

MUT/MIN/2011/2

COMMITTEE ON MUTAGENICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

Minutes of the meeting held at 10.30 am on Thursday 16th June 2011 in Room 102A Skipton House, Elephant and Castle, London, SE1.

Present:

Chairman: Professor P Farmer

Members: Dr B Burlinson
Dr G Clare
Dr D Gatehouse
Professor G Jenkins
Dr D Lovell
Dr A Lynch
Dr E Parry
Professor D Phillips
Mrs R Glazebrook

Secretariat: Mr J Battershill (HPA secretariat)
Dr L Hetherington (HPA secretariat)
Mr S Robjohns (HPA minutes)
Ms S Kennedy (HPA administration)
Dr D Parker (FSA)

Assessors: Dr R Shillaker (HSE CRD)

In attendance: Dr K Burnett (HPA – Tox unit)
Dr P Edwards (HPA)
Mr G Evans (VMD)

A G E N D A

Paragraph

Open session

1. Announcements/Apologies for absence 1
2. Minutes of the meeting held on 21st October 2011 (MUT/MIN/10/3) 4
3. Matters arising 5
 - 3.1 Testing and assessment of impurities

Closed session

4. Evaluation of additional genotoxicity data on fumagillin dicyclohexylamine

Open session

5. Revision of COM guidance on a strategy for testing of chemicals for genotoxicity 12
6. Further information on QSARs as used by the secretariat (MUT/2011/10) 24
7. Any other business 33
8. Date of next meeting: 20th June 2011 34

ITEM 1: ANNOUNCEMENTS/APOLOGIES FOR ABSENCE

1. The Chair welcomed Dr D Parker attending for Dr D Benford (FSA secretariat), Dr K Burnett (HPA Tox unit), Dr L Hetherington (HPA secretariat) Mr S Robjohns (HPA) and Mr Gary Evans (VMD).

2. Apologies for absence were received from the members Dr C Allen, Dr B Elliot and Professor D Kirkland. Apologies were also received from Dr D Benford (FSA) and the assessors Ms F Pollitt (HPA), Dr O Sepai (HPA), Dr A Smith (HSE), Ms C Pease (EA) and Dr H Stemplewski (MHCRA).

3. Members were reminded of the need to declare any interests before discussion of items.

ITEM 2: MINUTES OF MEETING ON 10th March 2011 (MUT/MIN/11/1)

4. Members agreed the minutes subject to some editorial changes.

ITEM 3: MATTERS ARISING NOT COVERED BY LATER AGENDA ITEMS

3.1 Testing of impurities (MUT/2011/09)

5. At the previous meeting the COM had discussed a strategy paper for the mutagenicity testing of impurities. Members agreed that that it would be prudent to await the publication of an ICH step 2 document (expected in 2012) and EFSA WG consideration of the threshold of Toxicological Concern (TTC) before finalising this guidance.

6. The committee reached a number of interim conclusions in March 2011, which were as follows:

- The Threshold of toxicological concern (TTC) was a useful concept in identifying impurities requiring genotoxicity testing, although reference needed to be made to chemical classes of concern e.g. aflatoxin-like, azoxy and N-nitroso-compounds.
- The combination of the Ames test and the *in vitro* micronucleus test (MNvit) would optimise the detection of the mutagenic hazard of impurities.
- Overall, impurities should be isolated and tested separately. QSAR data would be helpful, but would be limited in usefulness for novel structures and in some instances where metabolic activation of a pro-mutagen occurred.

7. The members were asked for their view on the revised flow diagram provided in MUT/2011/09 and for any information on international developments regarding impurities. It was suggested that the COM could provide interim guidance on the testing of impurities whilst awaiting the outcome of ICH and EFSA discussions.

8. The committee agreed that it would be useful to consult with colleagues with expertise in computational toxicology. If the QSAR predictions were outside the domain of applicability (as indicated by CEASAR) then the prediction could not be used. However, if the prediction was in the applicability domain and negative, then

there was no need for genotoxicity testing. If there was a positive QSAR prediction for an impurity or if impurities fell outside the applicability domain of the QSAR models, then testing would be required.

9. Members agreed that if an impurity would be present in a final product at a concentration below the TTC (i.e. equivalent to an estimated exposure of 0.15 micrograms per person per day) then testing would not be required (excluding the recognised chemical classes of concern e.g. aflatoxin-like, azoxy and N-nitroso compounds). Members noted the 'classes of concern' listed in the flow diagram would need to reflect all those in the published guidance on the TTC. If an impurity was present in a final product at a concentration where exposure would exceed the TTC for genotoxic substances and in the absence of reliable negative QSAR predictions, then testing would be required in the Ames assay and the MNvit.

10. Additionally, the committee agreed that expert judgement should be used on a case-by-case basis and that for any potentially mutagenic impurities the as low as reasonably practicable (ALARP) approach should also be adopted.

11. Overall, the COM agreed the proposed interim guidance and flow diagram on the testing of impurities which should be amended in-line with the above comments. A revised flow diagram would be circulated by post.

Closed session

ITEM 4: EVALUATION OF ADDITIONAL GENOTOXICITY DATA ON FUMAGILLIN DICYCLOHEXYLAMINE (MUT/2011/07)

Open session

ITEM 5: REVISION OF COM GUIDANCE ON A STRATEGY FOR TESTING OF CHEMICALS FOR GENOTOXICITY: POST CONSULTATION DRAFT (MUT/2011/08)

12. At the March COM 2011 meeting, the committee continued development of the Guidance document and started to review the comments received during the consultation process (MUT/11/03). Most of the significant comments were addressed during the meeting, but there was insufficient time to review all the comments in detail. It was decided that the remaining comments of a more editorial nature could be reviewed by a COM Working Group (WG).

13. The WG comprised the Chair and three other COM members, and representatives from the HPA secretariat and the HPA Toxicology unit. The final reviewed document produced from the WG meeting was provided with the new alterations present as track changes. Consultation comments that had been considered and rejected were highlighted.

14. Members were asked to consider the revised document and provide any comments.

15. The Chair thanked the Working Group members for their considerable work on the revised document.

16. Regarding a pre-meeting comment from the FSA secretariat that it would have been useful for the draft Guidance to provide advice on how to test chemicals with existing genotoxicity data, the committee reaffirmed its earlier decision to initially focus this guidance document on the testing of chemicals with no data and then to produce additional guidance on the testing and mutagenic hazard assessment for chemicals with limited or inadequate genotoxicity data. In the interim, the revised Guidance document could be used to aid the interpretation of chemicals with little or inadequate data and to determine further testing requirements.

17. The committee provided a number of comments on the draft document. The COM agreed that some of the current draft could be expanded to include some of the useful text on the definition of 'genotoxicity' from the previous draft document.

18. It was important to carefully check the context in which the terms 'genotoxicity' and 'mutagenicity' were used throughout the document.

19. Regarding *in vitro* tests, there was a need to explain 'sensitivity' in the text as had been done for 'specificity'. Members suggested the inclusion of 'precipitation' as one method for determining the highest concentration tested. There was also a request for suitable references to support the conclusion that the *in vitro* Comet assay detects gene mutations.

20. Regarding *in vivo* testing, it was agreed that the list detailing the specific questions addressed when undertaking *in vivo* tests was not exhaustive. It was agreed to remove the requirement for evidence of exposure in the in target tissues before a negative conclusion could be reached. This was because it was considered too difficult to do in all cases and systemic plasma exposure was normally considered sufficient as evidence of target tissue exposure. The Committee agreed that specific guidance to undertake kinetics studies in all cases was not appropriate. Members queried what the section on QSARs was attempting to achieve and whether this document was the right place to assess which models were suitable. Members also questioned whether it was appropriate to make distinctions between publicly available or commercial software. The Committee confirmed that QSAR approaches could be used where there were no other genotoxicity data on a compound. As this was a rapidly developing field, the COM agreed to consider QSARs further at a later date where it was hoped an expert in computational toxicology could attend.

21. Regarding the table on screening tests in the Annexes, members suggested some rearrangements of the tables. Also, that definitions of terms, such as, 'sensitivity' and 'specificity', would be helpful i.e. in a glossary

22. In relation to the 'executive summary', members agreed that this could be modified to include text taken from the summary of the *in vivo* section i.e. to indicate where the first *in vivo* study was negative, that there would only be a need for a second *in vivo* study, if there was a clear positive *in vitro* genotoxicity test with the chemical under consideration.

23. Members were asked to send any further editorial comments to the secretariat after the meeting. The Chair noted that a revised final draft document would be circulated by post. The document would be published in full on the publications section of the COM internet site. It would be subdivided into guidance statements covering each stage of the strategy on the 'Guidance statement section' of the COM internet site.

ITEM 6: FURTHER INFORMATION ON QSARs AS USED BY THE HPA SECRETARIAT (MUT/2011/10)

24. The COM briefly considered an initial assessment of the potential mutagenicity of chemicals arising from the test results of products that are intended to come into contact with drinking water. These chemicals were provided as illustrative examples of how QSAR approaches could potentially be applied to mutagenic hazard assessment.

25. Members had a number of reservations regarding the approach outlined in MUT/2011/04 and requested further information on the models that had been used. This was presented in MUT/2011/10 as an overview of a report by the EU Computation Toxicology Group of the Joint Research Centre (JRC), Ispra, Italy. The generic guidance from the COM has been that if there are no available genotoxicity data on a chemical, then at least publicly available QSAR modelling should be used to aid an initial assessment of potential mutagenicity.

26. The JRC developed a framework for assessing the usefulness of QSAR models based on published guidance adopted for use under REACH and also guidance published by the OECD. A review of publicly available and commercial models was conducted across all toxicology endpoints. Standardised documentation for demonstrating model validity was used (QSAR Model Reporting Format QMRF). The JRC undertook a survey of national regulatory bodies and international advisory organisations (in the field of food safety) to assess how QSAR approaches are used. There were responses from 38 organizations. Sixty percent of the organizations didn't use QSARs. In most cases the reason for this was lack of appropriate technical expertise. The most commonly used were: Derek; Multicase; OECD Toolbox; and Toxtree. The most highly developed models were for genotoxicity. The JRC undertook an assessment of commercial and publicly available genotoxicity models using a structurally diverse data set of 700 chemicals with genotoxicity test data. The correct identification of an Ames test data was highly reproducible. The JRC concluded that the use of pairwise combinations of the models increases the overall sensitivity and reduces the false negative rate. Further work could involve optimising data interpretation schemes.

27. CAESAR (Computer Assisted Evaluation of industrial chemicals According to Regulations) was developed as an EU funded project and is a statistically based model, developed using 4225 compounds from the Kazius-Bursi mutagenicity database. The correct classification rates for Ames test mutagenicity were 92.3% and 83.2% for the training and test sets, respectively. Toxtree was developed by the JRC as an EU project. It is a rule based approach using the Benigni-Bosa data base (an expansion of the Ashby super model) and the Tox Mic rulebase for the *in vivo*

micronucleus prediction. A combination of CAESAR and Toxtree had a false negative rate of 11%. The framework analyses for CAESAR and Toxtree were appended at Annex1.

28. A key part to any QSAR evaluation is a case-by-case judgement of the validity and the application of each QSAR model assessment. A number of guidance steps were outlined in MUT/2011/10. These briefly included the following; is the chemical within the scope of the model? is the defined endpoint suitable for the regulatory purpose?; how well does the chemical predict chemicals similar to the chemical of interest?; is the model prediction reasonable, taking into account other information? The secretariat proposed the use of the combination of CAESAR and Toxtree using a case-by-case approach where there is no genotoxicity available, noting the need for care in the interpretation of the results. Members were asked for their comments on the proposed approach.

29. One member had already sent written comments to the secretariat prior to the meeting, suggesting that chemical expertise or input from a chemist would be required to ensure an understanding of the relationship between mutagenic activity and chemical structure. It was considered that toxicological expertise was required to interpret the results of model predictions.

30. Members agreed that where negative predictions were made, this would provide some reassurance regarding the absence of mutagenic potential. It was also noted that Toxtree, being a rule based system, gave the reasons for its predictions. CEASAR gave the results for six other structural analogs, so some confidence could be obtained in positive predictions if the results for analogs were considered reasonable.

31. The representative from the FSA secretariat informed the committee that it had some experience in using QSARs. It was agreed that the FSA and HPA secretariats could liaise on further development of QSAR approaches.

32. The committee agreed that it would be useful for the COM to hear a presentation from a computational toxicologist with expertise in QSARs. Members agreed to provide the secretariat with suggested names of experts who could make a presentation to the COM on this topic.

ITEM 7: ANY OTHER BUSINESS

33. There was no other business.

ITEM 8: DATE OF NEXT MEETING

34. 20th October 2011

Item	Actions	Responsibility
Item 3: Testing of impurities	Revise and Provide interim guidance	Secretariat
Item 4: Fumagillin dicyclohexylamine	Finalise advice to VMD in form of statement	Secretariat
Item 5: Revision of COM Guidance: Consultation on a strategy for genotoxicity testing and mutagenic hazards of chemicals.	Finalise guidance document with one further postal circulation	Secretariat
Item 6: QSARs	Further information on QSARs. Arrange presentation to COM by computational toxicologist	Secretariat

