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Questions and answers for marketing authorisation holders/applicants on the CHMP Opinion for the Article 5(3) of Regulation (EC) No 726/2004 referral on nitrosamine impurities in human medicinal products

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Revision History

Rev.	Summary of changes made	Date
0	Replace obsolete Q&A published in 2019 to support the initial "call for review" with a new version reflecting the main principles agreed as part of the Article 5(3) referral which concluded in July 2020.	03rd August 2020
1	Update to Q&A 3 in order to clarify products in scope of the call for review. Update to Q&A 4 in order to add the link to the outcome of the referral under article 3 of Directive 2001/83/EC for ranitidine.	29th January 2021
2	Update to Q&A 3 on indicating testing timeline at the time of step 1 "risk identified" reporting.	24th February 2021
3	Update to Q&A 3 on the approach for non-marketed medicines. New Q&A 19 on the requirements for line extensions and variation applications.	15th April 2021
4	Update to Q&A 3 on combining step 2 response for multiple products from the same MAH.	18th May 2021
4*	Updates to Q&A 3 on when to perform step 2 confirmatory testing in order to meet the established deadline for step 3. Update and Q&A 10 to add an AI for NMOR.	29th June 2021
5	Update to Q&A 10 to add an AI for NNV.	21st September 2021
6	Guidance on confirmatory testing requirements for marketed (Q&A 8) and on-going applications (Q&A 14) to include cases where a potential nitrosamine impurity cannot be synthesised, and when a product is available in multiple strengths of the same dosage form.	14th October 2021
7	Inclusion of additional guidance on control strategies for products containing more than one nitrosamine impurity including examples (Q&A 10) and a decision tree (Annex I).	31st January 2022
8	Update to guidance on root causes and risk factors for nitrosamine contamination (Q&A 4) and on policy for confirmatory testing (Q&A 8) and dossier requirements (Q&A 15) to allow testing of intermediates, raw materials or API under certain circumstances.	24th March 2022
9	New Q&A 20 providing clarifications on what are the regulatory steps for dealing with scenario A cases and update Q&A10 with new AIs (N-nitrosomethylphenidate, N-nitrosopiperidine, N-nitrosorasagilene, 7-Nitroso-3-(trifluoromethyl)-5,6,7,8-tetrahydro[1,2,4]triazolo-[4,3-a]pyrazine, N-nitroso-1,2,3,6-tetrahydropyridine, N-nitrosonortriptyline, N-methyl-N-nitrosophenethylamine) and guidance on use of Ames test.	20th May 2022
10	Update to Q&A 5 to provide clarifications on the expectation for MAHs to continue to re-visit risk evaluations when new information becomes available with specific reference to API-nitrosamine risk. Update to Q&A 10 to include newly adopted AI for *N*-nitrosodabigatran and to indicate APIs where related nitrosamines have been identified. Clarification of how to set limits for products containing salt, hydrate or solvate forms of the API. Update to Q&A 14 to reference the new risk evaluation template for use in marketing authorisation applications.	23rd June 2022
11	Update to Q&A 3 on submission of amended step 1 response and extension of Step 3 deadline for chemical medicines.	29th July 2022
12	Update of Q&A 10 to add nitrosoduloxetine and introduction of Q&A 21 on approach to control presence of nitrosamine while the AI is being established.	10th October 2022
13	Update of Q&A 10 to add N-nitrosofluoxetine, N-nitrosoparoxetine, N-nitrosodiphenylamine, N-nitroso-mefenamic acid, N-nitrosopyrrolidine and N-nitrosodiethanolamine.	5th December 2022
14	Introduction of Q&A 22 on approach to control presence of N-nitrosamine exceeding the AI while CAPAs are being implemented. Update of Q&A 20 to consider the possibility of an interim limit based on the LTL approach during CAPA implementation. Update of Q&A 21	22nd December 2022

Rev.	Summary of changes made	Date
	for increased clarity on the application of the temporary universal limit.	
15	Amendment of Q&A 22 to indicate that no variation should be submitted to implement temporary above AI limits in specifications.	30th March 2023
16	Amendment to Q&A 10 to include the Carcinogenic Potency Categorization Approach (CPCA) and the enhanced Ames test (EAT) for establishing AIs for N-nitrosamines. Addition of Appendix 1, listing the nitrosamines for which AI have been established by the Non-clinical Working Party (NcWP), including new AIs for N-nitrosamines determined using the CPCA. Addition of Annex 2, describing the Carcinogenic Potency Categorization Approach for N-nitrosamines. Addition of Annex 3, describing the Enhanced Ames Test Conditions for N-nitrosamines.	7th July 2023
17	Amendment of Q&A 22 on approach to control presence of N-nitrosamine exceeding the AI while CAPAs are being implemented to extend the scope to authorised products for chronic use and clarify the applicable limits and exemptions. Amendment of Q&A 20 and Q&A 21 on approach to control presence of nitrosamine while the AI is being established to clarify that as the AIs can be established with the new carcinogenic category approach (CPCA) the approach for a universal temporary AI (t-AI) while a formal AI is established is no longer considered necessary.	28th July 2023
18	Update of Q&A 3 to highlighting the responsibilities of MAH(s) to control, report and mitigate the detection of Nitrosamine impurities throughout the product life-cycle, by using the established procedure.	2nd October 2023
19	Update of Q&A 10 to allow referral to other sources for CPCA categorisation, change reference from ICH M7(R1) to ICH M7(R2) guideline and removal of information in Annexes 2 and 3 to Appendices 2 and 3, respectively.	12th October 2023
20	Update of Q&A 3 and Q&A 10 to include guidance on non-mutagenic nitrosamine impurities (NMI) handling. Update to Q&A 9 to clarify sensitivity requirements for analytical methods. Update to Q&A 10 to include Ames test acceptability timelines.	15th January 2024
21	Updates to Q&As 8, 9, 10, 14 and 15 to clarify the expectations for risk assessments, confirmatory testing and dossier requirements for products where nitrosamine impurities can be controlled according to ICH Q3A/B limits. Update to Q&A 10 to update Ames test acceptability section. Update to Q&A 16 to clarify the responsibilities of MAHs and API manufacturers when reference is made to ASMFs and CEPs. Editorial correction of Q&A 4.	28th May 2024
22	Updates to Q&A 10 to clarify the applicable AI limit to different administration routes and accepted *in vivo* study type. Update to Q&A 22 to highlight the expectations for the submission of variations to shelf-life and storage conditions as needed to comply with interim limit during CAPA implementation.	19th May 2025
23	Update to Q&A 22 to clarify the timeframe of applicability of the interim limit during CAPA implementation.	10th October 2025

Introduction

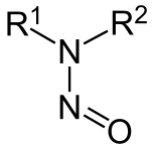
The assessment report of the CHMP's Article 5(3) of Regulation (EC) No 726/2004 opinion on nitrosamine impurities in human medicinal products provides general guidance and recommendations on mitigating and preventing the presence of nitrosamines in human medicinal products. In this context all MAHs/Applicants of human medicinal products should work with the manufacturers of their Active Pharmaceutical Ingredients (APIs) and finished products (FPs) in order to ensure that the presence of nitrosamine impurities in their medicinal products is mitigated as much as possible and controlled at or

below a limit defined based on ICH M7(R2) principles for substances of the "cohort of concern" reflected in this guideline and calculated considering a lifetime daily exposure and kept as low as possible and that appropriate risk mitigating measures are taken.

While the review by CHMP under Article 5(3) was ongoing, the regulatory authorities established in September 2019 a specific framework (hereinafter 'call for review')[1,2] for medicinal products containing chemically synthesised APIs, to provide details on the reporting to the authorities by the MAHs and set expectations regarding risk evaluation (step 1), risk assessment/confirmatory testing (step 2) and risk mitigation measures (step 3) to be carried out. Following the CHMP's Article 5(3) opinion, a similar exercise is launched for medicinal products containing a biological API, as further explained in this document. Further details are provided in Q&A 2 below.

The published CHMP Article 5(3) opinion, supplemented by the current Question and Answer document on its implementation, will replace the previous letter entitled 'Information on nitrosamines for marketing authorisation holders' (EMA/189634/2019, published on 19 September 2019).

The terms "nitrosamine" and "*N*-nitrosamine" are used interchangeably within this Q&A and related documents and should both be understood to refer to the following structure:



For the purpose of this Q&A please see definitions below:

Risk evaluation: all activities in step 1.

Risk assessment: all activities in step 2.

1. Should the risk of presence of nitrosamines be considered for all human medicinal products?

MAHs/Applicants of all human medicinal products should ensure that the presence of nitrosamines is controlled and kept as low as possible, irrespective of marketing status or the type of product (e.g. generics and over the counter (OTC) products).

For details on the approach required, please refer to Q&A 10 on the limits for nitrosamines and Q&A 12 on the measures to mitigate the risk of presence of nitrosamines.

MAHs/Applicants are reminded of their obligations to ensure that, in accordance with Article 23 and Annex I of Directive 2001/83/EC and Article 16 of Regulation (EC) No 726/2004, their medicinal products are manufactured and controlled by means of processes and methods in compliance with the latest state of scientific and technical progress.

Therefore, MAH/ Applicants shall:

- design their manufacturing processes and controls to prevent if possible or mitigate as much as possible the presence of *N*-nitrosamines in their API and FP(s);

[1] https://www.ema.europa.eu/en/documents/referral/nitrosamines-emea-h-a53-1490-information-nitrosamines-marketing-authorisation-holders_en.pdf
[2] https://www.ema.europa.eu/en/documents/referral/nitrosamines-emea-h-a53-1490-questions-answers-information-nitrosamines-marketing-authorisation_en.pdf

- assess the risk of presence nitrosamine impurities in their API(s) and FP(s) and introduce any resultant changes to the dossier as needed (e.g. changes to their manufacturing processes);
- ensure that active substances and excipients used in their FPs are manufactured in compliance with good manufacturing practices in line with Article 46(f) of Directive 2001/83/EC.

Compliance of the MAHs/Applicants with the above-mentioned obligations is subject to regular controls by competent authorities including during GMP inspections.

While the Article 5.3. recommendations on controlling nitrosamine impurities apply to all human medicinal products, the call for review applies only to human medicines containing chemically synthesised APIs or biological APIs, as further explained in Q&A 2 below.

2. What is the 'call for review'?

In September 2019, a 'call for review' was launched for medicinal products containing chemically synthesised APIs to request MAHs to review their manufacturing processes in order to identify and, if necessary, mitigate the risk of presence of nitrosamine impurities and report the outcome back to authorities. This exercise was started while the review by CHMP under Article 5(3) for Nitrosamine impurities in human medicinal products was ongoing.

Following the conclusion of the review under Article 5(3), the CHMP considered that there is also a risk of presence of nitrosamines in biological medicinal products, in particular for the biological medicines with the following risk factors:

- biologicals containing chemically synthesised fragments, where risk factors similar to chemically synthesised active substances are present;
- biologicals using processes where nitrosating reagents are deliberately added;
- biologicals packaged in certain primary packaging material, such as blister packs containing nitrocellulose.

For the above reasons the current call for review has been extended to cover also all biological medicinal products for human use. For further reference on what is considered to be a biological medicinal product for the purpose of this exercise, please consult the CMDh Questions & Answers on Biologicals.

The call for review consists of 3 steps:

- Step 1: MAHs to perform a risk evaluation to identify if APIs and/or FPs could be at risk of presence of nitrosamine;
- Step 2: if a risk is identified, MAHs to proceed with confirmatory testing in order to confirm or refute the presence of nitrosamines. MAHs should report outcomes as soon as possible;
- Step 3: if the presence of nitrosamine(s) is confirmed, MAHs should implement effective risk mitigating measures through submission of variation.

Please refer to Q&A 3 for further details on the 'call for review' including the timelines for chemicals and the timelines for biologicals.

For the specific case of sartans with a tetrazole ring that have been subject to a review under Article 31 of Directive 2001/83/EC, further guidance will be published soon.

3. For the 'call for review' for chemically synthesised and biological medicinal products, when and how should MAHs report steps 1 and 2 to competent authorities?

Submission of step 1 outcome

Products that have been approved after 26 September 2019 but for which a risk evaluation was not assessed within the MAA procedure should comply with the call for review deadlines, if not already done so.

For product containing **chemically** synthesised APIs, the step 1 risk evaluation should be concluded and reported at the latest by **31st March 2021**.

For product containing **biological** APIs, step 1 risk evaluation should be concluded and reported at the latest by **01st July 2021**.

The risk assessment has to be performed for all products for which a potential risk has been identified in step 1, irrespective of the marketing status of the product or whether any registered manufacturers are actively used in supply. However, it is recognised that step 2 may not be possible for medicines that are not marketed, including the case of manufacturers not actively used in supply, since there may be no finished product batches available for confirmatory testing. In these cases, i.e. where no batches of finished products are available, it would be acceptable to submit a written commitment that step 2 confirmatory testing will be conducted once finished product has been manufactured and/or the product is launched. The outcome of step 2 testing as well as any necessary variation(s) as part of step 3 will therefore need to be submitted and approved before the product can be placed on the market or the manufacturer can be actively used in supply, even if this is after the step 2 and 3 deadlines. MAHs'/Applicants' compliance with the above-mentioned obligations is subject to regular controls by competent authorities including during inspections.

All MAHs should inform the concerned Competent Authorities of the outcome of their risk evaluation (step 1) using the dedicated templates.

If a risk has been identified, the expected timeline for the testing activities should also be provided as foreseen in the dedicated template. No additional documentation is required at this stage. However, the risk review should be adequately documented, and related documentation should be made available upon request.

Step 2 should be started as soon as a risk is identified in API and/or FP and in accordance with product prioritisation (see Q&A 6).

If a risk has been identified for the API, the MAH is advised to report this outcome by using step 1 response template and to proceed directly to step 2 confirmatory testing of the FP. If no risk has been identified in the API, the MAH is advised to proceed with the risk evaluation of the FP and to present the result of Step 1 when a final conclusion has been reached on both the API and the FP. MAHs should inform the concerned Competent Authorities of the outcome of their risk evaluation (step 1) even if no risk has been identified in the API or FP.

It is acceptable for the submission of the outcome of step 1 to submit one email notification grouping products with identical outcome under the following provisions:

- For those Member States that have a dedicated portal, the MAH should submit the notification via this portal;

- If the outcome of step 1 is "risk identified", it is possible to provide a response by grouping these products. MAHs are still required to indicate the expected testing timeline on the related "Step 1 risk identified response template" excel file.

In specific cases it may be possible to correct a former step 1 outcome from "risk" to "no risk" by using the "Step 2 no nitrosamine detected response template". This template now contains a tick box for such cases. The possibility to amend the step 1 outcome may only be used in those cases where data was missing at the March 2020 deadline and is now available.

Submission of step 2 outcome

The step 2 confirmatory testing should be conducted in accordance with product prioritisation (see Q&A 6).

For nitrosamine impurities that are classified as non-mutagenic in Appendix 1 based on in-vivo mutagenicity studies, the submission of step 2 confirmatory testing is not required, and these impurities should be controlled according to ICH Q3A(R2) and ICH Q3B(R2) guidelines. For all other nitrosamine impurities the submission of step 2 confirmatory testing is required using the dedicated response templates.

For product containing **chemically** synthesised APIs, confirmatory testing activities at Step 2 are expected to be finalised at the latest by **26th September 2022.** MAHs should refrain from submitting incomplete step 2 outcomes.

The deadline for the submission of any changes required to Marketing Authorisations (Step 3, see Q&A 13) is by **1st October 2023**.

For product containing **biological** APIs, confirmatory testing activities at Step 2 and submission of any changes required to Marketing Authorisations (Step 3, see Q&A 13), are expected to be finalised at the latest by **1st July 2023.**

In order to meet the above deadlines for submission of any changes required to Marketing Authorisations at Step 3 for products containing chemically synthesised or biological APIs, it would be expected that confirmatory testing activities at Step 2 are finalised in advance of these deadlines.

MAHs should forthwith inform the competent authorities if tests confirm the presence of nitrosamine, irrespective of the amount detected and by utilising the dedicated reporting templates. The immediate risk to patients should be assessed based on the limits defined in Q&A 10 and appropriate action proposed to avoid or minimise the exposure of patients to nitrosamines.

For the submission of the outcome of step 2 confirmatory testing several products can be combined when the outcome is "no nitrosamines detected". When the outcome is "nitrosamines detected" all strengths and pharmaceutical forms of one marketing authorisation can be combined in one response template when the supporting documentation is completely identical for all products concerned; if not the response has to be submitted separately.

In case one or more nitrosamines are identified that exceed the limit defined in Q&A 10, the following supportive documentation is required at the time of reporting:

- testing results expressed in ng and ppm;
- interim investigation report including (preliminary) root cause, risk mitigating plan and benefit/risk assessment.

For their responses, MAHs are required to use dedicated templates and contact points as outlined on the EMA and CMDh websites.

Please note the set deadlines of the Call For Review (Step 1, 2 and 3) as described above for medicines containing chemically synthesised and biological active substances have passed. Any MAHs that have not reported identified Nitrosamine impurities to the relevant Competent Authority, should do so as a matter of priority, including any updates to previous notifications.

MAH(s) should report in accordance with the established limits and recommendations described in this guidance. MAH(s) should continue to use the response templates and available communication mechanisms previously established.

MAH(s) are reminded of their responsibilities to ensure the quality, safety and efficacy of their medicines and to adhere to the Nitrosamines guidance outlined by the EU Network. MAHs and Manufacturers should work together and take precautionary measures to mitigate the risk of presence of nitrosamines during the manufacturing and storage of all authorised medicinal products and throughout the lifecycle of the product if any changes are made.

Authorities in the EU will continue take all necessary measures to protect patients and ensure that medicines in the EU meet the required quality standards. The Authorities in the EU will also continue to collaborate with international partners on Nitrosamine to reflect scientific advances.

4. What are the currently identified risk factors for presence of nitrosamines?

N-Nitrosamines can be formed when an amine and nitrosating agent are combined under favourable conditions although other generation pathways are also possible, such as e.g. oxidation and reduction processes from hydrazine-type compounds and *N*-nitro derivatives.[3,4] Root causes for *N*-nitrosamines in medicinal products identified to date can be grouped as risk factors linked exclusively with the manufacturing process and storage of active substance and/or as risk factors associated with manufacture and storage of the finished product. Moreover, there are risk factors specifically linked to GMP aspects. Currently identified risk factors for *N*-nitrosamine impurities in medicinal products are listed below, along with some identified in the literature. However, the list is not exhaustive and further root causes may also be applicable – it is up to MAHs to determine if there is a risk with their product:

Risk factors related to the manufacture of the active substance:

1. Use of nitrite salts and esters (e.g. $NaNO_2$, alkyl nitrites), or other nitrosating agents (e.g. nitroso halides, nitrosonium salts, nitrogen oxides, nitro alkanes, halogenated nitro alkanes, Fremy's salt, nitroso sulfonamides),[3,4] in the presence of secondary or tertiary amines within the same or different steps of the manufacturing process. Sources for secondary or tertiary amines can also be starting materials, intermediates, reagents, solvents (e.g. DMF, DMAc and NMP) and catalysts, which contain amine functionality, amine impurities (e.g. quaternary ammonium salts) or which are susceptible to degradation to reveal amines.
2. Nitrite formation by oxidation of hydroxylamine or nitrite release from nitro-aromatic precursors (e.g. by fluoro de-nitration), in the presence of secondary or tertiary amines within the same or different steps of the manufacturing process (see 1).[5]

[3] Lessons learnt from presence of N-nitrosamine impurities in sartan medicines EMA/526934/2019.
[4] Org. Process Res. Dev. 2020, **24** (9), 1558–1585
[5] Chem. Rev. 2016, **116**, 422−518

3. Use of disinfected water (chlorination, chloro-amination, ozonisation) in the presence secondary or tertiary amines within the same or different steps of the manufacturing process (see 1).[6,7,8,9]

4. Oxidation of hydrazines, hydrazides and hydrazones by hypochlorite, air, oxygen, ozone and peroxides in the manufacturing process or during storage.[4]

5. Use of contaminated raw, recovered or recycled materials (e.g. solvents, reagents and catalysts) in the API manufacturing process.

6. Use of contaminated starting materials and intermediates supplied by vendors who use processes or raw materials which may contain residual nitrosamines or nitrosating agents.

7. Carry-over of nitrosamines deliberately generated (e.g. as starting materials or intermediates) during the manufacturing process.

Risk factors also related to the finished product:

8. Reaction of nitrosatable nitrogen functionality in APIs or their impurities/degradants with nitrosating agents present in components of the FP during formulation or storage. A particular risk of formation of nitrosamines should be noted for active substances that contain a nitrosatable amine functional group. Several examples have been reported where the amine functionality was shown to be vulnerable to nitrosation and formation of the corresponding *N*-nitroso impurity (i.e. NO-API). Secondary amines appear particularly vulnerable to this reaction although some cases with tertiary amines have also been observed. Vulnerable amines could also be formed by degradation (e.g. hydrolysis) during formulation or storage. Nitrites have been identified as impurities in many common excipients.[10] MAHs and/or applicants should be aware that *N*-nitroso API impurities can form at levels exceeding the AI even if nitrite levels in the excipients are very low. The overall nitrite content will also depend on the relative composition in terms of the excipients. As it has been reported that *N*-nitroso impurities can form from APIs or their impurities/degradants (containing amine functionality or susceptible to degradation to reveal amines) during manufacture of the finished product, as well as during storage, MAHs should give consideration to the stability of the finished product and should ensure that the AI of any *N*-nitrosamine impurity is not exceeded until the end of shelf life of the FP. For further information, please refer to the assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products.

9. Degradation processes of active substances, including those induced by inherent reactivity (e.g. presence of nitro-alkyl, oxime, or other functionality[3][11,4]) or by the presence of an exogenous nitrosating agent. This could potentially occur during both active substance and finished product manufacturing processes or during storage and could be influenced by crystal structure, crystal habit and storage conditions (temperature, humidity etc.). For more details, refer to page 6 of Referral under Article 31 of Directive 2001/83/EC for ranitidine and published literature.[11,12]

10. Oxidation of hydrazine or other amine-containing functional groups present in active substances or their impurities/degradants (e.g. from hydrazones and hydrazides), either in active substance manufacturing processes or during storage.[4] This root cause has also been observed during

[6] Crit. Rev. in Environ. Sci. 2017, 47, (24), 2448-2489
[7] J. Pharm. Biomed., 2019, **164**, 536-549
[8] Water Research, 2011, **45** (2), 944-952
[9] J. Org. Chem. 2021, **86**, 2037−2057
[10] *AAPS Pharm. Sci. Tech.* 2011, **12** (4), 1248- 1263
[11] *Org. Process Res. Dev*. 2020, **24** (12), 2915–2926
[12] *Chem. Pharm. Bull.,* 2021, **69**, 872–876

manufacture and storage of finished products containing such functional groups. Potential oxidants include oxygen and peroxides (common impurities in some excipients).[10]

11. Use of certain packaging materials. Relevant nitrosamine contamination has been observed in primary packaging of finished products in blister with lidding foil containing nitrocellulose. During the blister heat-sealing process, nitrogen oxides can be generated thermally from nitrocellulose. Under these conditions, nitrosamines have been shown to form from low molecular weight amines present either in printing ink or in the finished product and to transfer to the product and/or to the cavity via evaporation and condensation.

12. Reaction of amines leaching from quaternary ammonium anion exchange resins (e.g. used for purification steps) with nitrosating agents present in the liquid phase. A recent example of this was in the production of water for injections where residual chloramine used to disinfect incoming water reacted with dimethylamine leaching from the anion exchange resin used in the demineralisation step to form NDMA. In addition, disinfection procedures such as e.g. chlorination, chloro-amination and ozonisation can lead to significant *N*-nitrosamine generation as by-products in case vulnerable amines are present.[6,7,8,9] Given the source of contamination, risk is related to the concentration of the reactive agent(s) and thus to the volume of water in or used to dilute a particular product. The same risks could be associated with active substances or finished products manufactured using water purified using similar resins.

Risk factors related to GMP aspects:

13. Cross-contamination due to different processes being run successively on the same manufacturing line.

14. Carry-over of impurities between process steps due to operator-related errors or insufficiently detailed batch records such as inadequate phase separations during work-up procedures.

15. Use of contaminated recovered or recycled materials (e.g. solvents, reagents and catalysts) where the recovery is outsourced to third parties who are not aware of the content of the materials they are processing. Recovery processes carried out in non-dedicated equipment should also be considered.

5. What to do if after submission of step 1 and /or step 2 responses, new information (e.g. related to new potential risk factors or root causes) is identified?

MAHs together with API and FP manufacturers are expected to maintain the quality of their product throughout its lifecycle. Therefore, once step 1 and/or 2 responses are submitted, MAHs are expected to continue to review and re-visit the outcome of the risk evaluation as and when new information becomes available. MAHs are advised to routinely check this Q&A document and in particular Q&A 4 which will be kept up to date as regards newly identified risk factors for formation of nitrosamines, and also Q&A 10 concerning limits for nitrosamines.

In particular, MAHs should note the risk of formation of nitrosamine impurities from active substances (or their related impurities) containing a vulnerable amine during finished product formulation and/or storage due to the presence of traces of nitrites. This has been recently elaborated as a risk factor to Q&A 4 (bullet 8) based on understanding gained during the call for review. MAHs that did not take into account this risk as part of step 1 response for their products containing active substances with vulnerable amines should reconsider their original step 1 risk evaluations in light of this new information and proceed to step 2 confirmatory testing as appropriate (see also Appendix 1).

Appropriate timelines for reviewing the previous risk evaluation and for conducting confirmatory testing (if needed), should be followed depending on the risk identified.

The same approach should be followed for medicinal products granted a positive opinion and marketing authorisation during the call for review.

6. What factors should be considered in prioritising the risk evaluation?

When conducting the risk evaluation and risk assessment, MAHs should use a risk-based approach to prioritise products for evaluations and confirmatory testing. MAHs may consider factors such as the maximum daily dose taken for the concerned medicinal product, duration of treatment, therapeutic indication and number of patients treated. For example, medicinal products with higher daily dose and those for chronic use may take priority.

In order to undertake the analysis of the identified medicinal products at risk, MAHs can also use tools such as Failure Mode Effects Analysis (FMEA) and Failure Mode, Effects and Criticality Analysis (FMECA) as outlined in the ICH Q9 guideline on quality risk management.

7. How should the risk evaluation be performed?

MAHs/Applicants in collaboration with API, FP manufacturers and their raw material suppliers are required to perform risk evaluations using quality risk management principles, as outlined in ICH Q9 guideline. The principles described in ICH M7 guideline and in the Assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products in relation to the toxicology assessment, control strategy and changes to the manufacturing processes for active substances should also apply.

Manufacturers of active substances and FP and their raw material suppliers should provide MAHs/applicants with all information necessary for a comprehensive risk evaluation. If the risk of nitrosamine impurity formation was assessed during the development phase of the API/FP manufacturing processes, the information from this assessment can be used to support the risk evaluation.

MAHs/Applicants and manufacturers should consider as part of the risk evaluation all potential sources of contamination or formation of nitrosamine, notably the root causes listed under Q&A 4.

As MAHs/Applicants and manufacturers for products containing biological APIs should consider the following aspects that may increase the risks of nitrosamine presence in their products:

- biologicals containing chemically synthesised fragments, where risk factors similar to chemically synthesised active substances are present;
- biologicals using processes where nitrosating reagents are deliberately added;
- biologicals packaged in certain primary packaging material, such as blister packs containing nitrocellulose.

For further information on root causes, please refer also to the assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products.

If, after completion of the risk evaluation, a risk is identified in the API and/or the FP, MAHs/applicants must notify the competent authorities of the identified risk, proceed without further delay with confirmatory tests (see Q&A 8) and introduce any necessary changes to the dossier.

All MAHs should inform the concerned Competent Authorities of the outcome of their risk evaluation (step 1) even if a risk has not been identified, please see Q&A 3 for further details.

8. How should confirmatory tests be conducted by MAHs and manufacturers?

For the purpose of confirmatory testing as part of step 2 of the call for review to MAHs, testing should generally be carried out on the FP. Testing of the API, its intermediates, starting materials, solvents, reagents, excipients or any other raw materials for nitrosamines, amines, nitrites or other compounds with potential to generate nitrosamines is also recommended, if the risk assessment indicates that they are a potential source of nitrosamine impurities in the FP. In such cases, the results of testing API, intermediates or other relevant materials may be used to support root cause investigations and the development of a justified control strategy for nitrosamine impurities.

However, some root causes may only be linked to the API manufacturing process (see Q&A 4). In these cases, testing of the API or intermediates upstream of the active substance could be used as a surrogate for testing the finished product, provided that the risk assessment performed on the FP concluded no additional risk factors for formation of nitrosamine impurities in the finished product (see Q&A 4, risk factors related to the finished product). If testing is carried out on an intermediate, then there should also be no risk factors associated with subsequent steps in the API manufacturing process or the finished product. The confirmatory testing strategy is the responsibility of the MAH and should be justified based on the risk assessment for the finished product and documented in the MAH's pharmaceutical quality system. It should be clearly justified why testing of the active substance or intermediate is appropriate and why further risk of nitrosamine formation in the finished product or subsequent API manufacturing steps can be excluded. If nitrosamines are detected, then an appropriate control strategy should be implemented in the dossier.

In any case, if the control point of nitrosamines is not in the finished product, the responsibility for quality lies with the MAH.

The number of batches to be tested should be commensurate with the risk. MAHs and manufacturers should test a representative number of batches of FP and the relevant starting materials, intermediates, API or raw materials as applicable. If the source of risk has been identified and is well understood (e.g. by spike and purge studies) such that impurity levels are expected to be consistent from batch to batch, testing should be conducted on 10% of annual batches, or 3 per year, whichever is highest. This includes testing not only of newly produced batches but also retained samples of batches still within expiry date. If fewer than 3 batches are manufactured annually, then all batches should be tested.

If multiple manufacturers, manufacturing processes and/or sources of at-risk raw materials are used, (or were used historically for batches still within expiry date), then testing of additional batches would be necessary to cover these risk factors.

If a product is available in multiple strengths of the same dosage form with the same risk factors applicable to each, then testing could be rationalised by testing only the worst-case scenario strength.

The worst-case approach should be justified by the MAH on a case-by-case basis. The justification should be documented in the risk assessment in the MAH's pharmaceutical quality system.

During development of an analytical method, a reference standard of the relevant nitrosamine impurity is generally needed. If, despite extensive efforts, it becomes apparent that the relevant nitrosamine impurity cannot be synthesised, then this could be an indication that the nitrosamine either does not exist or that there is no risk of it being formed. In such cases, it may not be necessary to conduct confirmatory testing. This should be justified thoroughly on a case-by-case basis according to appropriate scientific principles. The justification could include relevant literature, information on structural/stereo-electronic features and reactivity of the parent amine, stability of the nitrosamine and experimental data to illustrate the efforts made to synthesise and to analyse the impurity. The justification should be documented in the risk assessment in the MAH's pharmaceutical quality system.

For products where nitrosamine impurities can be controlled according to ICH Q3A/B principles, see Q&A 10, confirmatory testing is generally not needed if the risk can be sufficiently mitigated based on scientific considerations that demonstrate that the relevant ICH Q3A/B thresholds will not be exceeded. In such cases, the justification should be documented in the risk assessment in the MAH's pharmaceutical quality system. For products with only advanced cancer indications as defined within the scope of ICH S9, and where the above principle has been applied to mitigate the potential risk of nitrosamine impurities, the approach should be revised should the indication ever move beyond the relevant ICH S9 setting. In these instances, confirmatory testing would generally be required considering the resultant lower applicable limits when ICH Q3A/B ceases to apply.

Methods for determination of various nitrosamines in sartans with a tetrazole ring, metformin and ranitidine have already been developed by the Official Medicines Control Laboratories and are available for reference on the European Directorate for the Quality of Medicines & HealthCare (EDQM) website. These may serve as a starting point for the development and validation of analytical methods for testing other APIs/FPs.

Appropriately sensitive analytical methods for determination of specific nitrosamines in other medicinal products should be developed, see Q&A 9 and validated accordingly before testing.

Given the trace levels of nitrosamines to be measured, the following technical aspects should be considered when developing analytical methods:

- Interference caused by presence of trace amounts of nitrosamines in testing materials utilised (e.g. water, airborne sources, plastics products and rubber/elastomeric products);
- Contamination during sample preparation (avoiding cross contaminations from gloves, membranes, solvents etc.) which could lead to false positive results;
- *In situ* formation of nitrosamines during analysis;
- Use of accurate mass techniques are required (MS/MS or high-resolution accurate mass systems) in order to overcome interference in the identification of the specific peak of a certain nitrosamine (e.g. false positives have been observed from DMF co-eluting with NDMA).

As a result of the above considerations, control experiments should be conducted such as analysing samples by orthogonal analytical methods.

Further details in relation to analytical methodology can be found on EDQM website and in the CHMP assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products.

9. What are the requirements of the analytical method(s)?

The analytical methods need to be sufficiently sensitive in order to adequately detect and quantify trace levels of nitrosamine impurities. When developing the analytical method, the required sensitivity should derive from the appropriate acceptable intake determined in line with the approaches described in Q&A 10. Appropriate development of the analytical method and the required sensitivity are the responsibility of the MAH/applicant. The following principles apply:

- The limit of quantification (LoQ) provides the minimum level at which an analyte can be quantified with acceptable accuracy and precision and should thus be used for impurity testing and decision-making;
- If quantitative testing is performed as a routine control, the LoQ should be ≤ of the acceptable limit based on the relevant acceptable intake (AI) for the respective nitrosamine impurity;
- If quantitative testing is performed to justify skip testing, the LoQ of the analytical procedure employed should be ≤ 30% of the acceptable limit based on the AI;
- If quantitative testing is performed to justify omission of specification, the LoQ of the analytical method employed should be ≤ 10% of the acceptable limit based on the AI;
- Exceptions are anticipated for medicinal products used at high daily doses (AI may be below technical feasibility of the method), or in case more than one nitrosamine is anticipated or identified in a given medicinal product.

Different analytical methods may be used for determination of multiple nitrosamines. If the same analytical method is used for multiple nitrosamines, the selectivity of the method should be demonstrated for each nitrosamine.

For products where nitrosamine impurities can be controlled according to principles of ICH Q3A/B, see Q&A 10, where tested the LoQ for the analytical procedure should be at or below the applicable reporting threshold for the respective drug substance and/or drug product.

10. Which limits apply for nitrosamines in medicinal products?

ICH M7(R2) guideline defines N-nitrosamines as substances of the "cohort of concern" for which limits in medicinal products refer to the so-called substance-specific acceptable intake (AI) (the Threshold of Toxicological Concern, TTC, value of 1.5 ug/day cannot be routinely applied) which is associated with a negligible risk (theoretical excess cancer risk of <1 in 100,000 over a lifetime of exposure). The calculation of AI assumes a lifelong daily administration of the maximum daily dose of the medicinal product and is based on the approach outlined in the ICH M7(R2) guideline as well as the principles described in relation to the toxicological evaluation in the assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products.

The 'less than lifetime' (LTL) approach should not be applied in calculating the limits as described above but can only be considered after consultation with competent authorities as a temporary measure until further measures can be implemented to reduce the contaminant at or below the limits defined above.

For products intended for advanced cancer only as defined in the scope of the ICH S9 guideline, N-nitrosamine impurities should be controlled according to ICH Q3A(R2) and ICH Q3B(R2) guidelines, as specified in the Q&A document to ICH S9 guideline. In addition, limits according to ICH Q3A(R2) and ICH Q3B(R2) apply to any impurities in Appendix 1 considered to be a non-mutagenic impurity (NMI) based on negative results from a well-conducted *in vivo* mutagenicity study.

If the active substance itself is mutagenic or clastogenic at therapeutic concentrations, N-nitrosamine impurities should be controlled at limits for non-mutagenic impurities according to ICH M7(R2).

The same risk approach is applicable to all routes of administration. Corrections to limits are generally not acceptable.

Establishment of the AIs

Two scenarios are foreseen for detection of new nitrosamines:

A. If N-nitrosamines are identified with sufficient substance specific animal carcinogenicity data, the TD50 should be calculated and used to derive a substance specific limit for lifetime exposure as recommended in ICH M7(R2) guideline.

B. If N-nitrosamines are identified without sufficient substance specific data to derive a substance specific limit for lifetime exposure as recommended in ICH M7(R2) guideline,

    1. The Carcinogenic Potency Categorization Approach (CPCA) for N-nitrosamines (Appendix 2) should be used to establish the AI, unless other robust data are available that would override this AI.

    2. A negative result in an GLP-compliant enhanced Ames test (EAT, Appendix 3) allows control of the N-nitrosamine at 1.5 µg/day. For substances testing positive, the AI should be established using options 1 or 3. For reporting requirements see Q&A 3 above.

    3. If a surrogate nitrosamine is available with sufficiently robust carcinogenicity data, the TD50 from the surrogate substance can serve as a point of departure for derivation of AI by SAR and read across.

    4. A negative result in a relevant well-conducted *in vivo* transgenic rodent (TGR) mutagenicity study (OECD 488) can allow control of the N-nitrosamine as a non-mutagenic impurity (NMI), i.e. according to ICH Q3A(R2) and ICH Q3B(R2) limits, irrespective of the limit calculated through option 1, 2 or 3. For substances testing positive, the AI should be established using options 1 or 3. For reporting requirements see Q&A 3 above.

Appendix 1 lists the nitrosamines for which acceptable intakes have been established by the Non-clinical Working Party. If the nitrosamine is not included in Appendix 1, MAH/MA applicants can also refer to a CPCA category from another source e.g. CPCA categories published by other regulatory authorities, but this will need confirmation to allow control of the substance at the level corresponding to that category.

All Ames assays initiated after August 2023 must comply with the requirements of the EAT protocol (Appendix 3).

Calculation of the limit when a single known nitrosamine is identified

The conversion to a specification limit in ppm for a particular medicinal product is calculated by dividing the respective AI in Appendix 1 (ng) by the maximum daily dose (mg) of a given product as reflected in the SmPC.

The maximum daily dose is defined in line with the definition of the product strength in the Guideline on the SmPC. Therefore, the limit in ppm should usually be expressed per active moiety (free base, free acid or anhydrous/non-solvated material) for control point in the FP. Exceptions to this are active substances in medicinal products where the strength has traditionally been expressed in the form of a salt or hydrate and active substances present in the formulation as ester or pro-drug.

For a control point in the API only, the limit should be expressed in general per drug substance (i.e. relating to form of salt, hydrate, solvate etc. where relevant).

Calculation of limit when more than one nitrosamine is identified in the same product

Note: This section does not apply to scenarios where the nitrosamine impurities are intended to be controlled according to ICH Q3A/B principles.

Please also refer to the decision tree in Annex 1 for further guidance.

For determining limits in the case of presence of more than one nitrosamine, two approaches are considered acceptable in order not to exceed the acceptable risk level of 1:100,000 as outlined in ICH M7(R2) guideline:

1. The total daily intake of all identified *N*-nitrosamines not to exceed the AI of the most potent *N*-nitrosamine identified, or

2. Total risk level calculated for all identified *N*-nitrosamines not to exceed 1 in 100,000.

The approach chosen needs to be duly justified by the MAH/Applicant.

Specifications for individual *N*-nitrosamines should generally include an AI limit expressed in ppm or ppb. The conversion to an AI limit in ppm/ppb for a particular medicinal product is calculated by dividing the respective above AI (in ng/d) by the maximum daily dose (in mg) of a given product as reflected in the SmPC. The calculation of the specification limit does not take into account the molecular weight of the *N*-nitrosamine.

It is considered that the presence of one or more *N*-nitrosamines at <10% of their respective AI constitutes a negligible toxicological risk, and as such, they do not need to be specified. *N*-Nitrosamines present below 10% of their respective AI do not need to be factored into the calculation of limits for individual or total *N*-nitrosamine(s).

However, the overall principle of the Article 5(3) referral should still be considered, notably that "*the presence of N-nitrosamines in human medicinal products shall be mitigated as much as possible*." Therefore, manufacturers are encouraged to improve their processes, even if they result in only very small amounts (<10% AI) of multiple nitrosamines, as processes and controls should be designed to prevent if possible or mitigate as much as possible the presence of *N*-nitrosamines in APIs and FPs (see Q&A 1).

For option 1, the AI limit for total *N*-nitrosamines should be set in ppm/ppb according to the most potent *N*-nitrosamine present at ≥ 10% of its AI. The most potent nitrosamine is the one with the lowest AI (see Appendix 1). Limits for individual *N*-nitrosamines can be defined but are not necessarily needed. However, it should be clearly stated which *N*-nitrosamines are included in the calculation of total *N*-nitrosamines.

For option 2, the limits for *N*-nitrosamines should ensure an overall risk of not more than 1 in 100,000. Different approaches can be employed to achieve this risk requirement:

Fixed approach: fixed AI limits (in ppm/ppb) are set for individual nitrosamines and no limit for total *N*-nitrosamines is needed. The limit for each *N*-nitrosamine should be set at a percentage of its AI limit such that the sum of the % AI limits for each specified nitrosamine does not exceed 100%.

Flexible approach: each *N*-nitrosamine should be specified at its AI limit in ppm/ppb and an additional limit for total *N*-nitrosamines is required. The calculation for total *N*-nitrosamines could be written as:

$$\sum_{i=2}^{n} \frac{Xi}{AIi} \; x \; 100\% \; \leq 100\%$$

Where Xi is the amount of each single *N*-nitrosamine i in ppm and AIi is the AI limit of each *N*-nitrosamine i in ppm.

For each batch, to determine whether the limit for total *N*-nitrosamines is met, the amount of each *N*-nitrosamine present (in ppm/ppb) should be converted to a percentage of its respective AI limit. The sum of % AI limits of specified *N*-nitrosamines should not exceed 100%.

Example of control options and specifications for multiple nitrosamines in the same finished product:

The case of two NAs:

Two NAs both at or above 10% of their respective AI

Example:
NDMA and NDEA are both detected at or above 10% of their respective AI) in a finished product with maximum daily dose of 300 mg.

AI limit

- NDEA: 26.5 ng/day / 300 mg/day = 0.088 ppm or 88 ppb = most potent N-nitrosamine
- NDMA: 96.0 ng/day / 300 mg/day = 0.32 ppm or 320 ppb

Specification possibilities for different control options:

Nitrosamine	**Option 1**	**Option 2 - Fixed** **Example 20:80 ratio**[2]	**Option 2 - Flexible**
NDMA	Not needed	NMT 64 ppb *(320 ppb x 0.2)*	NMT 320 ppb
NDEA	Not needed	NMT 70 ppb *(88 ppb x 0.8)*	NMT 88 ppb
Total Nitrosamines	NMT 88 ppb	Not needed	NMT 100%[1]

[1] $(\frac{[NDMA]\ ppb}{320\ ppb} + \frac{[NDEA]\ ppb}{88\ ppb})\ x\ 100\% \leq 100\%$
NMT 100% = 1:100,000 theoretical excess cancer risk.
[2] For option 2 fixed approach, a ratio of 20% NDMA to 80% NDEA (20:80) is used as an example only. Different ratios could be used in different situations dependent on relative amounts present, provided that the sum of the % AI limits for each specified nitrosamine does not exceed 100%.

Example of presentation of acceptable batch results for each control option:

Model data from 1 batch:

- NDMA found at 38 ppb
- NDEA found at 44 ppb

	Option 1		**Option 2 - Fixed** **Example 20-80 ratio**		**Option 2 - Flexible**	
	Limit	**Results**	**Limit**	**Results**	**Limit**	**Results**
NDMA	Not needed	-	NMT 64 ppb	38 ppb	NMT 320 ppb	38 ppb (12% of AI)
NDEA	Not needed	-	NMT 70 ppb	44 ppb	NMT 88 ppb	44 ppb (50% of AI)
Total NA	NMT 88 ppb	82 ppb	Not needed	-	NMT 100%	62%

Control options for Genotoxic APIs

Genotoxicity encompasses mutagenicity, clastogenicity and aneugenicity.

Mutagenic APIs are defined as substances having DNA-reactive properties as described in ICH M7(R2).

Clastogenic APIs are substances causing structural chromosomal aberrations.

Aneugenic APIs are substances causing numerical chromosomal changes.

The ICH M7(R2) guideline does not apply to drug substances and drug products intended for advanced cancer indications as defined in the scope of ICH S9 (Ref. 4). Additionally, there may be some cases where a drug substance intended for other indications is itself genotoxic at therapeutic concentrations and may be expected to be associated with an increased cancer risk. Exposure to a mutagenic impurity in these cases would not significantly add to the cancer risk of the drug substance. Therefore, impurities could be controlled at acceptable levels for non-mutagenic impurities. Below it is explained in more detail how this is applied to the control of nitrosamine impurities.

1. Policy for products not within the scope of ICH S9

   a. Containing mutagenic or clastogenic APIs:

      i. Control nitrosamine at or below ICH Q3A/B qualification threshold[1] when genotoxicity of API is considered to produce a significant risk for mutagenicity/clastogenicity at therapeutic exposures;

      ii. The rules established for the control of nitrosamines as explained in the Article 5(3) referral or elsewhere in the Q&A apply when mutagenicity/clastogenicity of API is considered not to produce a significant risk for mutagenicity/clastogenicity at therapeutic exposures.

   b. Containing aneugenic APIs:

      i. The rules established for the control of nitrosamines as explained in the Article 5(3) referral or elsewhere in the Q&A apply since aneugenicity of API is considered not to produce a significant risk for carcinogenicity at therapeutic exposures

   c. Containing non-genotoxic APIs

      i. The rules established for the control of nitrosamines as explained in the Article 5(3) referral or elsewhere in the Q&A apply

2. Policy for products within the scope of ICH S9

   a. Containing genotoxic or non-genotoxic APIs:

      i. Control nitrosamine at or below ICH Q3A/B qualification threshold.

Higher limits may be set for nitrosamines in certain cases. However, it is expected that the Applicant/MAH will ensure that the presence of nitrosamine impurities in their medicinal products is mitigated as much as possible.

[1] Wherever it is quoted ”Control nitrosamine at or below ICH Q3A/B qualification threshold”, this implies that control at the qualification threshold is justified from a safety perspective.

11. What should I do if a nitrosamine is detected in my medicinal product?

If one or several nitrosamine(s) is detected for the first time in my medicinal product:

The MAH/Applicant should forthwith inform the competent authorities, irrespective of the amount detected as described in Q&A 3 for medicinal products subject to the call for review.

The levels should be reported in ng and ppm, together with the corresponding calculations used to describe the potential exposure to the detected nitrosamine based on the maximum daily dosage recommended in the SmPC. If SmPCs differ between Member States, the calculations should be provided for each different maximum exposure. Sufficient details should be provided to enable the calculations to be reviewed and verified.

The calculated exposure(s) should then be compared to the limit defined in Q&A 10:

- If the limit is not exceeded for the detected nitrosamine or, in case of presence of multiple nitrosamines, if the total risk remains below a theoretical lifetime excess risk of $\leq$1:100,000, the MAH/Applicant shall control the nitrosamine(s) in the FP at or below this limit (see Q&A 10) and should take measures to mitigate the risk of nitrosamine formation or contamination in the medicinal product as much as possible (see Q&A 12).
- Where the limit defined in Q&A 10 for single or multiple nitrosamines is exceeded, the MAH/Applicant should submit forthwith an (interim) investigation report including (preliminary) root cause, risk mitigating plan and benefit/risk assessment. The competent authorities will then assess the impact on the benefit/risk balance and the consequent need for any action to be taken.

Please refer to the Assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products for further information.

Changes to the marketing authorisation related to measures to prevent or minimise the risk should be introduced without delay and in accordance with the guideline on classification of variation (please refer to Q&A 13).

If the presence of specific nitrosamine(s) in a medicinal product has already been reported to the authorities by the MAH and is below the limit defined in Q&A 10 or a limit approved by the authorities, there is no need for a further notification to the authorities.

Batch records are subject to inspection by competent authorities.

12. Which are the measures to mitigate the risk of presence of nitrosamines?

The presence of N-nitrosamines in the FP shall be mitigated as much as possible and shall be at or below a limit defined in Q&A 10.

MAHs shall design or adapt the manufacturing process of their medicinal products to prevent formation of and contamination with nitrosamines whenever possible.

MAHs should implement a control strategy regarding *N*-nitrosamines, which should include current and prospective measures to minimise the risk of generation of/contamination with nitrosamines (e.g. change of manufacturing process, change of raw material quality, introduction of appropriate specifications and development of appropriate methods, and measures on the premise and equipment such as cleaning procedures and environmental monitoring). MAHs should control nitrosamine levels in accordance with

the limits defined in Q&A 10 and any future changes that may impact on the risk (e.g. change of supplier, change of manufacturing process and change of packaging).

MAHs shall also ensure that active substances and excipients used in their FPs are manufactured in compliance with good manufacturing practices in line with Article 46(f) of Directive 2001/83/EC.

Please refer to the Assessment report of the CHMP's Article 5(3) opinion on nitrosamine impurities in human medicinal products for further information.

13. Which changes would be required for Marketing Authorisations?

MAHs should introduce changes to their API and/or FP (e.g. manufacturing process, controls and specification, product formulation, raw materials and packaging), through the timely submission of appropriate variation(s) in accordance with the guideline on classification of variations.

When nitrosamine(s) is (are) identified, the corresponding limit(s) as defined in Q&A 10 should be introduced in the specifications of the FP. Please refer to Q&A 15 for information on the test modalities.

The application for a variation should contain information on amendments to the marketing authorisation – i.e. in module 3 (3.2.S and 3.2.P), the active substance master files (ASMF) or the Certificate of Suitability to the monographs of the European Pharmacopoeia (CEP) that is necessary to control nitrosamine impurities in the active substance and/or FP. Variations should be submitted according to the existing variations classification guideline: EUR-Lex - 52013XC0802(04) - EN - EUR-Lex (europa.eu)

Depending on the root cause identified and extent of changes to be made, grouping of variations or use of work-sharing procedures might be applicable: https://www.ema.europa.eu/en/human-regulatory/post-authorisation/variations/worksharing-questions-answers.

14. What is the approach for new and ongoing marketing authorisation applications (MAA)?

Applicants shall design their manufacturing processes and controls to prevent if possible or mitigate as much as possible the presence of *N*-nitrosamines in their API and FPs (please refer to Q&A 12).

The potential presence of nitrosamines must be evaluated as part of the MAA as follows:

- **At the submission stage:**
  - For the risk evaluation, Applicants are required to follow the principles for step 1 as per Q&A 2. The risk evaluation should be submitted as an attachment to Module 1 with a corresponding reference in Module 3.2 of the marketing authorisation dossier. To supplement the detailed risk evaluation, the template located on the CMDh nitrosamine website (section "For additional specific information related to nationally authorised products (including MRP/DCP)") could also be submitted: https://www.hma.eu/human-medicines/cmdh/nitrosamine-impurities.html. The template is optional for CAPs. For NAPs, and DCPs, the template is mandatory and the CMDh practical guidance located in the same section of the same website should be followed.
  - If a risk of presence of nitrosamines in the medicinal product is identified, applicants are required to provide the risk assessment outlining the impact on the benefit-risk balance of the product and a risk mitigation strategy. Applicants should also submit confirmatory testing plans or confirmatory testing data as mentioned in step 2 (see Q&A 2).

- In case applicants have not submitted a risk evaluation and, if applicable, confirmatory testing plans with their MAA, these should be submitted during the marketing authorisation review procedure.

- **During the Marketing Authorisation (MA) evaluation procedure:**
  - If the risk evaluation was not submitted as part of the MAA, it will be requested during the MA review process. Risk evaluation will have to be adequately documented and, if applicable, supported by confirmatory testing in case a possible risk of presence of nitrosamines has been identified. This information should be submitted as part of the responses to the list of questions.
  - If the applicant is not able to provide satisfactory information and justification of a favourable benefit-risk profile of the product at this stage, a request to further assess the risk of presence of nitrosamine will be part of the further list of questions / outstanding issues depending on the stage of the MA procedure.
  - Any outstanding issues related to the quality requirements of the product would have to be addressed before the final opinion on the granting of the MA.

For new and on-going marketing authorisation applications, the number of batches to be tested as part of any confirmatory testing should be commensurate with the risk in line with ICH M7(R2) guideline. The source of risk has to be well understood (e.g. by spike and purge studies) such that impurity levels are expected to be consistent from batch to batch. Test results from a minimum of 6 pilot scale batches or 3 production scale batches may be sufficient. Depending on the risk factors for nitrosamine presence, e.g. with risk factors being closer to the FP, more batches may need to be tested. If multiple manufacturers, manufacturing processes and/or sources of at-risk raw materials are used, (or were used historically during development), then testing of additional batches would be necessary to cover these risk factors.

If a product is available in multiple strengths of the same dosage form with the same risk factors applicable to each, then testing could be rationalised by testing only the worst-case scenario strength. The worst-case approach should be justified by the MAH on a case-by-case basis.

During development of an analytical method, a reference standard of the relevant nitrosamine impurity is generally needed. If, despite extensive efforts, it becomes apparent that the relevant nitrosamine impurity cannot be synthesised, then this could be an indication that the nitrosamine either does not exist or that there is no risk of it being formed. In such cases, it may not be necessary to conduct confirmatory testing. This should be justified thoroughly on a case-by-case basis according to appropriate scientific principles. The justification could include relevant literature, information on structural/stereo-electronic features and reactivity of the parent amine, stability of the nitrosamine and experimental data to illustrate the efforts made to synthesise and to analyse the impurity. The justification should be included in the submitted risk assessment.

For products where nitrosamine impurities can be controlled according to ICH Q3A/B principles, see Q&A 10, confirmatory testing is generally not needed if the risk can be sufficiently mitigated based on scientific considerations that demonstrate that the relevant ICH Q3A/B thresholds will not be exceeded. In such cases, the justification should be documented in the risk assessment in the MAH's pharmaceutical quality system. For products with only advanced cancer indications as defined within the scope of ICH S9, and where the above principle has been applied to mitigate the potential risk of nitrosamine impurities, the approach should be revised should the indication ever move beyond the relevant ICH S9 setting. In these instances, confirmatory testing would generally be required considering the resultant lower applicable limits when ICH Q3A/B ceases to apply.

15. When should a test for nitrosamines be included in the MA dossier?

When a nitrosamine is identified after Step 2 confirmatory testing, a limit will usually need to be included in the specifications of the finished product and the product must comply if tested. If the root cause has been identified in the finished product manufacturing process or storage, or nitrosamines have been detected in the finished product, but the actual source of contamination remains unclear, routine testing of the finished product is required by default.

The control point (finished product, API or an intermediate) for nitrosamines should be selected in such a way that it will give assurance of presence of the impurity below the acceptable limit based on acceptable intake (AI) in the finished product. Testing is usually expected to be carried out in the finished product, however if the source of a nitrosamine impurity is identified in the active substance manufacturing process, control options 1 to 3 as stated in ICH M7(R2) guideline could be used to demonstrate that the nitrosamine will not be present above the acceptable limit based on AI in the finished product. Testing of raw materials (e.g. excipients) should also be considered if these are potential sources of nitrosamine impurities. Exceptions from routine testing may be possible, if the root cause of contamination is demonstrated to be well-understood:

- Only if the amount of nitrosamine present is consistently below 10% of the acceptable limit based on AI in the API or in the finished product, then a test for the nitrosamine could be omitted from the specification.
- Only if levels of a single nitrosamine are consistently below 30% of the acceptable limit based on AI in the API or the finished product, skip-testing according to the ICH Q6A definition could be acceptable.

Where nitrosamine impurities can be controlled according to ICH Q3A/B principles and the risk can be sufficiently mitigated based on scientific considerations that demonstrate that the relevant ICH Q3A/B thresholds will not be exceeded, then a specification limit in the dossier is not expected.

16. What are the responsibilities of MAHs for APIs with CEPs or ASMFs?

MAHs/Applicants, manufacturing authorisation holders and API manufacturers should work together and take precautionary measures to mitigate the risk of presence of nitrosamines during the manufacture and storage of all medicinal products containing chemically synthesised APIs.

MAHs/Applicants must ensure that appropriate and robust risk evaluations are carried out by the relevant manufacturing authorisation holders and API manufacturers (including ASMF or CEP holders) in accordance with Article 46 of Directive 2001/83/EC.

APIs documented in CEPs and ASMFs may be used in different finished products, potentially with different formulations, manufacturing processes and indications. To mitigate levels of nitrosamines impurities , a given finished product may need a particular quality of API with additional tests or tighter limits compared to the Ph. Eur. monograph of that substance, e.g. tighter limits for amine impurities that may be converted to nitrosamines during formulation and storage of the finished product. It is the responsibility of the MAH to work with its API suppliers to ensure that API of acceptable quality is used for a given finished product amongst any other measure that may be implemented to reduce nitrosamine impurities. Additional parameters and appropriate limits should therefore be included in the finished product manufacturer's active substance specification as required.

17. How does the lessons learnt exercise from presence of nitrosamines in sartans relate to the Article 5(3) Referral Outcome?

The lessons learnt exercise was conducted by experts from the EU Regulatory Network to determine which lessons can be learnt from the handling of the cases of sartans with nitrosamine impurities. The objective is to make recommendations on how to reduce the risk of such impurities in medicines and to ensure that regulators are better prepared to manage cases of unexpected impurities in the future. Although the exercise focussed on lessons learnt from the assessment conducted for the sartans with a tetrazole ring, the recommendations apply to all human medicines.

The recommendations set forward include new or additional guidance on areas such as the control of impurities (including cohort of concern compounds), Good Manufacturing Practice, the roles and responsibilities of manufacturers and MAHs/Applicants but also proposals for improvement of communication with patients and healthcare professionals and cooperation with international partners. The full recommendations are available on EMA's website. The European medicines regulatory network will develop an implementation plan and then work with the parties that will implement each action.

It should be noted that the lessons learnt exercise outcome has been taken into account in the Article 5(3) procedure. The implementation of recommendations of the lessons learnt exercise will strengthen the regulatory framework and complement the outcome of this Article 5(3) procedure which provides the scientific opinion on the presence of nitrosamine impurities in human medicines.

18. What about regulatory requirements in other regions?

Regulatory authorities in the EU have been cooperating with international partners in the United States, Canada, Japan, Singapore, Switzerland, Australia and other countries to mitigate presence of nitrosamines in medicinal products and to align requirements. For questions about regulatory requirements outside the EU, please contact the relevant authorities.

19. What is the approach for line extensions and variations applications not linked to changes required as part of article 5(3) recommendation?

No risk evaluation is generally necessary when submitting line extension or variation application. The risk evaluation is only required to be submitted for products in scope of the call for review as reported in Q&A 3.

Nevertheless, in some exceptional cases questions on the presence of nitrosamines in the product may be raised if a potential risk is identified during the assessment.

20. What are the regulatory steps taken by authorities following the identification of an N-nitrosamine exceeding the AI?

The regulatory process dealing with the outcomes of the call for review is outlined in European Medicines Regulatory Network approach for the implementation of the CHMP Opinion pursuant to Article 5(3) of Regulation (EC) No 726/2004 for nitrosamine impurities in human medicines.

Chapter 3.2 provides a description on how regulators will approach the outcome from the call for review in accordance with the different scenarios reported by MAHs.

In case of identification of one or more N-nitrosamine exceeding the AI in the finished product, or in case that the sum of all detected N-nitrosamines exceeds the 1 in a 100,000 lifetime risk (scenario A), the following steps are taken in order to protect public health and ensure availability of critical medicines:

- A lead authority is identified as responsible for reviewing the information available and for providing the (preliminary) assessment of the case. The lead authority is selected as outlined in chapter 5.1.
- The Rapid Alert Network (RAN) and the availability Single Point Of Contacts (SPOCs) are informed in order to determine the criticality of the product (in accordance with Criteria for classification of critical medicinal products for human and veterinary use).
- The feedback from RAN and availability SPOCs is taken into account by the lead authority when providing the preliminary recommendations on any interim or eventual required market actions and on the acceptability of corrective and preventive actions proposed by the MAH.
- The Incident Review Network (IRN) is consulted in order to facilitate the exchange of information and to evaluate whether additional measures are needed or whether a different regulatory pathway is warranted.
- If market actions are recommended, each National Competent Authority (NCA) will follow up in accordance with their national procedures and depending on the criticality of the product for their markets.
- The use of an interim limit based on the LTL approach during CAPA implementation, as described in Q&A 22, may be considered, as applicable, by the lead authority and NCAs on a temporary basis for market action purposes. Please refer to chapter 3.2.1.1 of the regulatory process dealing with the outcomes of the call for review referenced above.

21. What is the approach to control the presence of nitrosamines until a substance specific AI is established?

Considering the new approaches for setting nitrosamines limits using the carcinogenic potency categorisation approach (CPCA) and the enhanced AMES test (EAT) protocol (see Q&A 10 above), the approach for a universal temporary AI (t-AI) while a formal AI is established is no longer considered necessary, as such the contents of this question has been deleted in July 2023.

22. What is the approach to control presence of N-nitrosamine exceeding the AI during CAPA implementation? (Updated)

In accordance with the regulatory steps taken by authorities following the identification of an *N*-nitrosamine exceeding the AI and outlined in Q&A20, the less-than lifetime (LTL) concept or the use of interim limits may be considered by the lead authority and NCAs on a temporary basis in order to inform market actions and at the same time ensure availability of medicines. MAHs are expected to establish and implement corrective and preventive actions (CAPAs) in authorised medicines without any delays in order to ensure patients safety and product quality. Nevertheless, it is recognised that implementation of CAPAs may require some time before the MAH is able to mitigate the presence of the identified *N*-nitrosamine below the established AI. Therefore, in order to avoid unnecessary risk of supply disruptions,

a harmonised approach promoting the establishment of interim limits in a streamlined way is agreed. The approach is applicable to all authorised products that have:

- CAPA implementation timeline of up to 3 years from the date of publication of the initial AI (nevertheless MAHs are expected to expedite CAPAs implementation).

Treatment duration	Up to 12 months	>12 months
Interim limit	13.3 x AI*	6.7xAI*

*In any case the limit should not exceed 1.5 µg/day unless the established AI (Table 1, Q&A10) is > 1.5 µg/day or the nitrosamine concerns a category 5 according to CPCA or the nitrosamine is shown to be negative in an enhanced Ames test (EAT).

The approach is not applicable to the below instances where other approaches may be considered on a case-by-case basis in consultation with the appropriate regulatory authority:

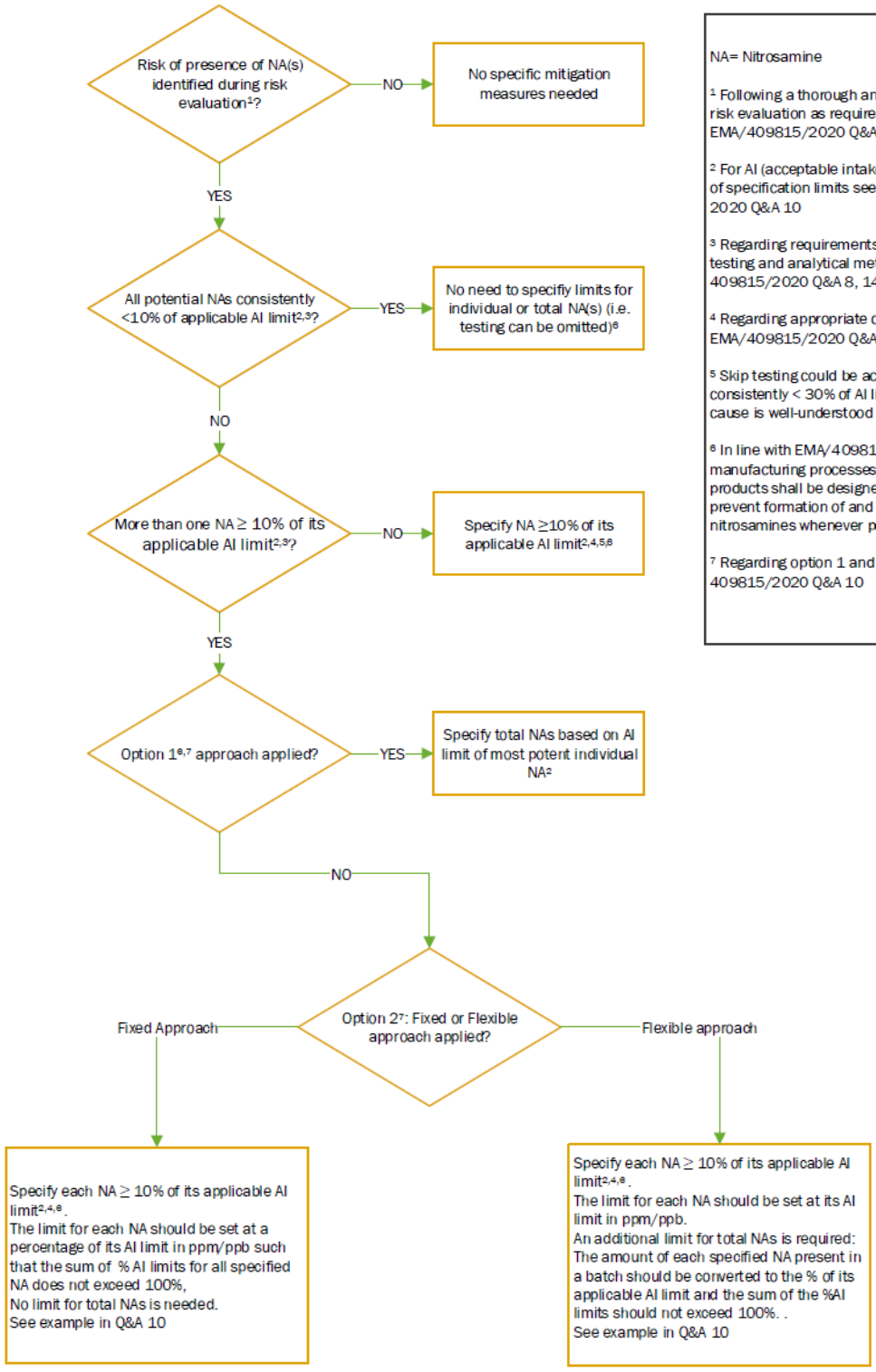
- CAPA implementation exceeding 3 years from the date of publication of the initial AI (this date which starts the 3-years period will not change if the AI is subsequently revised, even in case of publication of more restrictive AI). In cases where CAPAs are not implemented within the agreed timeframe please refer to Q&A 20 for the regulatory steps taken by authorities.
- New/ongoing regulatory applications.

The above interim limits are based on the LTL approach outlined in the ICH M7 guideline, using the two most conservative adjustment factors (6.7 and 13.3 x AI). The application of these adjustment factors would not be expected to exceed a theoretical excess cancer risk of 1 in 100,000 during the period of CAPA implementation.

The approach is intended to be evaluated by the lead authority during the assessment of the case and is expected to be communicated by the lead authority to the concerned MAH as part of assessment conclusions. In terms of retrospective application, where more restrictive interim limits were previously agreed for some products as part of case assessment, upon request from the MAH, the lead authority can re-assess interim limits taking into consideration this approach to control presence of N-nitrosamine exceeding the AI during CAPA implementation.

MAHs are expected to ensure that the implementation of adequate controls for the detected nitrosamines is done as a matter of priority. During the use of the interim limit, monitoring measures may be evaluated by the lead authority as required. However, it is not the expectation that MAHs include these interim limits in specifications via variation. Changes in shelf life and storage conditions to comply with the interim AI are evaluated by the relevant authority and are expected to be introduced without delay and in accordance with the guideline on classification of variation (please refer to Q&A 13).

Annex 1: Decision tree with control options for products containing multiple *N*-nitrosamines:



NA= Nitrosamine

[1] Following a thorough and comprehensive risk evaluation as required according to EMA/409815/2020 Q&A 7

[2] For AI (acceptable intakes) and calculation of specification limits see EMA/409815/2020 Q&A 10

[3] Regarding requirements of confirmatory testing and analytical methods see EMA/409815/2020 Q&A 8, 14 and 9

[4] Regarding appropriate control point see EMA/409815/2020 Q&A 15

[5] Skip testing could be acceptable, if NA is consistently < 30% of AI limit and the root cause is well-understood

[6] In line with EMA/409815/2020 Q&A 12 manufacturing processes of medicinal products shall be designed or adapted to prevent formation of and contamination with nitrosamines whenever possible.

[7] Regarding option 1 and 2 see EMA/409815/2020 Q&A 10