

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

Minutes of the meeting held on Tuesday 13 March 2001, at the Moller Centre, Cambridge.

1. Present

Chairman:	Professor Woods	
Members	Professor Aggett Professor Brown Professor Carthew Professor Chipman Dr Joffe Professor Renwick Professor Rowland Dr Rushton Ms Salfield Dr Smith Professor Strobel Dr Thomas Dr Tucker	
FSA Secretariat:	Dr Benford Mr Butler Dr Gott Mr Maycock Ms Mulholland Dr Shavila Dr Tahourdin Dr Thatcher	(Scientific) (Administrative)
DH Secretariat:	Mr Battershill	
Assessors:	Dr Efa Dr Cameron	PSD DETR
Also in attendance:	Dr Wadge Mr R Sinclair Dr Willets Dr Delic Mr Gem Dr Evans	(CST, FSA) (CST 4, FSA) Item 4 (CST 4, FSA) Item 4 (HSE) Item 4 (CD 'F', FSA) Item 6 (CST 5, FSA) Items 8&9

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Announcements

1. The Chairman informed members that the Secretariat had completed its review of the appointments due to expire on 31 March 2001 and with the approval of DH Ministers was about to begin recruitment of new members. The final stages were expected to be completed in the summer, when a formal announcement will be made. With the agreement of the Office of the Commissioner of Public Appointments (OCPA) all current appointments have been extended until 30 September 2001. The new appointments will be made from 1 October.
2. The Chairman reminded members that the Guidance issued by the OCPA sets a maximum length of 10 years service for members of advisory committees and that appointments were being reviewed on that basis. The Chairman added that as he had served as Chairman of the Committee since 1992, he would be retiring in March 2002 and that a successor was being sought.
3. The Chairman reminded those attending the meeting of the confidential nature of the proceedings and the content of some of the papers, and of the Agency's request that they declare any commercial or other interests that they have in any of the agenda items.

ITEM 1: APOLOGIES FOR ABSENCE

4. Apologies for absence were received from Professors Kimber and Timbrell and Dr Thomas.

ITEM 2: MINUTES OF THE MEETING HELD ON 6 FEBRUARY 2001: TOX/MIN/2001/01

5. The minutes of the meeting of 6 February were agreed subject to the following amendments:

Item 4– Paragraph 29 - delete last sentence and insert “Members asked whether the differences in sex ratio that had been reported in Seveso were supported by animal data. They were informed that data from animal

developmental studies did not show differences in the sex ratio of offspring. However, these studies were not specifically designed to address this issue.”

Item 4- Paragraph 32 first indent - delete “polymorphism of” and insert “animal strain differences in the”

Item 4- Paragraph 36: line 1 - after “sink” insert “for TCDD”;
line 2 – delete “endogenous” and insert “other”; delete 3rd sentence

Item 4 - Paragraph 37: line 3 – delete “effect” and insert “on the endpoint” after “dependent”

Item 4 - Paragraph 38: line 1 - insert “limited” before “evidence”;
line 2 - delete “is” and insert “may be”

Item 4 - Paragraph 41: line 4 - insert “not” before “considered”;
line 7 – delete “distinguish between responsive and non-responsive” and insert “show a good correlation with responsiveness in different”

Item 4 – Paragraph 45: line 3 – delete “effects” and insert “body burdens”;
line 7 - after “daily intake” insert “at lower doses”

Item 4 - Paragraph 48, line 4 – after “to” insert “limited data in”:
last line – at end of sentence insert “and the limited data from farmed and wild species did not indicate a difference.”

ITEM 3: MATTERS ARISING

6. The Chairman reported that the fifth draft PAH statement had been circulated to members as agreed, and that comments had been received. The Secretariat would consider these and prepare a revised draft for clearance by Chairman’s action. A final version would be sent to members.

7. Members were informed that, in the interests of openness, senior management at the Food Standards Agency had asked if draft minutes of expert committees and their working groups can be put onto the website after approval by the Chairman but before official approval by the full committee. This procedure is already followed by the ACNFP, whose minutes contain a rider that they are DRAFT and that “These minutes are subject to confirmation by the Committee at its next meeting.” The Chairman invited Members’ views on whether this would be acceptable for COT minutes.

8. Whilst members were in general agreement to this proposal, they expressed some concern that the DRAFT minutes may be taken out of context and portrayed as the final version. It was suggested the word 'DRAFT' be superimposed across every page. Subject to this, the Committee agreed to a trial for the remainder of the year.

ITEM 4: BISPHENOL A IN CANNED FOODS (TOX/2001/05)

9. Members were informed that bisphenol A (BPA) is manufactured by Dow Chemicals, Shell, Bayer and GE plastics, but it was not feasible to comprehensively record users of coated cans. No interests were declared.

10. The Committee's views were being sought on the health implications of the results of a survey by the Food Standards Agency (FSA) on (BPA) and bisphenol F (BPF) migration from can coatings into food. BPF was not detected in any of the samples and therefore the Committee's considerations focused on BPA. Members were also informed that a review of BPA was in progress under the Existing Substances Regulations (ESR). The February 2001 draft ESR Health Risk Assessment report, prepared by the UK Health and Safety Executive (HSE), had been provided to the Committee as background information to assist with the interpretation of the survey results. Dr Julian Delic of HSE attended for this item. Members agreed that the draft ESR report provided an accurate summary of the available data.

11. Members noted that most studies indicate a no-observed adverse effect level (NOAEL) of 50 mg/kg bw/day. However, a number of studies demonstrated effects at lower doses, but the functional significance of the effects was unclear and the studies were not designed to establish no-effect levels.

12. The available low dose studies on BPA were recently discussed at a National Toxicology Program (NTP) meeting in the USA. The NTP meeting noted that the data and conclusions of the low dose studies were valid; but the studies were not reproducible. Other well-conducted and reported studies had failed to reproduce the effects, using wider dose ranges (Cagen *et al* *Toxicological Sciences* 50: 36-44, 1999; Ashby *et al*. *Regulatory Toxicology and Pharmacology* 30: 156-166, 1999). However, there were variations in mouse strain and in the techniques used to carry out the investigations. The CF1 mice used in the studies by vom Saal and co-workers (Nagel *et al*.

Environmental Health Perspectives 105:70-76,1997; vom Saal *et al.* Toxicology and Industrial Health 14: 239-260, 1998) had been maintained as a closed colony and may have developed modified characteristics. However they are no longer available and therefore the results cannot be substantiated. Members considered that reproducibility is an important test of any scientific observation.

13. It was noted that the studies showing increased prostate weight in mice were not supported by histological evaluations and it was therefore not clear whether the effect had any functional significance. Prostate weight can be increased by infection, and histology is required to eliminate this possibility. In addition, the estimation of prostate size using three-dimensional reconstruction, has not been adequately validated and can result in measurements that are different to the actual size and shape in life. A more recent study (Gupta, Proceedings of the Society of Experimental Biology and Medicine 224: 61-68, 2000) added some support to the observation of increased prostate weight at low doses, although there were concerns about the adequacy of the study.

14. A very recent study by Soto's group showed early vaginal opening in mice treated with BPA at a very low dose and at a high dose but not in between, at five intermediate doses (Markey *et al.* Environmental Health Perspectives 109: 55-60, 2001).

15. The Committee noted that other compounds which show endocrine modulating activity at low doses also display developmental toxicity at higher doses. In contrast, BPA has not shown developmental toxicity, even at doses causing maternal toxicity, in well-conducted multi-generation studies. Rats are thought to be more sensitive to the effects of endocrine modulators than humans. The higher level of circulating oestradiol in humans compared to rodents during pregnancy could mean that small changes in total oestradiol-like activity brought about by endocrine modulators may be less relevant in humans.

16. Members considered that the effects on reproductive organs reported at low doses represented very sensitive intermediate biomarkers that indicate the need for further studies, but cannot be defined as "adverse". Overall the Committee concluded that it is not appropriate, at this time, to base human health risk assessment on these effects.

17. The BPA and BPF survey commissioned by the Food Standards Agency covered a range of canned foods (vegetables, beverages, soup, fish in aqueous media, desserts, fruit, pasta, meat products and infant formulae). Levels of BPA varied in different samples, with considerably higher levels in one type of canned ham. The Secretariat was asked and agreed to check whether the infant formulae tested in the survey were analysed in powder form or after reconstitution. It was noted that the samples would have been heated during the processing and sterilisation procedures, but BPA is known to be stable on heating.

18. The Committee considered a draft statement. Members made detailed recommendations for revision of the statement, in order to clarify the basis for their conclusions and the uncertainties that exist in the scientific understanding of potential endocrine effects of BPA. On present evidence the Committee concluded that the levels of BPA identified in canned foods analysed in the FSA survey are unlikely to be of concern to health. The Committee further agreed that it would be prudent to review this toxicological advice as further scientific evidence on possible low-dose effects of endocrine modulating substances unfolds.

19. It was agreed that the draft statement should be amended and circulated to Members for comment and clearance by Chairman's action.

ITEM 5: CONSIDERATION OF THE TDI FOR DIOXINS AND DIOXIN-LIKE PCBs

Previous COT considerations. (TOX/2001/10)

20. The Secretariat updated members on progress in compiling the requested structured summary of the exposure data from the human epidemiology studies. This summary would be discussed at the next meeting. Members were informed that, following the request by this Committee, the Committee on Carcinogenicity (COC) had agreed to review the US-EPA chapters on cancer and the actual risk assessment procedure adopted by the US-EPA. These will be discussed at the COC meeting on 22 March together with new information published since COC last considered the carcinogenicity of dioxins, following the IARC evaluation in 1998. The Secretariat will inform members of the progress of COC's deliberations and their conclusions at the next meeting.

21. Following the discussions at the February meeting, the Secretariat had recognised that providing members with a compilation of previous COT statements on dioxins and polychlorinated biphenyls (PCBs) (TOX/2001/10) would be beneficial in the development of the statement. Members noted that there remained an absence of data on dietary dioxin exposure in infants between 6 and 18 months. Members were informed that consideration was being given to a proposal for a survey on infant formula and follow-on foods.

Studies on endometriosis (TOX/2001/08)

22. Members were reminded that the recent WHO consultation and SCF opinion on dioxins and dioxin-like PCBs had considered endometriosis to be one of the most sensitive effects in experimental animals. Members had previously considered a study by Rier et al. (Fundamentals of Applied Toxicology 21: 433-441, 1993) which reported on endometriosis in monkeys 10 years after completion of a study in which TCDD (98% pure) was administered in the diet for a period of about 4 years. A recently published paper follows up the same groups of monkeys, reporting the incidence of endometriosis and serum levels of dioxins and PCB congeners, 13 years after completion of the dietary study (Rier et al., Toxicological Sciences 59: 147-159, 2001).

23. The recent study indicated that TCDD exposure and elevated serum TCDD concentration correlated with increased serum concentrations of triglycerides and a number of polyhalogenated hydrocarbons. Animals with elevated serum 3,3',4,4'-tetrachlorobiphenyl (TCB), 3,3',4,4',5-pentachlorobiphenyl (PnCB) and total TEQ exhibited a high prevalence of endometriosis. The severity of endometriosis correlated with serum TCB levels, but not TCDD levels. Elevated serum concentrations of specific PCB congeners were also found in animals housed in the same facility, which had not been treated with TCDD. The authors could not account for the source of PCB exposure, but concluded that PCBs may be involved in the pathogenesis of endometriosis in the rhesus monkey.

24. Members noted that there were a number of aspects of this observational study, which undermined confidence in these results and the earlier findings. These included the reported correlation between measured levels of TCDD and PCBs. Members suggested that this was indicative of subsequent exposure since the PCBs have shorter half-lives than TCDD and over 8 TCDD half-lives had elapsed since exposure ceased. Members noted

that animals involved in a study in which lead was administered were also reported to have PCB levels and endometriosis, however levels of lead did not appear to have been ascertained in the TCDD exposed animals.

25. Members concluded that it was not possible to draw reliable conclusions on endometriosis following dioxin exposure given the uncertainties arising from the reported serum levels and source of PCBs. Members agreed that, on the basis of these new data, endometriosis did not appear to be a key end-point in the animal studies. The data from the Seveso Women's Health Study on endometriosis would be crucial to further consideration of this end-point. The Secretariat agreed to approach the authors seeking further information regarding progress of publication of the results of this study. Members were informed that an expert in endometriosis had agreed to assist the Committee in evaluation of the human data.

26. Members noted that it was necessary to clarify whether PCB levels or only TCDD levels were determined in the study of behavioural effects in offspring from monkeys. Members noted they might need to reconsider this study in light of this clarification.

Outline of draft statement on dioxins (TOX/2001/14)

27. The Secretariat tabled a proposed outline for the draft statement. Members agreed that it would be necessary to produce a longer statement than previously, in order to set out clearly the Committee's interpretation of the data. Members suggested that the comparison of body burdens should be incorporated into the overall evaluation. Following this change members were content with the outline.

ITEM 6: PAHs IN SHELLFISH (TOX /2001/12)

28. No interests were declared

29. Members were reminded that the Committee considered a draft CEFAS report at the December 2000 meeting and concluded that, in order to support risk assessment, additional information was needed on consumption patterns for the general population and for potential high level consumers residing around coastal resorts.

30. Information on consumption patterns and estimations of intake of polycyclic aromatic hydrocarbons (PAHs) from bivalve molluscs were presented in TOX/2001/12. No data were available on whether PAH concentrations remained constant over time. Concentrations were therefore assumed to be representative except in the single example of a high level thought to be associated with a specific pollution incident.

31. Members were also provided with a JECFA review of benzo(a)pyrene (BaP) (WHO Food Additive Series 28: 301-363, 1991), noting the several orders of magnitude difference between human dietary intake and the level of BaP needed to induce tumours in experimental animals. Members were asked to consider whether the estimated intakes are sufficiently low as to not be a cause for concern, either for the general population or for consumers in coastal regions.

32. The data indicated that intakes of PAHs from bivalve molluscs are likely to be within the normal range for the general population, as assessed in Dennis *et al.* (Food Chemical Toxicology 21: 569-574, 1983), but may result in a higher level of intake for high level consumers in coastal regions with persistent PAH contamination.

33. The Committee was reminded that the COC considered that there were no thresholds for the carcinogenicity of BaP. The NOAELs that had been determined from animal studies were dependent on group size. In view of the apparent lack of epidemiological data on the consumers of shellfish and incidence of cancer, Members noted that it may be appropriate to commission "Small Area " studies. It was suggested that dietary intake of PAHs could be compared to exposure from tobacco smoke, however such a comparison would be complicated by the different routes of exposure, and the presence of other carcinogens in tobacco smoke.

34. Members noted that in this CEFAS survey, samples had been collected on the basis of microbiological concern. Thus, they were not necessarily representative of worst case situations in respect of PAH levels.

35. It was noted that the data indicated no major difference in consumption between coastal areas and the general population, but the data were based upon too few consumers to be reliable. It was also noted that it was not possible to determine the proportion of total exposure to PAHs from oysters

and other bivalve molluscs as total diet studies of PAHs have grouped shellfish with other fish and fish products.

36. In view of the limitations of the database on consumption patterns for consumers living around coastal areas, and given that the total exposures to individual PAHs were estimated from old data, Members agreed that it was not possible to comment on the health implications of the levels determined in the CEFAS study.

37. The Committee was informed that a Total Diet Study on PAHs is due to be commissioned later this year. The Secretariat agreed to give consideration to the design and sampling methodology of future surveys in order to assist with the Committee's interpretation of relevance to public health.

ITEM 7: PROPOSED JOINT MEETING OF COT/COC/COM (TOX/2 001/09)

38. In discussion of the report by Sir Robert May on the Review of Risk Procedures used by the Government's Advisory Committees dealing with Food Safety, members of the COT, COC and COM had identified a need for closer working of the three committees. Members had acknowledged that the expert committees could benefit from a greater degree of cross-fertilisation and welcomed a suggestion that they might meet other committees, at least on an occasional basis. Toxicogenomics and proteomics had been identified as a suitable topic for a joint meeting between the three committees, because it is a rapidly growing area with important implications for risk assessment and will have implications in the regulatory arena.

39. Members were provided with a number of recent publications, which discussed the proposed topic under consideration and highlighted some of the potential advantages and drawbacks of these allied fields. The Secretariat proposed that a joint meeting of the COT, COC and COM could be held in the autumn of 2001.

40. Members considered the proposal timely as this type of technology was already being used extensively, but its applications in risk assessment were still unclear. Members agreed that the emphasis should be on data analysis, not the technology. However a basic introductory overview would be required for the benefit of those not familiar with the technology. Members noted the need to consider how the data generated compare with the functional

endpoints assessed in conventional toxicity studies, and how the results could be integrated in the risk assessment process.

41. Other points raised included the importance of involving experts in bioinformatics, and the possibility of taking a strategic view of how the new technologies could be used in population studies as well as in toxicity studies.

42. Members noted that the International Life Sciences Institute (ILSI) was currently involved in a multi-centre initiative in this area, and may be able to help in identifying potential speakers.

43. Members also discussed the possible meeting format, and how this could be structured to integrate the three committees. One possibility might be to have a number of separate discussion groups. It was anticipated that other government departments would wish to send observers to the meeting. Consideration would also be given to inviting wider participation in the interests of openness.

44. Members agreed to advise the Secretariat of suitable speakers and the joint secretariats will then establish the date and venue. The Committee was informed that the secretariats aim to produce a publication from the meeting, although the format has yet to be considered.

ITEM 8: a) REVIEW OF ENZYME SUBMISSION: AMANO 90 (TOX/2001/11)

45. Members were reminded that this enzyme submission was considered in June 2000. The Committee had requested additional information on the assay methodology and inactivation of the enzyme during baking. The company had submitted new information intended to answer these points.

46. Members considered that the amount of detail in the new information was limited and not all the requested information had been provided. Members noted that the new information probably demonstrated that inactivation of the enzyme was occurring. However, Members remained concerned that there was still no information on the limit of detection, assays did not appear to have been conducted in duplicate and there was no explanation for the apparent increased sensitivity of the assay. Members were unable to recommend full approval of the hemicellulase enzyme Amano 90 in the absence of details of the assay and performance of the assay on duplicate samples, both of which the Committee had previously requested. The Secretariat will inform the

company of the Committee's conclusion and request more detailed information.

b) XYLANASE PREPARATION FROM GM *Aspergillus niger* (TOX/2001/13)

47. Professor Aggett declared a non-specific non-personal interest.

48. Members were informed that this was a new submission of a xylanase preparation from a genetically modified *Aspergillus niger* strain, and that the Advisory Committee on Novel Foods and Processes (ACNFP) had already considered the submission.

49. Members noted that the toxicity studies were well conducted and no dose-related findings were reported. There was a very large margin of safety between the no observed adverse effect level in the toxicity studies and the estimated intake values at the maximum application rates. Members noted that there were difficulties in demonstrating the lack of allergenicity requested by ACNFP and that methodology had not been specified in the Framework for the Assessment of Enzymes. However, several experimental methods are available to provide some evidence of absence of allergenicity.

50. Members agreed temporary approval for the use of the powdered and micro-granulated products in the formulation of enzyme preparations used in the production of baked goods produced using yeast, including bread and biscuits such as wafers and crackers. Approval is for 1 year in the first instance and it was not considered appropriate to specify a maximum application rate. The company will be requested to provide the following additional information (which included the points raised by the ACNFP);

- a) Testing for mycotoxins and other contaminants should ideally be conducted at regular intervals on representative batches of the xylanase preparation itself. Testing on composite products which contain the preparation would be an acceptable alternative, provided that at least one such test is carried out on each batch of the preparation;
- b) TOS and E values should be provided for at least three equivalent batches of the unstandardised, spray-dried product.
- c) All final enzyme preparations derived from the powdered and micro-granulated products should comply with the attached specification and that approval should be sought for any changes to the specification (e.g.

to allow the use of different strains of *Aspergillus niger* derived from XYL-2).

- d) Laboratory data should be provided to demonstrate that there is no residual enzyme activity/allergenic problem in the final food.
- e) Data on the genetic stability are required to support the claims made within the dossier on genetic and production stability of *Aspergillus niger* strain).

ITEM 9: SUBMISSION OF ADDITIONAL DATA ON ALITAME (TOX/2001/06)

51. Professor Renwick declared a personal, specific interest. Professor Rowland declared a non-personal, specific interest. Both left the meeting.

52. Members were reminded that alitame is an intense sweetener that had been discussed by the Committee on several occasions previously. In October 2000, Members had discussed a new study of alitame in diabetics. The statistical analysis of the study had been questioned and the results of the ANOVA requested. Concern was also expressed that the study had not attempted to investigate the possibility that alitame could induce drug metabolising enzymes in the liver.

53. The Company considered that the statistical analysis indicated that alitame was well-tolerated in diabetics with regard to cardiovascular effects. Members agreed that this study showed that alitame had no effects over the one year period of the study. However, long-term cardiovascular effects could not be excluded, though the relevant risk markers had not been affected by alitame.

54. Members noted that the company had provided analytical data on clinical chemistry parameters. This had been included in the report discussed previously and did not address the issue of enzyme induction. The Committee was informed that there were no biological samples available for further analysis. The ADI had been established on the basis of increased liver weights in dogs, in a study in which enzyme induction was also seen at higher doses, and it would be helpful to know the sensitivity of human liver enzyme induction compared with that in dogs, though this would require a specially designed study.

55. Overall, Members considered that the new data did not warrant a review of the ADI for alitame.

ITEM 10: DRAFT ANNUAL REPORT (TOX/2001/07)

56. The Committee was informed that the COT/COC/COM Secretariats had agreed a new format for the Annual Report. The main change concerned the Committee's statements, which were now contained in an Annex rather than in the text. Members were invited to comment on the text, having been reminded that the statements had previously been agreed and could not be altered. Although most had already done so, Members were also reminded to inform the Secretariat of any changes to their interests.

57. The introduction to the report referred to the Nolan principles for appointments, and it was noted that this should make clear that it relates to future appointments, and not the current membership.

58. Members were asked to forward any other comments to the Secretariat.

ITEM 11: ANY OTHER BUSINESS

59. There was no other business.

ITEM 12: DATE OF NEXT MEETING

60. The next meeting will take place on 1 May 2001. Members were informed that, subject to confirmation, Sir John Krebs, Chairman of the FSA, would attend for part of the meeting.