoassays that capture multiple analytes in their results.
21	4.3.1.4	T	Disagree with the situation defined in the 3rd example (quetiapine). The focus and significance of the analysis should be on the parent drug. If a more specific/more sensitive confirmatory test (i.e. LC/MS/MS) detected parent and metabolites, but only one metabolite was detected by a less sensitive screening test, this should still support identification of the parent drug in the confirmation.	Clarify rules to permit points to count towards identification of the parent drug when detected in a confirmation, but only a metabolite is detected by a screening/second test.	REJECT: Section 4.3.1.2 states: "Each identified analyte in each matrix shall independently meet the minimum point criteria."
52	4.3.1.4	T	Need a more objective guideline on what is considered "low cross-reactivity" and "good cross-reactivity" stated in the examples.	Add a requirement defining acceptable immunoassay cross-reactivity that can be counted for the identification points.	ACCEPT WITH MODIFICATION: The examples were modified to clarify the role that method validation plays in determining when points are allowed for immunoassays that capture multiple analytes in their results.
61	4.3.1.4	T	What is considered low cross-reactivity and high cross-reactivity?	Include cutoffs for low and high crossreactivity	ACCEPT WITH MODIFICATION: The examples were modified to clarify the role that method validation plays in determining when points are allowed for immunoassays that capture multiple analytes in their results.
62	4.3.1.4	T	Unclear why it is unacceptable that the presence of a metabolite detected by a less sensitive test should not be able to support the identification of a parent drug in confirmation.	Identification of the parent drug when detected in confirmation, but only a metabolite is detected by a less sensitive screening test should still be awarded points. Please clarify rules to allow for this.	REJECT: Section 4.3.1.2 states: "Each identified analyte in each matrix shall independently meet the minimum point criteria."
68	4.3.1.4	T	What if the diazepam result is 5 ng/mL and the alprazolam is 200 ng/mL? Immunoassays have extremely high cross reactivity to alprazolam and the response from the diazepam would not normally be detectable, if not for the presence of the alprazolam. The ASB is getting too far into the weeds and has made this document too restrictive and excessive. Why write a document that most laboratories will not be able to meet and claim it is a minimum standard? Is Forensic Toxicology that bad?	Revise to apply logic to this requirement.	ACCEPT WITH MODIFICATION: The examples were modified to clarify the role that method validation plays in determining when points are allowed for immunoassays that capture multiple analytes in their results.
22	4.3.2	T	The point system is confusing for some techniques, and could leave room for debate.	Include point totals for ALL routine hyphenated techniques, by themselves, in this table or in Annex B to avoid any confusion. Example: GC-MS, LC-MS/MS, LC-TOF, LC-QTOF, GC-MS/MS, GCxGC-MS, GC-MS/NPD, GC-FID, GC-NPD, etc.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. To list every possibility of combinations of chromatographic / electrophoretic systems with different detectors is overly burdensome. The goal is demonstrate how the points are combined so that laboratories are able to apply them to their own unique situations. The most common examples are included in Annex B.
23	4.3.2	T	The point system seems random and imbalanced. For example, why does a HRMS product ion scan yield 4 points on its own, even without chromatography?	Revise point totals, or add more specific scenarios to better aid in understanding. The point system should not permit for reporting on a single analysis - this exception should be clearly noted.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. Annex B contains over 20 of the most common scenarios found in forensic toxicology laboratories. Section 4.2.2 states: "At least one chromatographic or electrophoretic separation technique, including a concurrently analyzed reference standard/positive control of the analyte of interest, shall be performed to achieve identification." Further, Section 4.1.5 states: "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. As a matter of good laboratory practice, two aliquots of the same or different matrices from the same subject should be independently analyzed."
24	4.3.2	T	Is a "high resolution full scan" a precursor ion scan (i.e. a single stage HRMS scan like LC-TOF)? You would not use library matching to evaluate this data. You would use isotopic fit/modeling and internal mass error criteria for the observed precursor ion.	Remove this line from the table in the library matching category.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. A laboratory may conduct a full-scan HRMS library match and earn the allotted points, provided appropriate validation has demonstrated the limitations of the technique.
38	4.3.2	T	The points system is arbitrary and not well designed. How can a standard be developed with no scientific data to back it? Why is a total of 4 points needed? Does that really say anything about accuracy or reliability of results? Why can some analyses add up to nearly 10 points?	Revise based on scientific data (or comments below)	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. The point system was primarily based on the cited reference OJEC. "Commission Decision of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results (2002/657/EC). The Official Journal of the European Communities, 2002, L221/8. The concepts were modified to incorporate chromatographic separations, as well as other commonly used techniques (e.g., color tests, immunoassays, non-selective chromatographic detectors) into the point scheme. As specified in Section 4.2.1., points are applied to techniques validated according to ANSI/ASB 036 <i>Standard Practices for Method Validation in Forensic Toxicology Laboratories</i> .

39	4.3.2	T	What is the difference between all of the HR MS line items? There are not even this many options in the proposed Std 098 document. What is "High Resolution Full Scan"? This is essentially (or at least can be based on the 098 document) a precursor ion scan. Does a TOF-MS (precursor ion mass only) and a retention time really constitute identification and therefore reporting? The points do not align with logic.	Revise the points in relation to 098. Consider other scenarios that get you above 4 pts but should not constitute identification.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. ASB 098, Section 4.4 does allow for Single-Stage Mass Analyses Using High-Resolution Mass Analyzer. Further, as specified in Section 4.2.1., points are applied to techniques validated according to ANSI/ASB 036 Standard Practices for Method Validation in Forensic Toxicology Laboratories.
40	4.3.2	T	Why does the detection of HRMS fragment ions only add 0.5 points? A precursor ion on its own gives you 2.5 or 3.5 points (depending on how you categorize it), and then a product ion spectrum is 4 points. In most cases, the product ion spectrum is more specific than the precursor ion alone. This does not make sense and shows the value this points system offers to the practicality of forensic toxicology.	Revise points system (or make sure the 098 document is more properly laid out)	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. The value of HRMS is emphasized with the increased assignment of points compared to LRMS and the minimum identification threshold is 4 points.
63	4.3.2	T	The point system is confusing for some techniques, and seems unbalanced. Why would a HRMS product ion scan yield 4 points even without chromatography to separate isomers. What happens when only partial criteria is met? Does that allow for some point totals?	Revise point totals and include more specific examples to better aid in understanding.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. If only partial criteria are met, no points are awarded. For isomers, Section 4.3.1.5 states "Specific identification of an isomeric compound shall meet the minimum point requirements of this document (e.g., escitalopram, d-amphetamine). Unless differentiation is achieved, it is only acceptable to identify the mixed isomeric compound (e.g., amphetamine or d/l-amphetamine, methorphan or dextro/levomethorphan)."
64	4.3.2	T	Is a "high resolution full scan" a precursor ion scan (i.e. TOFMS)? As such, library matching would not be used for identification rather isotopic distribution based on theoretical isotopic patterns based on a chemical formula and mass error criteria for the precursor.	Remove this from the table in the library matching.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. Depending on how a HRMS full-scan is performed, a library match may be appropriate in an analytical scheme for identification. But if a laboratory is using this approach, they are required to validate their method and determine interfering substances that may limit identification. As this is an option in ASB 098, it must remain in this table.
73	4.3.2	T	Was there a formal evaluation of the points system to determine the 4 pt threshold for accuracy and reliability of results?	Evaluate if a points system is really the best way to determine reliability & accuracy of results; provide results of study, particularly with non-standard results.	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. The 4 point threshold was extensively challenged with different combinations of commonly used techniques to evaluate if appropriate for compound identification. See Annex B. It is also noted that the point system was primarily based on the cited reference OIEC. "Commission Decision of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results (2002/657/EC). The Official Journal of the European Communities, 2002, L221/8. The concepts were modified to incorporate chromatographic separations, as well as other commonly used techniques (e.g., color tests, immunoassays, non-selective chromatographic detectors) into the point scheme. As specified in Section 4.2.1., points are applied to techniques validated according to ANSI/ASB 036 Standard Practices for Method Validation in Forensic Toxicology Laboratories.
3	4.3.2 (table notes)	T	Concurrently analyzed reference standards are not always a feasible option for laboratories. Well-funded laboratories should not dictate the capabilities of all laboratories, nor should national standards be cost prohibitive to meet. National standards should be the minimum criteria, which should not be mistaken for a good practice. If this was a minimum criterion then I am sure the authors would have included it in the first draft as it would have been common knowledge. This requirement should be optional.	Remove note.	REJECT: This document specifically requires the use of a concurrently analyzed standard to determine the retention time in chromatographic analyses. To receive points assigned to chromatography requires the concurrently analyzed standard.
25	4.5.2	T	Unclear - where are different precursor ions originating from for the same compound? Is this discussing an in-source CID fragment? Or a C13 or halogen isotope (A+2)? Or a sodium adduct? Or is this addressing MS3 fragmentation and beyond?	Clarify or remove.	ACCEPT: Section 4.5.2 was clarified by adding examples.
41	4.5.2	T	Does this mean isotope precursor ions, such as the bromine M+2? Or other adducts? This is vague and confusing. What are these different precursor ions?	Revise (can not provide suggestion without better understand of context here)	ACCEPT: Section 4.5.2 was clarified by adding examples.
58	4.5.2	T	The use of different precursor ions for identification is confusing, but perhaps it is because we didn't design our methods this way.	Are the authors referring to different ionizations of the same drug? If so, I can see how that would fit and I would include an example that clarifies that here. Using a non-specific statement like "different precursor ions" could lead to misidentification.	ACCEPT: Section 4.5.2 was clarified by adding examples.

26	4.5.3	E	This comment applies both to this document and standard 098 for MS data. Here, the panel has recognized that a setting a number for a minimum recommended library match factor is not appropriate, as it will vary based on application, instrument technique, vendor platform, and software. It is left to the laboratory SOPs to define and verify through validation. If this is the case, why is a minimum mass error limit (5 ppm) set? This should also be left to internal laboratory SOPs and validations.	Apply consistent approaches between ASB standards. Remove mass error requirements from ASB standard 098.	REJECT: This recommendation is directed to ASB 098 and not this document.
27	4.5.4	T	Performing SIM and full-scan analysis simultaneously on the same sample with the same gradient does not provide additional specificity. It is unclear why this is listed as acceptable, but a separate SIM and full-scan analysis is not appropriate. Points should be awarded for one or the other mode of acquisition, but not both.	Remove sentence	ACCEPT: Sentence was removed.
42	4.5.4	T	By GCMS, this is really double dipping and should not be allowed. It does not add any specificity or accuracy.	Revise to not allow.	ACCEPT: Sentence was removed.
43	4.5.4	T	I think there is confusion about the data generated with HRMS systems here (unless this is not meant to include HRMS systems). Does this include HRMS systems? Remember: there are multiple mass spectra acquired. This still allows for using the TOF-MS and TOF-MS/MS data separately to rack up points.	Revise??? (unless the reader is unclear)	ACCEPT: Sentence was removed. Additionally, 4.5.5 was modified to include: "Points for both single- and multi-stage mass spectrometry with the same ionization process are allowed, provided different ions are monitored in each technique." This applies to low- and high-resolution instruments.
28	4.5.5	T	Ionization processes are able to change without chromatography changes, resulting in the potential for misidentification of analytes.	Remove sentence	Reject: Each of the different ionization techniques provides a different level of specificity. This is demonstrated through validation of the technique, as stated in Section 4.2.1. Points are only applied to techniques validated according to ANSI/ASB 036 Standard Practices for Method Validation in Forensic Toxicology Laboratories.
44	4.5.5	T	This really does not make sense. If you do not change your analytical the column, that is bad and not allowed. But if you switch the MS probe, that is good - and yet it still allows you to misidentify something in the same manner.	Remove this sentence.	Reject: Each of the different ionization techniques provides a different level of specificity. This is demonstrated through validation of the technique, as stated in Section 4.2.1. Points are only applied to techniques validated according to ANSI/ASB 036 Standard Practices for Method Validation in Forensic Toxicology Laboratories.
29	4.5.6	T	Based on the ASB 098 draft, 4.4d, "monitored diagnostic ions must be within 5ppm of the theoretical or measured m/z of a concurrently analyzed reference material." There are several situations where mass accuracy is between 5-10ppm. Are no points awarded for this identification?	Define point totals when 5ppm criteria are not met.	Reject: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. If the method was validated to meet the 5 ppm criteria and that is not met, then the points cannot be counted toward the identification of the compound.
45	4.5.6	T	Why only high res? Is this a typo?	Revise	Accept: The sentence was modified for clarity.
65	4.5.6	T	Based on the ASB 098 draft, "monitored diagnostic ions must be within 5ppm of the theoretical or measured m/z..." What happened when mass accuracy is between 5-10ppm, are no points awarded for this identification?	Clarification should be included on what the point totals would be if a 5 ppm criteria is not met. Other examples on what happens if criteria are not fully met should also be included in this document	Reject: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. If the method was validated to meet the 5 ppm criteria and that is not met, then the points cannot be counted toward the identification of the compound.
30	Annex B	T	Recommend modifying specific immunoassay techniques to immunoassay (IA) for the reader's ease since the different technologies all result in the same point totals (1)	Revise Annex B to remove all specific IA techniques (e.g., EMIT, CEDIA, ELISA)	ACCEPT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. Nonetheless, the CB has agreed to the suggested change, but differentiated immunoassays as instrumental or non-instrumental. An additional example was added for a non-instrument immunoassay.
31	Annex B	T	Recommend clarification in Annex B of the points for each item. This should be a standalone document and it would be beneficial to the reader to have the points listed next to each item. Two of the most common analyses are immunoassay paired with GC-MS and LC-MS/MS confirmation.	Immunoassay + LC-MS/MS (2 MRMs) = Immunoassay (1) + LC (1) + LR MS ² transitions (4) = 6 points Immunoassay + GC-MS (3 ions) = Immunoassay (1) + GC (1) + LR MS 3 ions (3) = 5 points	REJECT: Please note that comments on a re-circulation are generally accepted only on revised sections of a document. Comments made on text not revised from the previous public comment period are generally not accepted. It is noted that the CB did attempt to modify the Annex as the commentor suggested, but the table became even more difficult to read. The points listed in the second column of Annex B (Tabulation) are in the same order as the techniques listed in the first column. These values are defined in Section 4.3.2.

48	Annex B	T	<p>Scenario: LC-TOF-MS with full scan precursor ion match (precursor ion). This gives 1+3.5 points, enough for identification, but the detection could be non-specific as it does not allow for determination of isomers, etc. Many would agree that this would be non-specific, yet the points (and MS criteria document) allow it.</p>	<p>Many revisions may be necessary to both 113 and 098.</p>	<p>REJECT: No specific recommendation was made. It is noted that the points in the cited example, as interpreted, are correct but two additionally important concepts must be remembered: 1) Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed."; 2) Section 4.2.1 indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance, to include potentially interfering compounds. For isomers, Section 4.3.1.5 states "Specific identification of an isomeric compound shall meet the minimum point requirements of this document (e.g., escitalopram, d-amphetamine). Unless differentiation is achieved, it is only acceptable to identify the mixed isomeric compound (e.g., amphetamine or d/l-amphetamine, methorphan or dextro/levomethorphan)."</p>
49	Annex B	T	<p>Scenario: LC-TOF-MS with full scan precursor ion match (in source fragmentation gives rise to 91 and 119 ions). This again gives 1+3.5 points, enough for identification, but the detection could be non-specific (many amphetamine like compounds have similar fragments and no precursor ion was detected to distinguish formula). Many would agree that this would be non-specific, yet the points (and MS criteria document) allow it.</p>	<p>Many revisions may be necessary to both 113 and 098.</p>	<p>REJECT: No specific recommendation was made. It is noted that the points in the cited example, as interpreted, are correct but two additionally important concepts must be remembered: 1) Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed."; 2) Section 4.2.1 indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance, to include potentially interfering compounds. For isomers, Section 4.3.1.5 states "Specific identification of an isomeric compound shall meet the minimum point requirements of this document (e.g., escitalopram, d-amphetamine). Unless differentiation is achieved, it is only acceptable to identify the mixed isomeric compound (e.g., amphetamine or d/l-amphetamine, methorphan or dextro/levomethorphan)."</p>
50	Annex B	T	<p>Scenario: LC-TOF-MS with full scan precursor ion match (in source fragmentation gives rise to 91 and 119 ions). This could also give 1+2.5+2.5 points (for both ions), enough for identification, but the detection could be non-specific (many amphetamine like compounds have similar fragments and no precursor ion was detected to distinguish formula). Many would agree that this would be non-specific, yet the points (and MS criteria document) allow it. There is not criteria that says the precursor ion must be present. Also, these are (as currently defined) considered "diagnostic ions"</p>	<p>Many revisions may be necessary to both 113 and 098.</p>	<p>REJECT: No specific recommendation was made. It is noted that the points in the cited example, as interpreted, are correct but two additionally important concepts must be remembered: 1) Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed."; 2) Section 4.2.1 indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance, to include potentially interfering compounds. For isomers, Section 4.3.1.5 states "Specific identification of an isomeric compound shall meet the minimum point requirements of this document (e.g., escitalopram, d-amphetamine). Unless differentiation is achieved, it is only acceptable to identify the mixed isomeric compound (e.g., amphetamine or d/l-amphetamine, methorphan or dextro/levomethorphan)."</p>
51	Annex B	T	<p>Scenario: LC-QTOF-MS with product ion spectrum and library match (I have detected no precursor ions but have a fragment spectrum with 188 and 105 fragments). This could give 4 points (for lib match to fentanyl alone and ignore the chrom/RT because now it doesn't matter). As before, enough for identification, but the detection could be non-specific since I have no precursor ion data and 188/105 do not distinctively tell me what drug I detected (just maybe the drug class/subclass). Even though my "diagnostic ions" match fentanyl, they could also match many many other analogues (including methyl acetyl fentanyl which could have the same RT so that does not help). Many would agree that this would be non-specific, yet the points (and MS criteria document) allow it. I may have misidentified my drug as fentanyl. And the standard does not require me to run a second analysis so the problem goes unresolved (although a second analysis may not even help).</p>	<p>Many revisions may be necessary to both 113 and 098.</p>	<p>REJECT: No specific recommendation was made. Section 4.2.2 states: "At least one chromatographic or electrophoretic separation technique, including a concurrently analyzed reference standard/positive control of the analyte of interest, shall be performed to achieve identification." Further, Section 4.1.5 states that "Although one hyphenated instrumental technique (e.g., LC-MS/MS) may be sufficient to achieve identification, this alone does not ensure the reliability, reproducibility, quality, and integrity of results. Two aliquots of the same or different matrices from the same subject should be independently analyzed." Section 4.2.1 indicates that identification points are awarded only when "properly validated" methods are used. ASB 036 (Standard Practices for Method Validation in Forensic Toxicology) requires that a validation plan predefines acceptable method performance, to include potentially interfering compounds. For isomers, Section 4.3.1.5 states "Specific identification of an isomeric compound shall meet the minimum point requirements of this document (e.g., escitalopram, d-amphetamine). Unless differentiation is achieved, it is only acceptable to identify the mixed isomeric compound (e.g., amphetamine or d/l-amphetamine, methorphan or dextro/levomethorphan)."</p>

76	Annex B	T	There is no example for the most common technique in our field - LC-MS/MS with MRM.	Include this example to remove any such question/challenge in the future.	ACCEPT: The suggested example was added to Annex B.
----	---------	---	---	---	---