

US EPA ARCHIVE DOCUMENT

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June x, 2006

EPA-HSRB-06-01

George Gray, Ph.D.
Science Advisor
Office of the Science Advisor
1200 Pennsylvania Avenue, NW
Washington, DC 20460

Subject: April 4-6, 2006 Meeting EPA Human Studies Review Board Report

Dear Dr. Gray:

The United States Environmental Protection Agency (EPA or Agency) requested the Human Studies Review Board (HSRB) to provide advice on Agency scientific and ethics reviews of completed human studies concerning the following pesticide active ingredients: aldicarb, amitraz, azinphos-methyl, dichlorovos (DDVP), ethephon, methomyl, oxamyl, and sodium cyanide. The studies reviewed included both studies on which the Agency proposes to rely in actions under the pesticide laws and studies that the Agency has decided not to use in its risk assessments, either for ethical or scientific reasons. The enclosed HSRB report addresses the Board's response to EPA charge questions for the Board's consideration at its April 4-6, 2006 meeting.

The HSRB was extremely impressed with the high quality scientific and ethical review brought before the Board. A summary of the Board's conclusions on the scientific and ethical considerations of the human toxicity studies for the eight pesticides under review are provided below.

Aldicarb

Scientific considerations

- The cholinesterase data from the aldicarb human study were reliable for use in the aldicarb single chemical, aggregate risk assessment.
- The cholinesterase data from the aldicarb human toxicity study were reliable for use in the cumulative risk assessment for N-methyl carbamates.

1 Ethical considerations

- 2 • The aldicarb human toxicity study failed to fully meet the specific ethical standards
- 3 prevalent at the time the research was conducted.
- 4 • There was no clear or convincing evidence that the research was fundamentally
- 5 unethical--intended to seriously harm participants or that informed consent was not
- 6 obtained.
- 7 • There was no clear and convincing evidence of significant deficiencies in the ethical
- 8 procedures that could have resulted in serious harm (based on the knowledge available at
- 9 the time the study was conducted), nor that information provided to participants seriously
- 10 impaired their informed consent.

11
12 Methomyl

13
14 Scientific Considerations

- 15 • The methomyl human study could be appropriately applied to the inter-species risk factor
- 16 for methomyl and for use in cumulative risk assessment of N-methyl carbamates.

17
18 Ethical Considerations

- 19 • The methomyl human toxicity study failed to fully meet the specific ethical standards
- 20 prevalent at the time the research was conducted.
- 21 • There was no clear and convincing evidence that the research was fundamentally
- 22 unethical--intended to seriously harm participants or that informed consent was not
- 23 obtained.
- 24 • There was no clear and convincing evidence of significant deficiencies in the ethical
- 25 procedures could have resulted in serious harm (based on the knowledge available at the
- 26 time the study was conducted), nor that information provided to participants seriously
- 27 impaired their informed consent.

28
29 Oxamyl

30
31 Scientific Considerations

- 32 • Although the Board had some questions about the Agency's conclusions regarding lack
- 33 of sex difference and of the difference between brain and RBC ChE based on only one
- 34 species, the Board supported the Agency's conclusion that there were no study
- 35 deficiencies identified that would have affected the outcome of conclusions of this study.
- 36
- 37 • Considering the high quality of the design and the conduct of the study, the Board agreed
- 38 that this intentional human dosing study of oxamyl was sufficiently robust to be used for
- 39 reducing the 10x inter-species (i.e. animal to human) uncertainty factor in the cumulative
- 40 risk assessment for the N- methyl carbamates.

41
42 Ethical Considerations

- 43 • The oxamyl human toxicity study failed to fully meet the specific ethical standards
- 44 prevalent at the time the research was conducted.

- 1 • There was no clear or convincing evidence that the research was fundamentally
2 unethical--intended to seriously harm participants or that informed consent was not
3 obtained.
- 4
- 5 • There was no clear and convincing evidence of significant deficiencies in the ethical
6 procedures that could have resulted in serious harm (based on the knowledge available at
7 the time the study was conducted) nor that information provided to participants seriously
8 impaired their informed consent.
- 9

10 Azinphos-Methyl

11 Scientific Considerations

- 12 • Data from the 28-day repeat oral dose study of azinphos methyl should not be used in
13 developing a point of departure for extrapolation of risk to workers exposed to azinphos-
14 methyl via the dermal and inhalation routes.
- 15
- 16 • Data from the 28-day repeat oral dose study of azinphos-methyl cannot be used to inform
17 the inter-species factor in the cumulative risk assessment.
- 18

19 Ethical Considerations

- 20 • The AZM human toxicity study failed to fully meet the specific ethical standards
21 prevalent at the time the research was conducted.
- 22
- 23 • There was no clear and convincing evidence that the research was fundamentally
24 unethical--intended to seriously harm participants or that informed consent was not
25 obtained.
- 26
- 27 • There was no clear and convincing evidence of significant deficiencies in the ethical
28 procedures that could have resulted in serious harm (based on the knowledge available at
29 the time the study was conducted) nor that information provided to participants seriously
30 impaired their informed consent.
- 31

32 DDVP

33 Scientific Considerations

- 34 • The DDVP repeat dose human toxicity study was sufficiently robust for developing a
35 point of departure for estimating dermal, incidental oral, and inhalation risk from
36 exposure to DDVP in the single chemical risk assessment.
- 37
- 38 • The DDVP repeat-dose human toxicity study should not be used to support reducing the
39 default 10X inter-species factor in the cumulative risk assessment of the organophosphate
40 pesticides.
- 41
- 42 • The HSRB concluded that the other DDVP human toxicity studies available for the
43 Board's consideration should not be used for determining a reduction in the 10X
44
- 45

1 uncertainty factor to derive reference dose values for DDVP based on animal toxicity
2 endpoints.

3
4 Ethical Considerations

- 5 • The DDVP repeat dose oral human toxicity study failed to fully meet the specific ethical
6 standards prevalent at the time the research was conducted.
- 7
8 • There was no clear and convincing evidence that the research was fundamentally
9 unethical--intended to seriously harm participants or that informed consent was not
10 obtained.
- 11
12 • There was no clear and convincing evidence of significant deficiencies in the ethical
13 procedures that could have resulted in serious harm (based on the knowledge available at
14 the time the study was conducted) nor that information provided to participants seriously
15 impaired their informed consent.

16
17 Ethephon

18
19 Scientific Considerations

- 20 • The Board concluded that the scientific quality of both the ethephon 28 day oral toxicity
21 study and the ethephon 16 day human oral toxicity study was not adequate on its own.
22 The 16 day study can be used to inform the ethephon 28 day human oral toxicity study.
23 The Board approved the 28 day study for use in EPA risk assessments, emphasizing that
24 the dose level administered is almost certainly not the lowest dose at which adverse
25 effects are likely to be observed. However, its use in lieu of the animal studies will result
26 in greater protection for exposed human populations.

27
28 Ethical Considerations

- 29 • Both the 28 and 16 day oral human toxicity studies failed to fully meet the specific
30 ethical standards prevalent at the time the research was conducted.
- 31
32 • There was no clear and convincing evidence that the research was fundamentally
33 unethical--intended to seriously harm participants or that informed consent was not
34 obtained.
- 35
36 • There was no clear and convincing evidence of significant deficiencies in the ethical
37 procedures that could have resulted in serious harm (based on the knowledge available at
38 the time the study was conducted), nor that information provided to participants seriously
39 impaired their informed consent.

40
41 Hydrogen Cyanide

42
43 Scientific Considerations

- 44 • Data from the amygdalin trial could be used for establishing a point of departure in the
45 acute dietary risk assessment for hydrogen cyanide.

1 Ethical Considerations

- 2 • The hydrogen cyanide human oral toxicity study appeared to meet the specific ethical
3 standards prevalent at the time the research was conducted.
4
5 • There was no evidence that the research was fundamentally unethical--intended to
6 seriously harm participants or that informed consent was not obtained.
7
8 • There was no evidence of significant deficiencies in the ethical procedures that could
9 have resulted in serious harm (based on the knowledge available at the time the study was
10 conducted) nor that information provided to participants seriously impaired their
11 informed consent.
12

13 Amitraz

14
15 Scientific Considerations

- 16 • The results from the single oral dose study are informed by the human metabolism study
17 such that the single oral dose study is appropriate for developing a point of departure for
18 acute and chronic dietary risk and short-term oral exposure.
19
20 • The combined results from the single oral dose study and the human metabolism study
21 were not appropriate for developing a point of departure for chronic dietary risk, short-
22 term oral exposure, or inhalation risk.
23
24 • The majority of the Board concluded that the human dermal study was not appropriate
25 for developing a point of departure for dermal exposures of various durations.
26

27 Ethical Considerations

- 28 • The amitraz acute oral and dermal human toxicity studies failed to fully meet the specific
29 ethical standards prevalent at the time the research was conducted.
30
31 • There was no clear and convincing evidence that the research was fundamentally
32 unethical--intended to seriously harm participants or that informed consent was not
33 obtained.
34
35 • There was no clear and convincing evidence of significant deficiencies in the ethical
36 procedures that could have resulted in serious harm (based on the knowledge available at
37 the time the study was conducted), nor that information provided to participants seriously
38 impaired their informed consent.
39

40 In conclusion, the EPA HSRB appreciated the opportunity to advise the Agency on the
41 scientific and ethical aspects of human subjects and looks forward to future opportunities to
42 continue advising the Agency in this endeavor.
43
44
45
46

1 Sincerely,

2

3

4

5 Celia Fisher, Ph.D. Chair

6 EPA Human Studies Review Board

1 **NOTICE**

2
3 This report has been written as part of the activities of the EPA Human Studies Review
4 Board, a Federal advisory committee providing advice, information and recommendations on
5 issues related to scientific and ethical aspects of human subjects research. This report has not
6 been reviewed for approval by the Agency and, hence, the contents of this report do not
7 necessarily represent the view and policies of the Environmental Protection Agency, nor of other
8 agencies in the Executive Branch of the Federal government, nor does mention of trade names or
9 commercial product constitute a recommendation for use. Further information about the EPA
10 Human Studies Review Board can be obtained from its website at <http://www.epa.gov/osa/hsrb/>.
11 Interested persons are invited to contact Paul Lewis, Designated Federal Officer, via e-mail at
12 lewis.paul@epa.gov.

13
14 In preparing this document, the Board carefully considered all information provided and
15 presented by the Agency presenters, as well as information presented by public commenters.
16 This document addresses the information provided and presented within the structure of the
17 charge by the Agency.

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21 Agency, Washington, DC

22
23 * Not in attendance at April 4-6, 2006 Public Meeting

1	TABLE OF CONTENTS	
2		
3	INTRODUCTION	11
4	REVIEW PROCESS	12
5	CHARGE TO THE BOARD AND BOARD RESPONSE	13
6	1. ALDICARB.....	13
7	2. METHOMYL.....	19
8	3. OXAMYL.....	25
9	4. AZINPHOS-METHYL.....	32
10	5. DDVP.....	38
11	6. ETHEPHON.....	47
12	7. HYDROGEN CYANIDE.....	54
13	8. AMITRAZ.....	58

1 INTRODUCTION

2
3 On February 6, 2006 (71 FR 24, 6137), EPA published its final rule for protection of
4 subjects in human research. This new rule included creating an independent Human Studies
5 Review Board (HSRB) to provide advice, information and recommendations related to the
6 scientific and ethical issues of such research.

7
8 The Food Quality Protection Act (1996) mandated that the Agency reassess pesticide
9 tolerances (i.e., the legal limit of pesticides in food) by August, 2006. The Agency is in the final
10 stages of reassessing all tolerances and has elected for the inaugural April 4-6, 2006 HSRB
11 meeting to focus the first meeting of the Board on those pesticides which are subject to the
12 August, 2006 statutory deadline for tolerance reassessment and which have a human toxicity
13 study being considered for incorporation in quantitative human health risk assessment. The
14 HSRB reviewed toxicity studies involving intentional exposure of human subjects to eight
15 pesticide active ingredients: aldicarb, amitraz, azinphos-methyl (AZM), ethephon, dichlorvos
16 (DDVP), methomyl, oxamyl, and sodium cyanide. Three of these pesticides are members of the
17 *N*-methyl carbamate (NMC) common mechanism group (aldicarb, methomyl, oxamyl); two are
18 members of the organophosphorus pesticides (OP) common mechanism group (AZM, DDVP).
19 The three remaining pesticides (amitraz, ethephon, sodium cyanide) each have toxicity profiles
20 that differ from the other chemicals.

21
22 The Agency prepared a variety of documents which provided background information.
23 These included the proposed and final Human Studies Rule, report from the National Academy
24 of Sciences regarding human studies, EPA, Office of Pesticide Programs' (OPP's) policy
25 regarding the use of cholinesterase data in human health risk assessments, and a slide
26 presentation providing a broad overview of the OPP's human health risk assessments. The
27 Agency used the framework provided in Emanuel (2000) as the basis for its ethics reviews. The
28 Agency described the approach for performing scientific review of human studies and for
29 incorporating human toxicity data into risk assessment.

30
31 For each of the human studies under consideration, the Agency provided the complete
32 study report as submitted to the Agency. For each chemical, the Agency developed a review of
33 the ethical conduct of the study. Each ethics review identified any deficiencies noted in the
34 conduct of the specific study compared to both current ethical standards and the ethical standards
35 prevailing at the time the research was performed. The Agency has intentionally deferred
36 making a final determination of whether an individual study satisfied the ethical standards for
37 acceptability in 40 CFR sections 26.1704 – 26.1706, pending the advice of the Board.

38
39 For most studies, the Agency develops documents, called Data Evaluation Records
40 (DERs), containing a scientific review of the study. DERs contain summaries of the study
41 design, methods and results, describe potential deficiencies, and provide conclusions about the
42 usefulness of the study in risk assessment. In addition to the DERs, the Agency prepared for
43 each chemical a "Weight of Evidence" (WOE) memorandum that discusses the differences and
44 similarities between the human and animal toxic responses to each chemical and characterizes
45 the usefulness of the human toxicity studies for human health risk assessment. The WOE memos

1 express the Agency's most current scientific conclusions on which the Agency solicited
2 comments from the Board.

3
4 The HSRB reviewed both studies on which the Agency proposes to rely on actions under
5 the pesticide laws and studies that the Agency has decided not to use in its risk assessments,
6 either for scientific reasons or because they do not meet the standards in EPA's final human
7 studies rule. The Agency asked the HSRB to advise the Agency on a range of scientific and
8 ethics issues regarding how the studies should be assessed against the provisions in 40 CFR
9 sections 26.1701 – 26.1704 of EPA's final human studies rule. The Board anticipates having a
10 series of meetings to review pesticide human studies relevant to pending re-registration,
11 tolerance reassessment and new registration decisions, in accordance with the provisions of the
12 final rule. This report transmits the HSRB's comments and recommendations from its inaugural
13 April 4-6, 2006 meeting.

14 **REVIEW PROCESS**

15
16 On April 4-6, 2006 the Board had a public face-to-face meeting in Arlington, Virginia.
17 Advance notice of the meeting was published in the Federal Register "Human Studies Review
18 Board: Notice of Public Meeting and Proposed Candidates for Membership to the Board (71 FR
19 46, 12194). At the public meeting, following welcoming remarks from Agency officials, Celia
20 B. Fisher, HRSB Chair, proposed a set of scientific and ethics criteria consistent with the
21 language of 71 FR 24, 6137 to guide Board evaluation of each protocol. The Chair's scientific
22 criteria asked the Board to consider the following two questions: (1) Did the research design and
23 implementation meet scientific standards and (2) Did the data generated by the protocol have
24 implications for the Agency's Weight of the Evidence (WOE) review and when applicable
25 aspects of the risk assessment? The Chair's ethics criteria asked the Board to consider three
26 questions: (1) Did the study fail to fully meet specific ethical standards prevalent at the time the
27 research was conducted; (2) Was the conduct of the study *fundamentally unethical* (i.e.,
28 specifically was there clear and convincing evidence that the research was intended to seriously
29 harm participants or failed to obtain informed consent); and (3) Was the conduct of the study
30 *significantly deficient* relative to the ethical standards prevailing at the time (i.e., was there clear
31 and convincing evidence that identified deficiencies could have resulted in serious harm based
32 on knowledge available at the time the study was conducted *or* the information provided to
33 participants could seriously impair informed consent. The Board then heard presentations from
34 EPA on the following topics: (1) introduction (summary of EPA protections for subjects of
35 human research, science and ethics considerations of human subjects research); (2) carbamate
36 pesticides (science and ethics of aldicarb, methomyl and oxamyl human studies); (3)
37 organophosphate pesticides (science and ethics of azinphos methyl and DDVP human studies);
38 (4) other pesticides (science and ethics of ethephon, sodium cyanide and amitraz human studies).
39 The Board also heard oral public comments from the following individuals:

40 Carbamate pesticides

41 Mr. Angus Cameron representing BCG Europe

42 Neil Carmichael, Ph.D. representing Bayer Crop Science

43 Jennifer Sass, Ph.D. representing Natural Resources Defense Council

1 Organophosphate Pesticides

2 Monty Eberhart, Ph.D. and Mr. Dan Van Geothern representing Bayer Crop Science and
3 Mahktesheim Agan
4 Robert Levine, Ph.D. representing Amvac
5 Mr. Ian Chart representing Amvac
6 Laura Plunkett, Ph.D. representing Amvac
7 Thomas Starr, Ph.D. representing Amvac
8 Jennifer Sass, Ph.D. representing Natural Resources Defense Council
9 Ms. Shelley Davis representing the Farmworker Justice Fund

10
11 Other Pesticides

12 Neil Carmichael, Ph.D. representing Bayer Crop Science

13
14 Following Agency presentations and public comments, the Board deliberated on the
15 charge questions. For their deliberations, the Board considered the materials presented at the
16 meeting, written public comments and Agency background documents on each individual
17 pesticide (i.e., pesticide human study, Agency data evaluation record (DER) of the pesticide
18 human study, weight of evidence review, risk assessment and ethics review).
19

20 **CHARGE TO THE BOARD AND BOARD RESPONSE**

21
22 **1. Aldicarb**

23
24 **Charge to the Board**

25
26 Aldicarb is a *N*-methyl carbamate (NMC) pesticide whose primary toxic effect is
27 neurotoxicity caused by the inhibition of the enzyme acetylcholinesterase, via carbamylation
28 followed by rapid recovery. Aldicarb can, at sufficiently high doses, lead to a variety of clinical
29 signs. The Agency is conducting an acute, aggregate (single chemical, multi-route) risk
30 assessment of aldicarb. In addition, aldicarb is a member of the *N*-methyl carbamate common
31 mechanism group and is thus included in the cumulative (multi-chemical, multi-route) risk
32 assessment for the NMCs.
33

34 **1.1 Scientific considerations**

35
36 The Agency's "Weight of the Evidence" (WOE) document and Data Evaluation Records
37 (DERs) for aldicarb describe the study design and results of the aldicarb acute oral, human
38 toxicity study. The WOE document also discusses the Agency's conclusions regarding the
39 usefulness of the human study in the acute, aggregate, single chemical risk assessment and in the
40 cumulative risk assessment for the NMCs. Regarding the aldicarb human study, the Agency has
41 concluded that the study is sufficiently robust for reducing the inter-species (i.e., animal to
42 human) uncertainty factor in the aggregate and the cumulative risk assessments.
43

1 The Board was asked to comment on the scientific evidence that supports whether the aldicarb
2 human study is sufficiently robust for reducing the inter-species (i.e., animal to human)
3 uncertainty factor in:
4 a. single chemical, aggregate risk assessment and
5 b. cumulative risk assessment.

7 **Board Response to the Charge**

9 Brief Overview of the Study

10
11 The study (Wyld et al. 1992) consisted of a double blind, placebo-controlled, single oral
12 dose of aldicarb in orange juice, taken during the course of a meal. A variety of both subjective
13 and objective observations were accumulated, including red blood cell (RBC) cholinesterase
14 activities, blood pressure, and respiratory parameters among others. Several dose levels were
15 investigated and several time points were also investigated.

17 Critique of Study

18
19 The strengths of the study were: it was designed with multiple doses so a dose-response
20 relationship could be studied; it was double blind; a large number of parameters that are relevant
21 to anticholinesterase were studied (including both subjective data such as headache and several
22 objective physiological measures such as blood pressure, pulse, pulmonary function and saliva
23 production); there were exclusion criteria; there were frequent measurements of cholinesterase
24 over a 1 day period in addition to pre-treatment measurements; the observations of clinical signs
25 were made by trained observers; both sexes were studied (although there were fewer doses and
26 individuals in the female group); and the cholinesterase depression was both dose- and time-
27 dependent in both males and females.

28
29 The weaknesses of the study were: there were typically only 4-8 individuals per dose;
30 there were fewer female subjects and they were only tested at the middle dose levels; there were
31 a large number of reported signs across all groups, including the placebo, making interpretation
32 difficult; the cholinesterase methodology may have not assayed all of the cholinesterase in
33 RBCs; and the first time point of cholinesterase measurement may have been after the time of
34 peak cholinesterase inhibition.

35
36 The Board also noted that, with respect to statistical analysis, the study was likely to be
37 under-powered. With the large number of endpoints and the relatively large between-subject
38 variances in almost all endpoints, the number of subjects was probably insufficient to guarantee
39 adequate statistical power. In addition, no adjustments were made for multiple comparisons.
40 Since each endpoint was analyzed separately and because many comparisons were carried out
41 across genders, time points, doses, etc., the overall confidence level is probably much below the
42 nominal 95%. It was also noted that the univariate statistical models that were fitted to the data
43 account for the repeated measures structure of the design, but do not account for the multivariate
44 structure in the measurements. Because endpoints were measured on the same subjects, one
45 could expect meaningful correlations between the various endpoints that could contain additional
46 information about the treatment effects and the effects of covariates. Consequences of not using

1 multivariate methods on this data set include a decrease in the power of statistical tests and
2 erosion of confidence levels that occur when many models are fitted to many endpoints
3 independently as was done in this study. These considerations throw some doubt on some of the
4 study results that suggest no statistically significant treatment effects.

5
6 Nevertheless, while it is clear that the statistical power of the study was low, the data do
7 show a very clear and predictable dose- and time-dependency in the RBC cholinesterase data.
8 The response of the cholinesterase data is what would be expected of a transient carbamate
9 anticholinesterase. Additionally, it would be expected that blood cholinesterase would be the
10 most sensitive endpoint. RBC cholinesterase is well recognized as a sensitive biomarker of
11 exposure to anticholinesterases. Its inhibition is not responsible for clinical signs of
12 anticholinesterase toxicity, so no cause-and-effect relationship should be expected between RBC
13 cholinesterase inhibition and changes in any of the physiological parameters measured.
14 Therefore, it is also reasonable that inhibition of the RBC cholinesterase would occur whereas
15 the other parameters related to clinical signs, such as salivation or blood pressure, would not.
16 Lastly, the occurrence of numerous clinical signs in the placebo group gives credence to the
17 conclusion that the RBC cholinesterase inhibition was a treatment-related effect whereas clinical
18 signs were not.

19
20 The Board concluded that the results of the study could be used in the WOE analysis to
21 determine a NOEL for RBC cholinesterase and clinical signs in males and that the RBC
22 cholinesterase demonstrated a dose-dependent and time-dependent pattern of inhibition in both
23 males and females.

24 25 HSRB Consensus and Rationale

26
27 The Board concluded that the cholinesterase data from the aldicarb human study were
28 reliable for use in the aldicarb single chemical, aggregate risk assessment.

29
30 The Agency Data Evaluation Report for Aldicarb (Sette 1992) suggested that the NOAEL
31 and LOAEL were based upon sweating in males. While sweating is a possible clinical sign
32 resulting from cholinesterase inhibition, the responses in the subjects were not consistently dose-
33 related. Although sweating is an objective endpoint, the WOE document indicates that the RBC
34 cholinesterase inhibition would be the more appropriate objective endpoint. There are additional
35 reasons why there is more confidence in the cholinesterase data. First, the blood cholinesterase
36 data, despite their weaknesses, are the most consistent responses to aldicarb exposure, because
37 they follow time- and dose-dependent patterns, as would be expected of a transient
38 anticholinesterase. Second, the cholinesterase data, though probably based on incomplete
39 fractions of the entire RBC cholinesterase population due to the nature of the analysis method,
40 would still be expected to be internally consistent within this study between samples of treated
41 and placebo individuals, and therefore the inhibition levels can probably be believed. There was
42 a dose- and time-to-recovery-related relationship to cholinesterase inhibition in both sexes, so the
43 cholinesterase data are also consistent between the sexes.

44
45 The Board concluded that the cholinesterase data from the aldicarb human toxicity study
46 were reliable for use in the cumulative risk assessment for N-methyl carbamates. Aldicarb,

1 being an N-methyl carbamate, belongs to this common mechanism group which acts via
2 acetylcholinesterase inhibition. Therefore, the endpoint in the cumulative risk assessment must
3 be cholinesterase inhibition. Moreover, the dose-response data from the human study appeared
4 such that BMD and BMDL can be calculated. The HSRB concluded that the aldicarb human
5 study appears to be a scientifically valid study, suitable for use in both the aggregate risk
6 assessment and the cumulative risk assessment.

7
8 While the Board concluded that the cholinesterase data could be reliably used in the
9 cumulative risk assessment, it did note some limitations. The animal data were not supplied so
10 the HSRB cannot address the accuracy of the corresponding animal data. Because of the rapid
11 reactivation of carbamylated cholinesterase, it is unclear whether accurate cholinesterase
12 inhibition values were obtained in the human study or that peak inhibition was measured because
13 the earliest time point measured was 1 hour post-dose. The WOE document stated that the
14 human and rat cholinesterase inhibition were comparable at the 0.05 mg/kg dose level; a
15 question arises as to whether this comparison was made at the same point in the
16 inhibition/recovery patterns of both species. However, it should be possible to make
17 extrapolations of the possible peak from animal data on the time course pattern of cholinesterase
18 inhibition and recovery, and the Agency is urged to make certain that BMD and BMDL
19 comparisons between animals and humans are being made on comparable times in the
20 inhibition/recovery pattern.

21 22 **1.2. Ethical considerations**

23 24 **Charge to the Board**

25
26 a. The Agency requests that the Board provide comment on the following:
27

28 In light of the ethics committee's instruction that the lay summary be "greatly expanded,"
29 and the fact that the materials used to obtain informed consent listed a limited range of
30 symptoms of carbamate toxicity (excluding some reported as adverse effects in the study),
31 included multiple references to the test material as a drug, and failed to identify dose levels to be
32 administered to male subjects, whether, the materials used to obtain informed consent should be
33 considered significantly deficient relative to the ethical standards prevailing when the study was
34 conducted

35
36 Whether the absence from the protocol of discussion of the potential risks to subjects or
37 benefits to society of conducting the proposed research (as required by the 1989 Declaration of
38 Helsinki, Principle # 4, with which the research asserted compliance) should be considered
39 significantly deficient relative to the ethical standards prevailing when the study was conducted;
40

41 b. The Agency asks that the Board provide comment on the following, taking into account all
42 that is known about the ethical conduct of this study:
43

44 OPP's conclusion that there is not clear and convincing evidence that the conduct of the
45 research was fundamentally unethical.
46

1 Whether there is clear and convincing evidence that the conduct of the study was
2 significantly deficient relative to the ethical standards prevailing when the study was conducted.

3
4 **Board Response to the Charge**

5
6 Study Overview

7
8 The study was completed on March 11, 1992. The study sponsor was located in France
9 and the performing laboratory was located in Scotland. The protocol documents specifically
10 stated that the research was conducted in compliance with the Principles of Good Clinical
11 Practice as promulgated by the EMEA CPMP in May 1990, and the Declaration of Helsinki (in
12 either the 1983 or 1989 version, as cited in two different portions of the study documents).
13 Although the May 1990 document on good clinical practice was the precursor for the subsequent
14 1997 document promulgated by the International Conference on Harmonization, the earlier
15 document does not include any specification of the contents of the informed consent form.
16 However, the supporting and supplementary study documents claimed that the research ethics
17 committee providing independent review and oversight of the research functioned according to
18 the standards of the FDA regulations found in 21 CFR 56. These regulations cross-reference the
19 informed consent requirements found in 21 CFR 50 Both FDA regulations were effective on
20 January 27, 1981. As such, the research ethics committee should have applied the FDA
21 regulations from nearly a decade before the conduct of this particular research protocol. It
22 should be noted that the sponsor subsequently claimed that the study also was performed
23 according to the requirements of 40 CFR 26, based on a retrospective analysis published in 2003.

24
25 Critique of Study

26
27 The Board concurred with the Agency on the factual observations of the strengths and
28 weaknesses of the study, as detailed in Carley (2006a). However, further comments are
29 warranted on (1) the documentation and process of informed consent, and (2) the minimization
30 of risk with respect to the study design.

31
32 It is clear that the written documentation for informed consent is not up to the standards
33 found in the above regulations. The Volunteer Information that was given to research subjects as
34 an appendix to the signed informed consent document did not have an adequate discussion of the
35 risks of the research. At the request of the research ethics committee (dated December 24,
36 1991), the Volunteer Information included the possibility that administration of the test product
37 could result in "gut motility effects (abdominal pains), effect on eye pupil size and muscle
38 weakness"). In addition to the concerns that the risk information was insufficient in the written
39 consent documentation, selected subjects were enrolled a second time. This was not discussed or
40 in the protocol or the informed consent documentation and therefore raises doubts to the Board
41 about the adequacy of the consent process. In effect, the re-dosing of selected subjects suggests
42 that the investigator was unblinded to group assignment, reducing the supposedly double blind
43 study to a single-blind study and raising doubts (unanswered by the documentation) about the
44 sufficiency of informed consent. In particular, the re-use of selected subjects suggests that some
45 study participants were randomized to receive placebo versus active compound while other

1 subjects were not. As a result, the burden of risks was unlikely to have been distributed evenly
2 among all study participants.

3
4 There were several observations that raised concerns about the extent to which the study
5 design minimized risk to research subjects. First, the dosing schedule did not adhere to a strict
6 dose escalation design. As a result, it is possible that subjects dosed at higher levels would
7 experience significant adverse events that could have been anticipated in a more traditional dose
8 escalation design. The Board recognized that the public comments by Bayer CropScience
9 presented during the public comment period at the HSRB meeting suggests that there was
10 previous human testing data available to indicate that such risks would be unlikely. Second, the
11 stopping rule based on a 70% inhibition of cholinesterase activity is fairly liberal (i.e. not
12 protective), given that 70% inhibition would likely result in significant adverse effects.
13 Although no subject in the research actually experienced this level of cholinesterase reduction,
14 the selection of such a stopping rule increases the risk to subjects. Finally, the letter to the
15 subjects' general practitioner does not contain any information about the actual study and the
16 product being administered other than the name of the compound. As such, this minimizes the
17 extent to which the opinion of the general practitioner could protect the research subject.

18 HSRB Consensus and Rationale

19
20
21 The Board concluded that:

22
23 a) The aldicarb human toxicity study failed to fully meet the specific ethical standards
24 prevalent at the time the research was conducted.

25
26 b) There was no clear or convincing evidence that the research was fundamentally
27 unethical--intended to seriously harm participants or that informed consent was not obtained.

28
29 c) There was no clear and convincing evidence of significant deficiencies in the ethical
30 procedures that could have resulted in serious harm (based on the knowledge available at the
31 time the study was conducted), nor that information provided to participants seriously impaired
32 their informed consent.

33
34 Informed Consent: While informed consent was obtained, the informed consent
35 documents did not include adequate discussion of the research risks as documented by Carley
36 (2006a) and did not fully meet ethical standards prevalent at the time the study was conducted.
37 There was a suggestion that additional aspects of risk may have been discussed in the Volunteer
38 Information. Although the Board recognized that the documentation would not meet today's
39 standards, the lack of documentation does not in and of itself demonstrate a significant
40 deficiency that would have seriously impaired the participants' ability to provide informed
41 consent.

42
43 Participant risk: The Board concurred that the stopping rule based on 70% reduction in
44 cholinesterase activity raised the possibility of exposing subjects to inappropriate risk. However,
45 the Board acknowledged that data existed at the time to suggest that the doses used in the study

1 would likely not achieve this level. Thus, there is no evidence that serious harm was intended nor
2 that serious harm could have resulted from the implementation of the study.

3
4 The Board concluded that there was no obvious reason why the Agency cannot rely on
5 the results of this study, as appropriate under current pesticide laws, given the absence of clear
6 and convincing evidence that the research was fundamentally unethical or significantly deficient
7 relative to the ethical standards prevailing when the study was conducted.

8 9 **2. Methomyl**

10 11 **Charge to the Board**

12
13 Methomyl is a member of the *N*-methyl carbamate (NMC) common mechanism group
14 based on its ability to inhibit acetylcholinesterase via carbamylation. The Agency has
15 previously completed the acute, aggregate (single chemical, multi-route) risk assessment of
16 methomyl. At the present time, the Agency is considering the use of the methomyl acute oral,
17 human toxicity study to inform the inter-species uncertainty factor used in the cumulative risk
18 assessment of the NMCs.

19 20 **Scientific considerations**

21
22 The Agency's WOE document and DER for methomyl describe the study design and
23 results of the methomyl acute oral human study. The WOE document also discusses the
24 Agency's conclusions regarding the usefulness of the human study in the cumulative risk
25 assessment for the NMCs. For methomyl, the Agency has concluded that the human toxicity
26 study supports a 10X inter-species uncertainty factor for methomyl in the cumulative risk
27 assessment of the NMCs.

28
29 Please comment on the scientific evidence that supports this conclusion.

30 31 **Board Response to the Charge**

32 33 Study Overview

34
35 A study with methomyl was conducted in human subjects using a double-blind, placebo-
36 controlled, single ascending dose design (McFarlane et al. 1998). The defined goal of the study
37 was to determine an acute no-adverse effect level (NOAEL) based on inhibition of plasma and
38 RBC cholinesterase, and in the original protocol, the doses were projected to be 0.1, 0.3, 0.5,
39 0.75, 1 and 1.5 mg/kg. Methomyl was administered as a bolus dose in a capsule immediately
40 (about 5 minutes) following a meal, and post-dose blood sampling commenced at 15 minutes
41 after dosing. Collection of blood samples for analysis of plasma and RBC acetylcholinesterase
42 activity was continued for 24 hours after dosing. Plasma and RBC acetylcholinesterase activities
43 were compared to a baseline measurement, determined from two predose samples collected 16
44 hours and 30 minutes prior to dosing, for each subject. The results were compared to the placebo
45 group. In addition, numerous physiological parameters including ECG, heart rate, pulse, blood
46 pressure, body temperature, clinical chemistry and hematology parameters, determination of

1 pupil size and salivation were assessed during the entire 24 hour period after dosing. The
2 criterion for not escalating to the next dose was inhibition of RBC cholinesterase exceeding 40%.

3 4 Critique of Study

5
6 The scientific validity of this study was determined by its comparative strengths and
7 weaknesses. The strengths of this study are determined by the comprehensive analysis of
8 cholinesterase inhibition that was obtained including:

- 9
- 10 • The design and results established a clear dose-response and time-dependent relationship
11 for inhibition of RBC cholinesterase. This was based on the execution of three dose
12 levels in the ascending dose paradigm. The dosages evaluated were 0.1, 0.2 and 0.3
13 mg/kg.
 - 14 • The analysis of the temporal pattern of RBC cholinesterase inhibition was comprehensive
15 and complete. Specifically, blood samples were collected at 15 minutes after dosing,
16 with 15 minute sampling intervals for the first 2 hours, followed by sample collection for
17 evaluation at 2, 3, 4, 6, 8, 12 and 24 hours after dosing. This time course established the
18 peak of cholinesterase inhibition and assured recovery of enzyme activity in all subjects.
 - 19 • The inclusion of clinical endpoints associated with increased cholinergic stimulation as
20 additional endpoints provided additional relevant measures of potential adverse effects
21 during the course of the 24 hour study.

22
23 Several weaknesses of the study were noted, and these weaknesses are also based on aspects
24 relating to its design and conduct and include:

- 25
- 26 • All subjects were male, with no data obtained from females
 - 27 • The group size (5/dose level) was small
 - 28 • The establishment of the baseline RBC cholinesterase level from which all subjects were
29 compared was the mean of 2 determinations, which, given the general pattern of
30 variation, reduces the confidence in the calculation of percent inhibition of the enzyme.

31
32 One additional issue concerning the design and execution of this study was the initial
33 proposed dose escalation which ranged from 0.1 to 1.5 mg/kg (0.1, 0.3, 0.5, 0.75, 1, and 1.5
34 mg/kg) and the criterion to not escalate dose was inhibition of RBC cholinesterase exceeding
35 40%. There was one instance in which this criterion was not adhered to. In the first session of
36 the study, one subject was dosed with placebo and the other received methomyl at 0.1 mg/kg.
37 The subject receiving methomyl showed evidence of RBC cholinesterase inhibition exceeding
38 40% (approximately 43%), but this change was determined at 8 hours after dosing. The study
39 sponsor concluded that, based on the general knowledge and understanding of the rapidity of
40 enzyme inhibition observed with the carbamates, this finding was likely to be a spurious result.
41 In review of the data for RBC cholinesterase inhibition for this session, it was also noted that the
42 placebo-treated subject in this session showed a 20% decrease in baseline enzyme activity,
43 thereby suggesting a possible, albeit unconfirmed, issue regarding the accuracy and reliability of
44 the analysis of enzyme activity in these samples. Therefore, the sponsor's conclusion that the
45 effect observed in the subject dosed with 0.1 mg/kg methomyl was most likely to be a spurious

1 result was considered scientifically reasonable and justifiable, and the decision to escalate the
2 dose in the next session was not in violation of the protocol.

3
4 In the second session of the study, the lead subject dosed with methomyl at 0.3 mg/kg also
5 showed a greater than 40% inhibition of RBC cholinesterase. Consequently, the next session
6 completed the five subjects to be used in this group but did not escalate the dose further as per
7 the study protocol. Therefore, the highest dosage evaluated in the study was 0.3 mg/kg, which
8 was much lower than the original intended escalation up to 1.5 mg/kg. To establish a better
9 dose-response relationship, the study was unblinded, and a dosage of 0.2 mg/kg was added to the
10 protocol after which the study was re-blinded and completed.

11 HSRB Consensus and Rationale

12
13
14 The Board concluded that the methomyl human study could be appropriately applied to the
15 inter-species risk factor for methomyl and for use in cumulative risk assessment of N-methyl
16 carbamates.

17
18 The HSRB concluded that data from the study in human subjects exposed to methomyl
19 provided a meaningful assessment of the dose-response relationship and time course for
20 inhibition of RBC cholinesterase in human subjects. The strengths of the study design and
21 conduct outweigh the indicated weaknesses, and the results are scientifically valid. Therefore,
22 the data can be used to inform the assessment of interspecies uncertainty factors for methomyl.
23

24 **Ethical considerations**

25 **Charge to the Board**

26
27
28 2.2.1. The Agency requests that the Board provide comment on the following:
29

- 30 • Whether the investigators' decision to administer a dose to additional subjects in session
31 3, when one subject receiving that dose in session 2 displayed RBC ChEI greater than
32 40%, a response that triggered the protocol's anti-escalation provision, should be
33 considered significantly deficient relative to the ethical standards prevailing when the
34 study was conducted;
- 35
36 • Whether the timing of the investigators' report to the ethics committee of the adverse
37 effects observed in one subject during session 2 should be considered significantly
38 deficient relative to the ethical standards prevailing when the study was conducted;
39
- 40 • Whether the failure of the investigators to request approval from the ethics committee for
41 certain amendments to the approved protocol, as required by the protocol, when the
42 changes were administrative and had no effect on the safety of the subjects should be
43 considered significantly deficient relative to the ethical standards prevailing when the
44 study was conducted; and
45

- Whether the absence from the protocol of discussion of the potential risks to subjects or benefits to society of conducting the proposed research (as required by the Declaration of Helsinki, Principle # 5) should be considered significantly deficient relative to the ethical standards prevailing when the study was conducted; and

2.2.2. The Agency asks that the Board provide comment on the following, taking into account all that is known about the ethical conduct of this study:

- OPP's conclusion that there is not clear and convincing evidence that the conduct of the research was fundamentally unethical.
- Whether there is clear and convincing evidence that the conduct of the study was significantly deficient relative to the ethical standards prevailing when the study was conducted,

Board Response to the Charge

Brief Overview of the Study

The study was performed by a commercial research organization, Inveresk Clinical Research of Edinburgh, Scotland and sponsored by the Haskell Laboratory for Toxicology and Industrial Medicine, E.I. du Pont de Nemours and Company, of Newark, Delaware. The protocol documents specifically state that the research was conducted in compliance with the Principles of Good Clinical Practice promulgated by the International Conference on Harmonization (CMPM/ICH/135/95), and the 1996 version of the Declaration of Helsinki.

Critique of Study

The Board concurred with the Agency's factual observations of the strengths and weaknesses of the study, as detailed in Carley (2006b). However, further comments are warranted on certain events that took place during the conduct of the study.

1. Determination That Certain Data Were Spurious

In session one of the study, one of the subjects exhibited a greater than 40% inhibition in RBC cholinesterase activity at one point, without any signs or symptoms. According to the protocol, such inhibition of cholinesterase was a criterion for not escalating to the next dose level. Because this occurred at 8 hours post dose, study personnel considered this to be a spurious result and went ahead with escalation to the next dose without consulting with the ethics review board (It is somewhat unclear when the results of the assay became available; the observation about this result is described as being noted "retrospectively at the time of reporting, when there was no sample available for retesting."). If this determination of spuriousness had been incorrect, then giving the following subject an escalated dose might have placed that subject at considerable risk.

1 The Board concluded that the determination of spuriousness was reasonable at the time,
2 given the late timing of the change in cholinesterase levels, and the similar change observed in
3 cholinesterase levels for a subject in session one who received placebo. Nonetheless, the Board
4 determined that a far more appropriate action would have been for the researchers to have first
5 consulted with the ethics review committee about repeating that 0.1 mg/kg dose before
6 proceeding with dose escalation. Moreover, the report of the study results stated that “laboratory
7 tests showing abnormal values for any subject were repeated as often as deemed necessary by the
8 clinical investigator until the test values returned to accepted limits or until an explanation other
9 than compound effect was given.” In spite of this, it does not appear that any additional tests
10 were performed; study participants only received scheduled laboratory tests at the 12 hour and 24
11 hour points. Although there is a possibility that these events might have placed subjects at an
12 increased risk of harm, the Board determined that there was not clear and convincing evidence
13 that this could have resulted in serious harm to subjects, particularly given the information
14 known about the compound being tested and the other protections in place as part of the
15 protocol.

16
17 2. Decision to Proceed with Third Session

18
19 In session two of the study, one subject who received an escalated dose of 0.3 mg/kg
20 exhibited a greater than 40% inhibition in RBC cholinesterase, together with self-reported
21 clinical symptoms (i.e. headache). As noted above, the protocol indicated that dose escalation
22 would not occur if any subject experienced such a level of inhibition. The protocol was followed
23 in this regard: additional subjects were tested at the 0.3 mg/kg dose, but not at any higher doses.
24 However, the protocol also stated that the study would stop if a subject both had greater than
25 40% inhibition and also demonstrated signs or symptoms of carbamate toxicity (one of which
26 might be considered to be a headache). Although the Board was concerned that this decision to
27 continue the study and to test additional subjects at the 0.3 mg/kg dose was made without
28 consulting the ethics review committee, it determined that the provisions of the protocol were
29 sufficiently unclear as applied to these facts (particularly regarding what constitute signs and
30 symptoms of toxicity); the actions of the investigators were thus consistent with the protocol as
31 written. In addition, given the protections that were in place as part of the protocol, and the
32 information then known about the compound being studied, the Board concluded that there was
33 not clear and convincing evidence that this decision presented a risk of substantial harm to the
34 subjects. Nonetheless, the Board concluded that a more appropriate course of action would have
35 been for the researchers to have consulted with the ethics review board before proceeding with
36 the study. This conclusion and the desirability of having taken similar actions after the
37 apparently “spurious” result in session one is underscored by public comments presented to the
38 Board that the observation of acetylcholinesterase depression at 0.3 mg/kg was “surprising.”
39 There is a danger in explaining away unexpected findings based on prior expectations, and ethics
40 review boards should play an important role in preventing investigators from falling prey to such
41 expectations.

42
43 3. Amendment of the Protocol

44
45 Following the testing of the subjects at the 0.3 mg/kg level as part of session three, the
46 protocol was amended to allow additional subjects to be tested at the 0.2 mg/kg level. This

1 change was approved by the chair of the ethics review committee, consistent with procedures for
2 the committee which allowed the chair to approve amendments that did not increase the risk
3 level (i.e., did not involve testing subjects at a dose level above those approved by the convened
4 board). Although the Board did not conclude this was a formal violation of then-existing ethical
5 standards, some members of the Board were concerned that the ethics committee chair's actions
6 were inappropriate and that the protocol change should have been reviewed by the full
7 committee because additional subjects were being exposed to a study compound beyond those
8 contemplated by the unamended protocol.

9
10 HSRB Consensus and Rationale

11
12 The Board concluded that:

13
14 a. The methomyl human toxicity study failed to fully meet the specific ethical standards
15 prevalent at the time the research was conducted.

16
17 b. There was no clear and convincing evidence that the research was fundamentally
18 unethical--intended to seriously harm participants or that informed consent was not obtained.

19
20 c. There was no clear and convincing evidence of significant deficiencies in the ethical
21 procedures could have resulted in serious harm (based on the knowledge available at the time the
22 study was conducted), nor that information provided to participants seriously impaired their
23 informed consent.

24
25 (1) The Board concluded that the decision to administer a dose to additional subjects in
26 session three could not be determined to be significantly deficient relative to the ethical standards
27 prevailing when the study was conducted because, as discussed above, that decision could be
28 interpreted as consistent with the terms of the protocol and was not likely to expose the subjects
29 to additional serious harm.

30
31 (2) The Board concluded that the timing of the investigators' report to the ethics
32 committee regarding what happened to the subject who experienced cholinesterase inhibition
33 during session two should not be considered significantly deficient relative to the ethical
34 standards prevailing when the study was conducted, given the subject's minimal symptoms and
35 the fact that RBC cholinesterase inhibition by itself is not considered an adverse event.

36
37 (3) The Board determined that the failure of the investigators to request approval from the
38 ethics committee for certain amendments to the approved protocol, while deficient relative to the
39 ethical standards prevailing at the time the study was conducted, should not be considered
40 significantly deficient, given that these changes were purely administrative.

41
42 (4) The Board determined that the absence from the protocol of discussion of the
43 potential risks to subjects or benefits to society of conducting the proposed research (as required
44 by the Declaration of Helsinki, Principle # 5) should not be considered significantly deficient
45 relative to the ethical standards prevailing when the study was conducted. The potential risks and
46 benefits are described in the research subject information sheet/informed consent document.

1 While not in the protocol itself, they are nonetheless included in the materials provided to the
2 ethics committee (and to the potential research subject). Thus, the ethics committee and the
3 potential research subjects had the opportunity to weigh the risks and benefits.
4

5 The Board concluded that there was no obvious reason why the Agency cannot rely on
6 the results of this study, as appropriate under current pesticide laws, given the absence of clear
7 and convincing evidence that the research was fundamentally unethical or significantly deficient
8 relative to the ethical standards prevailing when the study was conducted.
9

10 **3. Oxamyl**

11 **Charge to the Board**

12
13
14 Similar to aldicarb and methomyl, oxamyl is a member of the *N*-methyl carbamate
15 (NMC) common mechanism group based on its ability to inhibit acetylcholinesterase via
16 carbamylation and is thus included in the NMC cumulative risk assessment. The Agency has
17 previously completed the acute, aggregate (single chemical, multi-route) risk assessment of
18 oxamyl. The Agency is now considering the use of the oxamyl acute oral, human toxicity study
19 to inform the inter-species uncertainty factor in the cumulative risk assessment of the NMCs.
20

21 **3.1. Scientific considerations**

22
23 The Agency's WOE document and DER for oxamyl describe the study design and results
24 of the oxamyl acute oral human toxicity study. The WOE document also discusses the Agency's
25 conclusions regarding the usefulness of the human study in the cumulative risk assessment for
26 the NMCs. For oxamyl, the Agency has concluded that the human toxicity study is sufficiently
27 robust for reducing the 10X inter-species (i.e., animal to human) uncertainty factor in the
28 cumulative risk assessment.
29

30 Please comment on the scientific evidence that supports this conclusion.
31

32 **Board Response to the Charge**

33 Study Overview

34
35
36 The stated primary objective of this randomized, double-blind, ascending oral dose study
37 with oxamyl (McFarlane and Freestone 1999a) was to estimate the no-observable-adverse effect
38 level (NOAEL) for oxamyl in humans after oral administration.
39

40 The study used 40 healthy male subjects, aged 19-39. Each was given a single oral dose
41 of oxamyl in a gelatin capsule at doses of 0 (placebo), 0.005, 0.015, 0.03, 0.06, or 0.15 mg/kg
42 BW. With the exception of dose sessions 1 (2 placebos), 2 (1 placebo and 1 at 0.005), 7 (1
43 placebo and 4 at 0.09), 8 (1 placebo and 1 0.15), and 9 (1 placebo and 4 at 0.15), all other dose
44 sessions had 1 placebo, 4 at current dose, and 1 at the next higher dose. Plasma and RBC
45 cholinesterase activity were assayed at screening and 2 days, 16 hours and 30 minutes prior to
46 dosing and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2, 3, 4, 6, 8, 12, and 24 hours post dose and 7

1 days post dose. Other clinical parameters indicative of carbamate exposure, such as urinalysis,
2 pupillometry, saliva increase etc were also measured.

3
4 No treatment related effects for ECG, heart rate, pulse, blood pressure, respiratory rate,
5 body temperature, hematology, clinical chemistry, urinalysis, or pupillometry were recorded.

6
7 At 0.09 mg/kg b.w., 7-12% plasma and RBC ChE inhibition was observed with three of
8 five volunteers exhibiting greater than 20% plasma ChE inhibition. Therefore, 0.06 mg/kg b.w.
9 was considered the NOAEL. The Agency generated BMD and BMDL estimates of 0.083 mg/kg
10 and 0.069 mg/kg, respectively, based on the RBC ChE data from the study

11 Critique of the Study

12
13
14 The scientific validity of this study was analyzed by weighing its strengths and
15 weaknesses in order to determine if the study was sufficiently robust to be of use in the Agency's
16 risk assessment for oxamyl.

17
18 Appropriate endpoints for this study were selected. Acetyl cholinesterase inhibition in
19 the nervous system was viewed as a key molecular event in the mechanism of toxicity of
20 carbamate compounds. Measurements of acetyl cholinesterase inhibition in both RBCs and
21 brain are considered to be biomarkers of exposure for these compounds. In its policy on the se
22 of data on cholinesterase inhibition for risk assessments of organophosphorus and carbamate
23 pesticides (USEPA, 2000), the Agency stated that it will treat cholinesterase inhibition in the
24 blood as a surrogate measure for the peripheral nervous system in animals and for the peripheral
25 and central nervous system in humans.

26
27 The animal data for oxamyl were not available to the Board to allow it to make an
28 independent judgment as to whether the methods used to determine acetyl cholinesterase
29 inhibition in the animal studies and the human study were comparable. According to EPA,
30 however, the methods used in the animal studies are comparable to that in the human study.

31
32 Previous animal data was used to predict that the starting dose 0.005 mg/kg would have
33 no detectable effect on humans. Oral administration of the compound is the most predictive
34 mode of administration since exposure to residues would come from food consumption. There
35 was no justification for the sample size of 4 for oxamyl for each dose session.

36
37 Forty healthy males of similar age and weight were used in this study. A weakness of
38 this study is that no female subjects were used in this study and there was no indication that any
39 human female data exists from other studies. However, the Agency advised the Board that, based
40 upon its evaluation of the animal data, no sex differences would be expected.

41
42 The specific schedule for dosing was described in protocol. It was an ascending double
43 blind, placebo controlled study. Dose escalation did not occur if there were clinically significant
44 symptoms/signs of toxicity. Stopping rules were equal to or greater than 40% inhibition of RBC
45 cholinesterase at a single time point or 25% at two consecutive time points. If there were
46 associated symptoms/signs of carbamate toxicity, no further dose escalation would occur.

1
2 A repeated measures analysis of variance was used to analyze the percentage change
3 from baseline in BRC and plasma cholinesterase activity, pupillometry and saliva collection. At
4 each time point of measurements, a test for linear trend with dose was performed using a linear
5 contrast. If the test for linear trend was significant at the 5% significance level, the pairwise
6 comparisons at each timepoint were not adjusted for multiple comparisons. Otherwise, a
7 Bonferroni adjustment was applied to the pairwise test at the 0.83% significance level.
8

9 At 0.09 mg/kg b.w., 7-12% plasma and RBC ChE inhibition was observed with three of five
10 volunteers exhibiting greater than 20% plasma ChE inhibition. Therefore, 0.06 mg/kg b.w. was
11 considered the NOAEL. The Agency generated BMD and BMDL estimates of 0.083 mg/kg and
12 0.069 mg/kg, respectively, based on the RBC ChE data from the study
13

14 The Board highlighted the strengths of the study as follows:
15

16 (a) The study design, including both the increasing dose levels and the time point of
17 observations/measurements, was very rich in information content to allow evaluation of dose-
18 response and estimation of the NOAEL and the dose-escalation in a step-wise fashion was
19 scientifically sound considering the uncertainties in the risk.
20

21 (b) The study had a very clearly defined dose-escalation scheme with properly justified
22 stopping rules. The schedule of observation and measurements were frequent enough to give
23 sufficient information on the time-course effect of oxamyl on subjects.
24

25 (c) The statistical methods used for analysis, including the adjustment for multiple
26 comparisons between placebo and active doses when there was no statistical significance in the
27 linear trend test, were quite adequate for the study.
28

29 The Board highlighted the weaknesses of the study as follows:
30

31 (a) There appears to be no specific justification for the sample size of four for oxamyl for
32 each dose session and no traditional statistical test of effect size regarding hypotheses. Because
33 the study was exploratory versus confirmatory, the lack of justification was not critical.
34 Nevertheless it would have been helpful to know what effect size could be detected between the
35 placebo and the active dose groups given the sample size of the study.
36

37 (b) Because of lack of justification for the sample size, the lack of statistical significance
38 in the observed difference between placebo and active doses could not be interpreted
39 appropriately. Specifically the statement in the Agency Data Evaluation Report (Moyer and
40 Milznez 2001) that "RBC cholinesterase activity was statistically similar to placebo at all time
41 points in the 0.005, 0.015, 0.03, and 0.06 mg/kg dose groups" was not justified as the study was
42 not designed to test for statistical similarity.
43

44 (c) The Agency concluded that "there is no indication from the available rat data for
45 either oxamyl or methomyl to suggest a sex difference." The animal data were not supplied to

1 the Board and therefore the Board could not independently verify this conclusion. However the
2 Board was not uncomfortable with accepting EPA's conclusions on this matter.

3
4 The Agency also concluded that "Given the similarity between brain and RBC ChE in
5 rats, the Agency is not aware of any biological or physiological reason that human brain ChE
6 would be more sensitive than the rat brain." Again without seeing the animal data, the Board
7 could not verify this conclusion about rats, but the conclusion about human brain and RBC
8 cholinesterase is reasonable scientifically

9 10 HSRB Consensus and Rationale

11
12 Although the Board had some questions about the Agency's conclusions regarding lack
13 of sex difference and of the difference between brain and RBC ChE based on only one species,
14 the Board supported the Agency's conclusion that there were no study deficiencies identified that
15 would have affected the outcome of conclusions of this study.

16
17 Considering the high quality of the design and the conduct of the study, the Board agreed
18 that this intentional human dosing study of oxamyl was sufficiently robust to be used for
19 reducing the 10x inter-species (i.e. animal to human) uncertainty factor in the cumulative risk
20 assessment for the N-methyl carbamates.

21 22 **3.2. Ethical considerations**

23 24 **Charge to the Board**

25
26 a. The Agency requests that the Board provide comment on the following:

- 27
28
- 29 • Whether inclusion in the protocol submitted to the ethics committee of a factually
30 inaccurate statement regarding unavailability of data on accidental or incidental
31 exposure to oxamyl should be considered significantly deficient relative to the
32 ethical standards prevailing when the study was conducted;
 - 33 • Whether the absence from the protocol of any discussion of the potential risks to
34 subjects or benefits to society of conducting the proposed research (as required by
35 the Declaration of Helsinki, Principle # 5) should be considered significantly
36 deficient relative to the ethical standards prevailing when the study was
37 conducted; and
- 38

39 b. The Agency asks that the Board provide comment on the following, taking into
40 account all that is known about the ethical conduct of [this/each] study:

- 41
42
- 43 • OPP's conclusion that there is not clear and convincing evidence that the conduct
44 of the research was fundamentally unethical.

- Whether there is clear and convincing evidence that the conduct of the study was significantly deficient relative to the ethical standards prevailing when the study was conducted.

Board Response to the Charge

Study Overview

The study was performed by a contract research organization, Inveresk Clinical Research of Edinburgh, Scotland and sponsored by the Haskell Laboratory for Toxicology and Industrial Medicine, E.I. du Pont de Nemours and Company, of Newark, Delaware. The protocol documents specifically state that the research was conducted in compliance with the Principles of Good Clinical Practice promulgated by the International Conference on Harmonization (CMPM/ICH/135/95), and the 1996 version of the Declaration of Helsinki. Comments provided by du Pont Crop Protection (Dupont 2006), also assert compliance with the 1996 version of the Declaration of Helsinki.

Critique of Study

The Board concurred with the factual observations of the strengths and weaknesses of the study, as detailed in Carley 2006b). However, further comments are warranted regarding: 1) whether the exclusion of data on accidental or incidental exposure to oxamyl meets the legal definition of research misconduct prevalent at the time the study was conducted; 2) whether the escalating-dose oral-exposure protocol used was designed to minimize risks to study participants; and 3) whether the documentation and process of study subject enrollment was sufficient to meet prevailing standards of voluntary informed consent.

1) Exclusion of Available Data on Accidental or Incidental Exposure

Page 15 of the original study protocol provided to the independent ethics review committee established by Inveresk Clinical Research included the following statement: “No data on accidental or incidental exposure of people to oxamyl is available” (McFarlane and Freestone 1999). This statement is false. Prior to the start of the Randomised Double Blind Ascending Oral Dose Study with Oxamyl in late 1998, Du Pont submitted eleven adverse event reports involving exposure of humans to oxamyl for inclusion in the EPA’s Office of Pesticide Programs (OPP) Incident Data System (IDS). Between 1982 and 1994, the State of California Department of Food and Agriculture’s Pesticide Illness Surveillance Program recorded an additional 61 incidents, including 30 cases involving occupational exposure to oxamyl alone. In comments provided to the EPA on March 31, 2006, Du Pont Crop Protection stated that “most of the accidents or incidents reported in these databases lack confirmation of the product involved or the amount of product involved in the exposure. Also, some reports lack information on symptoms or details of the exposure. Therefore, none of this data was useful for designing the study.” (DuPont 2006). This latter statement may be true, but the discounting of the relevance of adverse event data from accidental or incidental exposures is ill-advised and borders on the unprofessional. This may have been an appropriate decision but the data, evaluation and assessment should have been discussed in the protocol and the consent document. Furthermore,

1 the statement in the original study protocol remains untrue. The question before the Board,
2 therefore, was whether or not this misstatement met the legal definition of research misconduct.

3
4 The Agency has accepted the federal policy on research misconduct (Classification No.:
5 3120.5; Approval Date: 03/18/2003; Review Date: 03/18/2006), as defined in 42 CFR 93.103:

6
7 Research misconduct means fabrication, falsification, or plagiarism in proposing,
8 performing, or reviewing research, or in reporting research results.

9
10 (a) Fabrication is making up data or results and recording or reporting them.

11
12 (b) Falsification is manipulating research materials, equipment, or processes, or changing
13 or omitting data or results such that the research is not accurately represented in the research
14 record.

15
16 (c) Plagiarism is the appropriation of another person's ideas, processes, results, or words
17 without giving appropriate credit.

18
19 (d) Research misconduct does not include honest error or differences of opinion.
20 Although the failure to include the incidental exposure data may meet this definition of
21 falsification, a finding of research misconduct made under 42 CFR 93.104 requires that: 1) there
22 be a significant departure from accepted practices of the relevant research community; 2) the
23 misconduct be committed intentionally, knowingly, or recklessly; and 3) the allegation be proven
24 by a preponderance of the evidence. Under the standards outlined in 42 CFR 93.103-104, given
25 the limitations in the studies cited by Du Pont, and lack of evidence that the absence of
26 information was intentional, knowing, or reckless, it is the opinion of the Board that this
27 omission does not meet the evidentiary threshold necessary for a finding of research misconduct
28 based on falsification.

29
30 2) Minimization of Risks to Study Participants

31
32 Reasonable exposure levels appear to have been chosen using available animal data.
33 Although human exposure data from the EPA OPP IDS and California's Pesticide Illness
34 Surveillance Program also may have been useful in designing the study so as to minimize risk to
35 research subjects, there is no clear and convincing evidence of willful or blatant disregard for
36 participant safety. Adherence to a strict dose escalation protocol, with a stopping rule based on
37 40% inhibition of RBC cholinesterase, reduced the likelihood that study subjects would
38 experience unanticipated adverse clinical events. Based on the knowledge available to
39 investigators at the time the study was conducted, this double-blind ascending-dose oral
40 exposure trial appears to have been designed to adequately minimize potential risks to study
41 participants.

42
43 3) Voluntary Informed Consent
44

1 It is clear that the written documentation for informed consent failed to meet the
2 standards outlined in Principles of Good Clinical Practice (CMPM/ICH/135/95), and the 1996
3 version of the Declaration of Helsinki.

4
5 The information sheet that was given to research subjects as an appendix to the signed
6 informed consent document lacked adequate discussion of the potential risks of participating in
7 the research study. Several examples of what Board members consider inadequate discussion of
8 potential risks are illustrated below:

9
10 Data on incidents involving accidental oxamyl exposure were available through both the
11 EPA's OPP IDS and the California Pesticide Illness Surveillance Program, as previously
12 discussed. These data included reports of adverse clinical events, including detailed reports of
13 systemic effects, eye effects and dermatological effects resulting from exposure to oxamyl.
14 These data were not provided to study participants.

15
16 The background information provided to study participants enrolling in both the original
17 and amended study included the statement that "results from this study would further confirm
18 that the use of oxamyl for crop protection would not pose an unreasonable risk to human health."
19 (McFarlane and Freestone 1999). Although the informed consent document clearly states that
20 this is the first study involving intentional exposure of human subjects, particularly given the
21 incident data discussed above, this statement seems to be written in order to downplay any fears
22 that study participants might have with regard to participation.

23
24 Study participants enrolling in the amended study, designed to look for clinical or
25 biological outcomes associated with exposure to 0.15 mg/kg body weight of oxamyl, were
26 informed that "no significant side effects [were] noticed in any of [the previous lower] dose
27 levels." (McFarlane and Freestone 1999). Previous trial participants receiving 0.09 mg/kg body
28 weight of oxamyl exhibited 4-24% and 12-22% RBC and plasma cholinesterase inhibition. The
29 study investigators concluded that these observed levels of cholinesterase inhibition did not
30 qualify as adverse events because of the variability in the data, but they also state that the level of
31 inhibition observed in study participants receiving 0.09 mg/kg body weight of oxamyl achieved
32 statistical significance. Because the consent form described cholinesterase inhibition as a side
33 effect of oxamyl exposure, it can be argued that this information should have been provided to
34 subjects participating in the 0.15 mg/kg dose study. The information sheet provided to
35 volunteers, however, specifically describes cholinesterase inhibition as one of the side effects of
36 oxamyl exposure.

37
38 The letter to the subjects' general practitioner contained no information about the actual
39 study and the product being administered other than the name of the compound. It thus
40 minimized the extent to which the opinion of the general practitioner could protect the research
41 subject.

42
43 In hindsight, it is clear that the documentation provided failed to rigorously meet the
44 standards of voluntary informed consent applicable to studies conducted in Scotland in 1998-
45 1999. However, there was no clear and convincing evidence that these deficiencies knowingly
46 and seriously impaired the informed consent process.

1
2 HSRB Consensus and Rationale

3
4 The Board concluded that:

5
6 a) The oxamyl human toxicity study failed to fully meet the specific ethical standards
7 prevalent at the time the research was conducted.

8 b) There was no clear or convincing evidence that the research was fundamentally
9 unethical--intended to seriously harm participants or that informed consent was not obtained.

10 c) There was no clear and convincing evidence of significant deficiencies in the ethical
11 procedures that could have resulted in serious harm (based on the knowledge available at the
12 time the study was conducted) nor that information provided to participants seriously impaired
13 their informed consent.
14

15 Although the study failed to meet the specific ethical standards prevalent at the time the
16 research was conducted, including those cited in the study protocol itself, the Board concurred
17 with the assessment of the Agency that there was no clear and convincing evidence that the
18 conduct of the research was fundamentally unethical in that it was not intended to harm subjects
19 nor failed entirely to obtain informed consent.
20

21 It is believed that potential risks to study participants were inadequately addressed in the
22 documents provided to research participants at enrollment, thus impairing the informed consent
23 process. Nevertheless, the Board believed that there was no clear and convincing evidence to
24 suggest that the study investigators failed to obtain voluntary informed consent from all research
25 participants.
26

27 The Board was of the opinion that the deficiencies noted above are unlikely to have
28 resulted in serious harm to study participants, based on the knowledge available to the
29 investigators at the time.
30

31 The Board concluded that there is no obvious reason why the Agency cannot rely on the
32 results of this study, as appropriate under current pesticide laws, given the absence of clear and
33 convincing evidence that the research was fundamentally unethical or significantly deficient
34 relative to the ethical standards prevailing when the study was conducted.
35

36 **4. Azinphos-Methyl**

37
38 **Charge to the Board**

39
40 Azinphos methyl (AZM) is an organophosphate pesticide (OP). Consistent with other
41 OPs, AZM elicits neurotoxicity through the inhibition of the enzyme, acetylcholinesterase, via
42 phosphorylation of the active site. At sufficiently high doses, exposure to AZM can lead to a
43 variety of clinical signs. The Agency is developing an assessment to estimate risk to workers
44 from exposure to AZM. In addition, AZM is a member of the OP common mechanism group
45 and is thus included in the cumulative risk assessment for the OPs.
46

1 Scientific considerations

2
3 The Agency's WOE document and DER for AZM describe the study design and results
4 of the AZM repeat dose, oral, human toxicity study. The WOE document also discusses the
5 Agency's conclusions regarding the usefulness of the human study in the worker risk assessment
6 and in the cumulative risk assessment for the OPs. For AZM, the Agency has concluded that the
7 human toxicity study is appropriate for developing a point of departure for extrapolation of risk
8 to workers exposed to AZM via the dermal and inhalation routes. For the cumulative risk
9 assessment, the Agency has determined that because no cholinesterase inhibition was seen in the
10 human toxicity study, it is not possible to evaluate whether steady state had been reached in
11 humans at 28 days of exposure. Thus, the Agency has concluded that the AZM repeat dose, oral,
12 toxicity study is not sufficiently robust for informing the inter-species factor in the cumulative
13 risk assessment of the OPs.

14
15 Please comment on the scientific evidence that supports the conclusions for:

- 16
17 a. the use of the human toxicity study to develop a point of departure for extrapolation of
18 risk to workers in the worker risk assessment and
19
20 b. the determination that the human toxicity study cannot be used to inform the inter-
21 species factor in the cumulative risk assessment.
22

23 **Board Response to the Charge**

24
25 Brief Overview of the Study

26
27 In a repeat dose oral human toxicity study, conducted in 1999 by the Inveresk Laboratory
28 (McFarlane & Freestone, 1999b), four males were dosed with a placebo (lactose) and eight
29 males were dosed with 0.25 mg/kg/day of AZM for 28 consecutive days. The sole dose of 0.25
30 mg/kg was chosen based on the results of subchronic rat study and an earlier study with human
31 subjects that indicated this would be a NOAEL for inhibition of plasma and RBC cholinesterase.
32 Previous human studies were conducted in 1972 and 1974 and regulatory agencies reluctant to
33 rely on the data "due to deficiencies in documentation, specifically the availability of raw data
34 and the lack of GLP compliance."
35

36 Dosing was orally by capsule and the subjects were housed in a clinic throughout the
37 dosing period. The subjects were monitored for reactions and samples of blood were taken at
38 predose (eight times) and daily throughout the dosing period. On some days, a blood sample was
39 taken both prior to administering the dose and four hours later. Both plasma cholinesterase (ChE)
40 and RBC acetylcholinesterase (AChE) were measured. In addition, hematology, clinical
41 chemistry, and urinalysis were evaluated as well as blood pressure and electrocardiogram (ECG).
42 There were no reactions to treatment and blood pressure and ECG were not affected. Eight of
43 the 12 volunteers were noted to have symptoms of a viral infection and some were treated with
44 paracetamol. The postdosing group mean data were compared three ways in an effort to see if
45 AZM caused inhibition of either plasma ChE or RBC AChE. None of the comparisons indicated
46 inhibition of either measure. Also, samples taken four hours after dosing did not indicate

1 inhibition. It was concluded that a dose of 0.25 mg/kg/day for 28 days did not result in either
2 clinical symptoms or inhibition of either plasma ChE or RBC AChE in human males, so that this
3 dose could be considered a NOAEL.

4 5 Critique of the Study

6
7 One strength of the study was its design as a randomized, double blind study including a
8 placebo group. The clear inclusion and exclusion criteria for subject selection appeared to have
9 been chosen to reduce the likelihood of confounding. The subjects were in good health, were not
10 using prescription or illicit drugs, were non-smokers, and were not agricultural workers or
11 pesticide applicators who might have experienced OP exposure within the preceding month.
12 Subjects resided in the clinic from the day before study start to 24 hours after the last dose was
13 administered, received a standardized diet, and were under medical and nursing supervision
14 throughout.

15
16 The assessment of critical outcomes was comprehensive, involving a general clinical
17 exam, measurement of systolic and diastolic blood pressure, the administration of an ECG, and
18 various blood and urine studies. Multiple measurements were made of plasma ChE and RBC
19 AChE, namely on 8 pre-dose days then daily 4 hours after dosing. Moreover, on 12 post-dosing
20 days, two blood samples were taken, one before the dose and one 4 hours afterwards.

21
22 Possible group differences in plasma ChE and RBC AChE were evaluated in several
23 ways, both in grouped and individual data)

- 24 a) Comparison of the day-to-day ChE activities within the AZM group to the same day
25 values in the control group
26 b) Comparison of same day predosing ChE activities with inhibition 4 hours after dosing
27 c) Comparison of the day-to-day ChE activities values within the AZM group to
28 predosing baseline value.

29
30 Several weaknesses were identified in the study. First, the subjects were all male,
31 however there have been no consistent sex differences in sensitivity observed in animal models.
32 The sample size was not large, involving only 12 adult males (four in the placebo group, eight in
33 the AZM group), raising a concern that the statistical power might have been inadequate to
34 detect a modest degree of inhibition in the AZM group.

35
36 The standard deviations of the ChE levels were large, as great as 20% for plasma ChE
37 and 15% for RBC ChE, reducing confidence in the consistency of these endpoints.

38
39 A variety of symptoms were reported over the course of the study, with the majority
40 occurring in subjects administered AZM. The conclusion drawn in the Agency's Data
41 Evaluation Record (Doherty 1999) that none of the symptoms likely reflected effects of AZM is
42 not verifiable, nor is it credible.

43
44 Certain aspects of the statistical analyses are problematic. For the analyses of percentage
45 change each day from baseline (predosing) plasma and RBC cholinesterase activity in the
46 subjects receiving AZM, the choice of repeated measures ANOVA as the statistical approach

1 was appropriate given the structure of the data. This would have allowed each subject to be used
2 as his own control, but this does not appear to be the way the analyses were done. The pooled
3 baseline RBC and plasma ChE values were used as covariates rather than individual subject
4 values. This essentially ignores the value of the individual levels in increasing statistical power
5 by taking account of pre-dosing inter-individual differences in ChE levels. The other two types
6 of analyses conducted were not limited by this deficiency, but the analysis of within-subject
7 change from baseline is potentially the most sensitive analysis in this study.
8

9 In proposing to use the results of this study for dermal and inhalation risk assessment, the
10 Agency applied a dermal absorption factor of 42% based on a rat study and an inhalation
11 absorption factor of 100%. Bayer Crop Science commented that data are available supporting a
12 lower estimate of human dermal absorption (21.5%) but these data were not available for
13 evaluation by the Board. Another concern was whether the effects of a single oral dose
14 administered daily provide a reasonable estimate of the effects of the continuous dosing via all
15 routes of exposure that would be experienced by agricultural workers.
16

17 The major concern regarding this human study was it was designed to demonstrate a
18 NOAEL. No LOAEL could be established to validate the NOAEL because only one dose was
19 used. The fact that inhibition was not observed at the single dose used raised concern about
20 sensitivity of the methods used. The Agency was reassured by the laboratory's observation of
21 plasma and RBC cholinesterase inhibition in other studies of carbamates, including aldicarb,
22 methomyl, and oxamyl. Nevertheless, it is not possible to be sure if the absence of an effect in
23 this study was because the methods used were, for some reason, not up to the standards of those
24 used in the other studies, and therefore less sensitive.
25

26 HSRB Consensus and Rationale

27
28 The Board concluded that:

29
30 a. Data from the 28-day repeat oral dose study of AZM should not be used in developing
31 a point of departure for extrapolation of risk to workers exposed to AZM via the dermal and
32 inhalation routes.
33

34 b. Data from the 28-day repeat oral dose study of AZM cannot be used to inform the
35 inter-species factor in the cumulative risk assessment.
36

37 Despite the study's strengths, the HSRB had only limited confidence in the inferences
38 that can be drawn from this study, in large part because only one AZM dose was used. In order
39 to use these data, one must assume that the lab would have detected inhibition had it been present
40 at the single dose administered. The conclusions drawn could be correct, but this cannot be
41 determined with certainty. The conclusion that this study is not appropriate for developing a
42 point of departure for either the worker risk assessment or the cumulative risk assessment is
43 consistent with the recommendation of the NAS committee (NAS 2001) that the Agency should
44 not consider data from NOEL-only studies.
45

46 **Ethical considerations**

1
2 **Charge to the Board**

3
4 a. The Agency requests that the Board provide comment on the following:

5
6 Whether the informed consent materials – which refer to “the company” and “supervising
7 doctor” without further identification, and contain no discussion of who would benefit from the
8 research – should be considered significantly deficient relative to the ethical standards prevailing
9 when the study was conducted; and,

10
11 Whether the absence from the protocol of any discussion of the potential risks to subjects
12 or benefits to society of conducting the proposed research (as required by the 1996 Declaration
13 of Helsinki, Principle # 5, with which the research asserted compliance) should be considered
14 significantly deficient relative to the ethical standards prevailing when the study was conducted;
15 and

16
17 b. The Agency asks that the Board provide comment on the following, taking into
18 account all that is known about the ethical conduct of [this/each] study:

19
20 OPP’s conclusion that there is not clear and convincing evidence that the conduct of the
21 research was fundamentally unethical.

22
23 Whether there is clear and convincing evidence that the conduct of the study was
24 significantly deficient relative to the ethical standards prevailing when the study was conducted.

25
26 Brief Overview of the Study

27
28 As noted above, the study was conducted in 1999 by a contract research organization,
29 Inveresk Clinical Research of Edinburgh, Scotland, at its Elphinstone Research Centre in
30 Tranent. The study sponsor was the Bayer Corporation, Agricultural Division, of Stilwell,
31 Kansas. The protocol documents specifically state that the research was conducted in compliance
32 with the guideline for Good Clinical Practice, and in accordance with the guidelines of the
33 Declaration of Helsinki, 1964, as amended through 1996.

34
35 Critique of Study

36
37 The Board concurred with the factual observations of the strengths and weaknesses of the
38 study, as detailed in Carley (2006d). However, further comments are warranted with regard to
39 whether the documentation and process of study subject enrollment was sufficient to meet
40 prevailing standards of voluntary informed consent.

41
42 It is clear that the written documentation for informed consent failed to meet fully the
43 standards contained in the 1996 version of the Declaration of Helsinki.

44
45 In particular, the information sheet that was given to research subjects as an appendix to
46 the signed informed consent document contained the following statement under the subheading

1 Aim: “The aim of the study is to establish a recommended daily intake following long term
2 dietary exposure and to determine the most appropriate way of assessing workers exposure.” By
3 using the phrase “recommended daily intake,” this statement would appear to suggest that it is
4 actually desirable to consume a daily minimum amount of the study compound. This sentence is
5 thus misleading. However, other portions of the information sheet made it clear that the study
6 compound was a pesticide, and that consuming it is not good for people, thus leading the Board
7 to conclude that this one sentence by itself does not provide clear and convincing evidence that
8 the consent process as a whole was significantly deficient relative to the ethical standards
9 prevailing at the time the study was conducted.

10
11 HSRB Consensus and Rationale

12
13 The Board concluded that:

- 14
15 a) The azinphos-methyl human toxicity study failed to fully meet the specific ethical
16 standards prevalent at the time the research was conducted.
17
18 b) There was no clear and convincing evidence that the research was fundamentally
19 unethical--intended to seriously harm participants or that informed consent was not obtained.
20
21 c) There was no clear and convincing evidence of significant deficiencies in the ethical
22 procedures that could have resulted in serious harm (based on the knowledge available at the
23 time the study was conducted) nor that information provided to participants seriously impaired
24 their informed consent.

25
26 The Board determined that the informed consent materials – which refer to “the
27 company” and “supervising doctor”, without further identification, and which contain no
28 discussion of who would benefit from the research – did not fully comply with the ethical
29 standards at the time. However, the Board did not find clear and convincing evidence that the
30 level of non-compliance should be considered significantly deficient relative to those then-
31 prevailing ethical standards. With regard to the use of the terms “the company” and “supervising
32 doctor,” this type of language, though highly undesirable, is unfortunately not uncommon, even
33 at the present. With regard to the benefit issue, the Board determined that the consent materials,
34 while far from ideal, did in fact somewhat address this issue. They indicate, for example, that the
35 study may lead to further information about the risk that this compound poses to workers and
36 consumers.

37
38 The Board determined that the absence from the protocol of discussion of the potential
39 risks to subjects or benefits to society of conducting the proposed research (as required by the
40 Declaration of Helsinki, Principle # 5) should not be considered significantly deficient relative to
41 the ethical standards prevailing when the study was conducted. The potential risks and benefits
42 are described in the research subject information sheet/informed consent document. While not in
43 the protocol itself, they are nonetheless included in the materials provided to the ethics
44 committee (and to the potential research subject). Thus the ethics committee and the potential
45 research subjects had the opportunity to weigh the risks and benefits.
46

1 The Board concluded that there was no obvious reason why the Agency cannot rely on
2 the results of this study, as appropriate under current pesticide laws, given the absence of clear
3 and convincing evidence that the research was fundamentally unethical or significantly deficient
4 relative to the ethical standards prevailing when the study was conducted.

5 6 **5. DDVP**

7 8 **Charge to the Board**

9
10 Like AZM, DDVP is an organophosphate pesticide (OP) which elicits neurotoxicity
11 through the inhibition of acetylcholinesterase, via phosphorylation of the active site. The
12 Agency is conducting an aggregate (single chemical, multi-route, multi-duration) risk assessment
13 of DDVP. In addition, DDVP is a member of the OP common mechanism group and is thus
14 included in the cumulative (multi-chemical, multi-route) risk assessment for the OPs.

15 16 **5.1. Scientific considerations**

17
18 a. The Agency's WOE document and DER for DDVP describe the study design and results
19 of the DDVP repeat dose oral human study. The WOE document also discusses the Agency's
20 conclusions regarding the usefulness of this study in the aggregate risk assessment and in the
21 cumulative risk assessment for the OPs. For the single chemical risk assessment, the Agency has
22 concluded that the human study is sufficiently robust for developing a point of departure for
23 estimating dermal, incidental oral, and inhalation risk from exposure to DDVP in the single
24 chemical risk assessment. For the cumulative risk assessment, the Agency has determined that
25 results of the DDVP multi-dose human toxicity study do not support reducing the default 10X
26 inter-species factor in the cumulative risk assessment of the OPs.

27
28 Please comment on the scientific evidence that supports the conclusions for:

29
30 a) the Agency's conclusions for use of the human study for developing a point of
31 departure for estimating risk in the single chemical, aggregate risk assessment and

32
33 b) the Agency's determination that the human study cannot be used to reduce the
34 interspecies factor in the cumulative risk assessment.

35 36 **Board Response to the Charge**

37 38 Study Overview

39
40 A single blind, randomized placebo-controlled oral study was conducted with DDVP in
41 which six healthy male volunteers were administered a daily dose of DDVP at approximately
42 0.1mg/kg/day and three volunteers were administered a placebo (Gledhill 1997). DDVP was
43 dissolved in corn oil and administered daily in a gelatin capsule to fasted subjects (subjects did
44 not eat after midnight prior to each morning dose) for a total of 21 days. Blood samples were
45 collected for analysis of red blood cell (RBC) cholinesterase activity. For each subject, the
46 baseline level of RBC cholinesterase activity was established from seven pre-dose samples, and a

1 single blood sample was collected approximately 24 hours after dosing (just prior to dosing on
2 the following day) on days 1, 2, 4, 7, 9, 11, 14, 16 and 18 of the study. No blood samples were
3 collected on the last three days of the study (days 19-21). After cessation of dosing, subjects
4 returned approximately 1 week later (on or about day 25) for a follow-up analysis of RBC
5 cholinesterase activity. The criterion for withdrawal from the study was a inhibition of RBC
6 cholinesterase activity of 20% or more on a given day's analysis followed by a further decrease
7 in activity in the next successive sample. A linear mixed model was fitted to the cholinesterase
8 activity and significant differences were found within subjects and also between groups
9 (treatment versus placebo subjects) for pre-dose and post-dose cholinesterase activity.

10 11 Critique of Study

12
13 The scientific validity of the repeated dose study for DDVP was determined by the
14 relative strengths and weakness of the design and conduct of the study.

15
16 The strengths of the study were considered to be as follows:

- 17 • The repeat dosing paradigm affords the opportunity to more critically evaluate the
- 18 sustained nature of RBC cholinesterase inhibition associated with OPs.
- 19 • The analysis of RBC cholinesterase activity is fairly robust, given the assessment of
- 20 predose baseline values on 7 separate occasions. The baseline values were fairly
- 21 consistent both within and between subjects (16,000 – 21,000 IU).
- 22 • A low, but statistically significant effect was observed in RBC cholinesterase inhibition.

23
24 There are, however, numerous weaknesses of the study that are relevant to the utility of these
25 data in establishing a point of departure for use in single chemical, aggregate and cumulative risk
26 assessments for DDVP. These weaknesses included:

- 27
28 • A single dosage was used, preventing establishment of dose-response relationships.
- 29
30 • The sample size was small and included only male volunteers. In this regard, it is not
- 31 clear whether the study was properly powered, given that no sample size calculations
- 32 seem to have been used in order to arrive at the number of volunteers used in the study.
- 33
34 • RBC cholinesterase inhibition was determined only at 24 hours post dosing. Although
- 35 organophosphates are known to show prolonged inhibition of cholinesterase activity, it
- 36 was not clear whether the peak of RBC cholinesterase was established.
- 37
38 • The sponsor did not include the analysis of plasma cholinesterase measurements in the
- 39 study. The reason for this omission is not clear, and it was noted that virtually every
- 40 toxicological and exposure assessment study that has focused on cholinesterase inhibition
- 41 due to organophosphorus insecticides includes measurement of both plasma (serum)
- 42 cholinesterase and RBC cholinesterase. The combination of these measurements has been
- 43 used routinely for risk assessments by numerous regulatory agencies, including the EPA,
- 44 as well as by clinicians who diagnose pesticide poisonings. Current worker surveillance
- 45 programs in some states (California and Washington) require measurement of both
- 46 plasma and erythrocyte cholinesterase, and both measurements are used in combination

1 to initiate workplace investigations and remove workers from pesticide-related job
2 activities until enzyme activities return to baseline.
3

- 4 • Sampling was incomplete because the last day of analysis of enzyme inhibition was day
5 18 but dosing continued until day 21. It is not clear whether any additional decrease in
6 enzyme activity would have been observed on days 19-21. In the sampling schedule
7 outlined above, the maximum time gap between samples during the dosing period (days
8 1-21) was three days (between days 11 and 14). Logically then, an additional sample
9 would have been taken on day 21 or 22, the time in the study when maximum effects
10 might be observed. In this regard, two subjects presented with inhibition values of 22
11 and 23%, exceeding the 20% cut-off for subsequent analysis and consideration for
12 withdrawal on day 18 of the study. The lack of additional measurements in these subjects
13 during the remainder of the dosing period is scientifically inadequate and ethically
14 troublesome.
15
- 16 • It is neither clear nor convincing that steady state inhibition of RBC cholinesterase
17 activity was achieved in the study. The data suggest that the level of enzyme inhibition
18 was still increasing, particularly in the time period from 11-18 days, with a difference of
19 16% in activity on Day 18 compared to pre-dose values. Dosing was stopped on Day 21,
20 with no further analysis until approximately 4 days after cessation of dosing. Although
21 the difference in activity between day 18 post-dose and the average of the pre-dose
22 measurements was deemed to be “not-biologically significant” by the study sponsor, the
23 possibility that RBC cholinesterase inhibition may have reached or exceeded 20% by the
24 end of the study cannot be excluded.
25
- 26 • There was a lack of appropriate follow-up in all subjects in this study. As noted earlier,
27 two subjects presented with inhibition values of 22 and 23%, exceeding the 20% cut-off
28 for subsequent analysis and consideration for withdrawal on Day 18. However, there
29 were no additional analyses of RBC cholinesterase activity in these individuals on days
30 19-21 of dosing. Additionally, a blood sample was collected within 4 to 10 days
31 following completion of the 21-day dosing period for all subjects. The mean inhibition
32 of the enzyme observed at seven days following cessation of dosing was 83%, suggesting
33 little if any recovery of enzyme at this time, but these data were not analyzed for
34 statistical significance. Furthermore, one subject ended his participation in the study (day
35 25) with RBC cholinesterase inhibition greater than 20%. In all cases, there was no
36 additional follow-up of the subjects after the single post-dose analysis of cholinesterase
37 activity.
38

39 HSRB Consensus and Rationale

40
41 The Board concluded that:

- 42
43 a) The DDVP repeat dose human toxicity study was sufficiently robust for developing a point
44 of departure for estimating dermal, incidental oral, and inhalation risk from exposure to DDVP in
45 the single chemical risk assessment.
46

1 In specific response to the Agency's charge question, the HSRB concluded that there are
2 numerous technical limitations to the data obtained in the study. In addition to the weaknesses of
3 the study design and execution outlined above, the HSRB concluded that the evaluation of only a
4 single dose level of DDVP along with the omission of plasma cholinesterase measurements
5 greatly limit value of this study in terms of producing new knowledge that can be used in the
6 regulatory process and in medical diagnosis. Furthermore, study investigators have an obligation
7 to design a study that provides appropriate oversight of subjects until indications of the effects of
8 the administered dose are no longer present, and the Board considered that the design of this
9 study which continued intentional dosing without collection of blood samples for cholinesterase
10 analysis was not defensible scientifically. Following considerable discussion, the HSRB
11 concluded that the dosage evaluated in the repeat dosing human study can be used as a LOAEL
12 for the single chemical aggregate risk assessment for DDVP. Although a study using a single
13 dose level is not ideal for establishing a LOAEL, there was general consensus that RBC
14 cholinesterase is a well-characterized endpoint for compounds that inhibit acetylcholinesterase
15 activity and therefore, because the decreased activity in RBC cholinesterase activity observed in
16 this study was at or near the limit of what could be distinguished from baseline values, it was
17 unlikely that a lower dose would produce a measurable effect in RBC cholinesterase activity.
18 However, the HSRB strongly recommended that, in most cases, studies in human subjects
19 designed to define the NOAEL or LOAEL for a compound should include more than a single
20 dose level.

21
22 b) Results of the DDVP repeat-dose human toxicity study should not be used to support
23 reducing the default 10X inter-species factor in the cumulative risk assessment of the
24 organophosphate pesticides.

25
26 Although the data obtained in this study can be used to establish a point of departure for the
27 single chemical aggregate risk assessment, the consensus of the HSRB was that the scientific
28 limitations of the study design do not justify its use in the cumulative risk assessment. In
29 particular, the lack of sample collection through to the completion of dosing, the lack of a clear
30 demonstration that steady state inhibition of RBC cholinesterase inhibition had been achieved,
31 and the lack of any dose-response data limit the overall utility of the human data. Accordingly,
32 the HSRB recommended that the default interspecies uncertainty factor should be applied for the
33 cumulative risk assessment for DDVP.

34 35 **Charge to the Board**

36
37 b. The Agency has concluded that other human studies made available to the Board do not
38 provide sufficient scientifically sound information to warrant any reduction in the 10X inter-
39 species uncertainty factor used to derive reference dose values for DDVP based on animal
40 toxicity endpoints.

41
42 Please comment on the scientific evidence that supports these conclusions.

43 44 **Board Response to the Charge**

45 46 HSRB Consensus and Rationale

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The HSRB concluded that the other DDVP human toxicity studies available for the Board's consideration should not be used for determining a reduction in the 10X uncertainty factor to derive reference dose values for DDVP based on animal toxicity endpoints.

The Board's deliberations for this question focused on the acceptability or not of these human studies for the Agency's intended purposes, rather than on the evaluation of the magnitude of the interspecies uncertainty factor. Since the results of animal studies or benchmark dose modeling were not provided to the Board, the review was restricted to the evaluation of the acceptability of the human studies for the intended purposes. The Board concluded these other human studies did not warrant use in determining the inter-species uncertainty factor because of policy considerations (e.g., exposures involving infants, children, pregnant women) or due to uncertainty associated with the study itself (e.g., inappropriate duration, questionable route, exposure data of limited utility, lack of time course data on critical effect). Board concerns regarding these other human studies are provided in Table 1.

While the studies reviewed are not useable either because they are unacceptable due to policy considerations or due to uncertainty associated with the study itself, the Board also suggested that the Agency should take a critical hard look at those studies that are ethically acceptable (e.g., the ones that did not include exposures of children and pregnant women), to the extent to which they can be informative about the magnitude of the interspecies uncertainty factor. In making such evaluations, the Agency should consider taking into account: (1) applicability of Haber's law, (2) nature of the toxic moiety, and (3) the relative importance of interspecies differences in pharmacokinetics and sensitivity. Further, consideration should be given to the manner in which the data from the different species are pooled and analyzed (e.g., continuous response versus maximal response).

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Table 1. Evaluation of Specific DDVP Human Studies Provided to the HSRB

Reference	Observations/Concerns
Glendhill Acute toxicity study	Time course measurements of cholinesterase
Cavagna (1970)	Conducted in newborns
Slomka (1981)	Exposure through plastic bead (polyvinyl resin) formulation; rate of absorption not known; data provided as ranges
Cervoni (1969)	Efficacy trial; measurement of cholinesterase inhibition not the main focus; no assurance of consistent methods of measurement of inhibition
Leary (1974)	Exposure via insecticide strips; possibility children were involved, but composition of families not given; inhalation route – caveat of dose calculations; no information on consent of subjects
Smith (1972)	Performed in altitude chamber; short term inhalation study; no information on consent of subjects
Cavagna (1969)	Exposure via insecticide strips included pregnant women, infants and children in a hospital
Pena-Chavarria (1969)	Exposure of patients (most of them anemic, treated for parasite infection); not designed to identify NOAEL or LOAEL
Stein (1966)	Insufficient information on the time course of cholinesterase inhibition; doses not quantitated
Funckes (1963)	Study population included children, some below six years; time course evaluation on inhibition insufficient
Witter (1961)	Simulated aircraft cabin; one hour exposures; time course information lacking
Gratz (1963)	Inadequate information to evaluate dose and NOAEL; inadequate information about how many people exposed at each concentration; single application-based exposures

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5.2. Ethical considerations

Charge to the Board

a. The Agency requests that the Board provide comment on the following:

1 Whether references to the test material as a drug and other statements that could indicate
2 the study constituted medical research, that appear in the materials used to obtain informed
3 consent should be considered significantly deficient relative to the ethical standards prevailing
4 when the study was conducted;

5
6 Whether the administration of the test material for three additional days without
7 monitoring subjects' cholinesterase levels following the detection of cholinesterase inhibition
8 greater than 20 % in some subjects should be considered significantly deficient relative to the
9 ethical standards prevailing when the study was conducted; and

10
11 Whether the lack of medical surveillance of subjects, following the termination of dosing,
12 to establish the subjects' cholinesterase levels returned to normal should be considered
13 significantly deficient relative to the ethical standards prevailing when the study was conducted;
14 and

15
16 b. The Agency asks that the Board provide comment on the following, taking into account all
17 that is known about the ethical conduct of the Gledhill repeated dose study:

18
19 OPP's conclusion that there is not clear and convincing evidence that the conduct of the
20 research was fundamentally unethical; and

21
22 Whether there is clear and convincing evidence that the conduct of the Gledhill repeat
23 dose study was significantly deficient relative to the ethical standards prevailing when the study
24 was conducted.

25 26 **Board Response to the Charge**

27 28 Brief Overview of the Study

29
30 As noted above, the study was conducted in 1996 by the Zeneca Central Toxicology
31 Laboratory, Cheshire, United Kingdom. The study sponsor was the Amvac Chemical
32 Corporation. The protocol documents specifically state that the research was conducted in
33 compliance with the guideline of the Declaration of Helsinki, 1964, as amended through 1989.

34 35 Critique of Study

36
37 The Board concurred with the Agency's factual observations of the strengths and
38 weaknesses of the study, as detailed in Carley (2006e). However, further comments are
39 warranted with regard to: 1) whether the provisions in the protocol for monitoring the subjects
40 adequately protected them, and 2) whether the documentation and process of study subject
41 enrollment was sufficient to meet prevailing standards of voluntary informed consent.

42 43 1. Monitoring of Subjects While Receiving Study Compound

44
45 The protocol permitted subjects to continue to be administered the study compound for
46 three days following the detection of RBC cholinesterase inhibition of greater than 20%. Given

1 that study subjects were not monitored during most of the study (and were not full-time residents
2 at the study laboratory), it was possible that they might develop clinically significant symptoms
3 during this period and no one would be available to detect that and to treat them. The Board
4 concluded that this aspect of the study design was inadequate. However, given that there had
5 been a prior similar study at triple the dose used in this study, and that study had demonstrated
6 minimal symptoms, the Board could not conclude that there was clear and convincing evidence
7 that this element of the study design could have resulted in serious harm based on the knowledge
8 available to the investigators at the time.

9
10 2. Monitoring of Subjects at End of Study

11
12 The protocol also provided that, at the end of the study, inhibition levels would not be
13 retested for periods as long as seven days even though some subjects had RBC cholinesterase
14 inhibition of greater than 20%. During this period of time, due to the long-acting nature of the
15 study compound, subjects' cholinesterase inhibition levels could have been increasing, and thus
16 symptoms might have developed and, theoretically have become serious at a time when they
17 were being infrequently monitored. The Board determined that this aspect of the study design
18 was inadequate. However, as noted above, given that there had been a *prior* similar study at
19 triple the dose used in this study, and that study had demonstrated minimal symptoms, the Board
20 could not conclude that there was clear and convincing evidence that this element of the study
21 design could have resulted in serious harm based on the knowledge available to the investigators
22 at the time.

23
24 The inadequate monitoring during and at the end of the study undermines the scientific
25 value of the study. The fact that the research subjects were unsupervised during the study, and
26 the ascertainment of adverse event data was through self-report, renders the data establishing a
27 NOAEL suspect. The risk of a "false negative" conclusion is quite high. In addition, the failure
28 to achieve a steady state in the inhibition of acetyl cholinesterase activity may render these data
29 inadequate as well, and of limited utility. This fact, also noted in the Board's scientific review,
30 can negatively influence the prospective benefit to risk ratio.

31
32 3. Informed Consent

33
34 It is clear that the written documentation for informed consent failed to fully meet the
35 standards outlined of the 1989 version of the Declaration of Helsinki.

36
37 In particular, the information sheet that was given to research subjects as an appendix to
38 the signed informed consent document under the subheading Possible Adverse Events noted that
39 if "any symptoms possibly due to enzyme blocking did occur, they could rapidly be reversed by
40 a specific antidote." Although that statement is a correct statement of the scientific issue, it failed
41 to take account the fact that this study did not involve subjects remaining under 24 hour
42 supervision. For most of the time, the subjects would not be under any supervision. Given that
43 enzyme inhibition could build up over time due to the long-acting nature of the compound being
44 studied, symptoms could likely occur at a time when there was no one available to administer the
45 antidote. Telling the subjects that an antidote could rapidly reverse symptoms without explaining

1 that there were circumstances in which that antidote would not be readily available was not an
2 adequate way to inform subjects of the true nature of this risk.

3
4 In hindsight, it is clear that the documentation provided fails to rigorously meet the
5 standards of voluntary informed consent applicable to studies conducted in the United Kingdom
6 in 1996. However, there was not clear and convincing evidence that these deficiencies
7 knowingly and seriously impaired the informed consent process.

8
9 HSRB Consensus and Rationale

10
11 The Board concluded that:

12
13 a) The DDVP repeat dose oral human toxicity study failed to fully meet the specific
14 ethical standards prevalent at the time the research was conducted.

15
16 b) There was no clear and convincing evidence that the research was fundamentally
17 unethical--intended to seriously harm participants or that informed consent was not obtained.

18
19 c) There was no clear and convincing evidence of significant deficiencies in the ethical
20 procedures that could have resulted in serious harm (based on the knowledge available at the
21 time the study was conducted) nor that information provided to participants seriously impaired
22 their informed consent.

23
24 The Board determined that there was not clear and convincing evidence that references to
25 the test material as a drug and other statements that could indicate the study constituted medical
26 research, that appear in the materials used to obtain informed consent should be considered
27 significantly deficient relative to the ethical standards prevailing when the study was conducted.
28 These statements are highly undesirable, and should not be used as part of modern-day practice
29 in writing such consent materials. Nonetheless, it was unfortunately not uncommon for such
30 wording to have been used in consent materials in the past. In addition, other wording in the
31 consent materials clearly advised subjects that this was a study involving consuming an
32 insecticide. The Board, however, specifically rejects the arguments of the study sponsor that it
33 was appropriate to use the "drug" terminology because the consent materials were generic
34 materials used for a variety of studies. It also rejects the study sponsor's argument that because
35 the test material is sometimes used as a drug, it was appropriate to use that terminology in this
36 study even though this study in no way related to its use as a drug.

37
38 For the reasons discussed above, the Board determined that there was not clear and
39 convincing evidence that the administration of the test material for three additional days without
40 monitoring subjects' cholinesterase levels following the detection of cholinesterase inhibition
41 greater than 20 % in some subjects could have led to serious harm. There the study should not be
42 considered significantly deficient relative to the ethical standards prevailing when the study was
43 conducted.

44
45 (3) For the reasons discussed above, the Board determined that there was not clear and
46 convincing evidence that following the termination of dosing, the lack of medical surveillance to

1 establish the subjects' cholinesterase levels returned to normal could have led to serious harm.
2 Therefore the study should not be considered significantly deficient relative to the ethical
3 standards prevailing when the study was conducted.
4

5 The Board concluded that the Agency can rely on the results of this study, as appropriate
6 under current pesticide laws, given the absence of clear and convincing evidence that the
7 research was fundamentally unethical or significantly deficient relative to the ethical standards
8 prevailing when the study was conducted.
9

10 **6. Ethephon**

11 **Charge to the Board**

12
13
14 Ethephon is an organophosphorus compound that, upon absorption into plants, forms
15 ethylene gas which is an important component of the plant hormone complex. The Agency is
16 conducting an aggregate (single chemical, multi-route) risk assessment of ethephon.
17

18 Scientific consideration

19
20 The Agency's WOE document and DERs for ethephon describe the study design and
21 results of the ethephon repeat dose, oral, human toxicity studies. The WOE document also
22 discusses the Agency's conclusions regarding the usefulness of the human studies in the
23 aggregate, single chemical risk assessment. The Agency has concluded that the 28-day human
24 study is sufficiently robust to establish a point of departure for extrapolating acute and chronic
25 dietary risk.
26

27 Please comment on the scientific evidence that supports this conclusion.
28

29 **Board Response to the Charge**

30 Brief Overview of the Study

31
32
33 According to the Agency's weight of evidence document (WOE) (Kent 2006) ,
34 "ethephon is an organophosphorus compound that, upon absorption into plants, forms ethylene
35 gas which is an important component of the plant hormone complex. The production of ethylene
36 by plants occurs naturally as crops mature but it can be slow during periods of unfavorable
37 weather. Ethylene generated from ethephon is absorbed by the plant tissues and moderates the
38 growth process. It is not similar in structure to other organophosphate pesticides. Ethephon
39 registrants have submitted many toxicity studies to support the continued pesticidal use of the
40 chemical, and among these studies are two involving direct dosing of human subjects that OPP is
41 considering using to establish endpoints to assess risk from exposure to ethephon. The WOE
42 document compares the strengths and weaknesses of the human and animal toxicity studies and
43 discusses how the human studies fit in with the animal studies, i.e., are the human data consistent
44 with animal data in terms of types of effects and effect levels or are there notable differences
45 between animals and humans."
46

1 According to the WOE document, “as a strong acid, ethephon is corrosive to the skin and
2 eyes (Category I). In acute and subchronic neurotoxicity studies in rats, there were signs of
3 neurotoxicity at doses of 500 mg/kg and above (acute) and 300 mg/kg (subchronic). In dogs
4 there were no clinical signs of neurotoxicity evident at 75 mg/kg/day although plasma and RBC
5 cholinesterase were inhibited in animals at doses as low as 7.5 mg/kg/day.”
6

7 There are two human studies under consideration, both involve direct dosing of human
8 subjects and are repeated dose oral studies, one for 28 days duration (Reese 1972) and the other
9 for 16 days (Weir 1977).

10 11 28 Day Oral Toxicity Study

12
13 In a non-guideline oral toxicity human study (Reese 1972), ethephon was administered
14 by capsule in a powdered formulation (10% a.i. weight/weight) of hydrated silica and starch to
15 human adults (5/sex) at an average dose of 1.8 mg a.i./kg/day or placebo 0 mg/kg/day (3/sex) for
16 28 consecutive days. Administration of the test material or the placebo was given orally by
17 capsule divided into three daily doses. Each subject received 2 capsules postprandially for the
18 first two dosing periods; the third dose was given at the end of the work day in order to simulate
19 as closely as possible ingestion of the material as a crop residue. The capsules were dispensed
20 daily (5 days/week) by an assistant who observed the subjects while they took each capsule and
21 recorded solicited comments related to the ingestion of the material during the first 8 hours. The
22 subjects and the assistant did not know which capsules were placebo or the test material. Each
23 Friday, the subjects received a supply of capsules to be taken during the two day weekend. All
24 subjects were monitored regularly for adverse effects. Hematology measurements, clinical
25 chemistry and urinalysis were conducted initially and on Days 7, 14, 21, 28 and 2 weeks
26 following the last administration. ChE (plasma and RBC) determinations were conducted using
27 the pH/hr method (modified Michel method) initially, and on Days 1, 2, 7, 14, 28 and 2 weeks
28 after the last administration.
29

30 Critique of 28 Day Oral Toxicity Study MRID 00036510

31
32 The ethephon subjects, though exhibiting normal physical appearance and behavior,
33 developed symptoms of diarrhea and increased urination and bowel movement. Three females
34 and one male experienced a sudden onset of diarrhea or an urgency of bowel movement of 1-4
35 days duration during the first week of the compound administration; the one male also
36 experienced loose stools for 2 weeks. Another male complained of stomach cramps or gas for
37 the duration of the study. One subject in the ethephon group was asymptomatic. Five subjects
38 experienced increased urgency and frequency of urination, 4 of whom experienced this symptom
39 throughout the study. Two subjects experienced decreased appetite while a third (with diarrhea)
40 experienced increased appetite during the first 2 days of the study. It was stated that the
41 symptoms were “consistent with exposure to a cholinesterase inhibitor.” The WOE document
42 stated “the clinical signs observed in the study, diarrhea and increased frequency of urination, are
43 consistent with a cholinergic mode of toxicity, but other signs associated with a cholinesterase
44 inhibitor such as pinpoint pupils (miosis), sweating, runny nose, tearing, salivation, respiratory
45 distress and muscle fasciculation (twitching), were not seen [sic]; [and] the test subjects reported
46 symptoms as they considered appropriate, and it is difficult to interpret what the self-reported

1 symptoms mean in terms of a biological response.” In fact, it is unknown if symptoms or signs
2 associated with a cholinesterase inhibitor were seen; they may not have been monitored or
3 specific requests for their reported made by the investigators. However, these symptoms,
4 probably related to irritant effects, are important results.

5
6 The Agency’s Data Evaluation Record (Khasawinah) and the WOE documents both
7 stated “None of the placebo subjects had similar complaints.” This is probably not correct since
8 according to the investigators at least two of these controls were symptomatic as well some test
9 and control subjects also exhibited some atypical lymphocytes of unknown origin.

10
11 The WOE document states “Unexpectedly, plasma and RBC ChE activities were similar
12 or slightly higher than initial values in the test subjects. ([The] details of the ChE determinations
13 were not provided.)” However, the ChE levels were not related to the diarrhea occurring in the
14 subjects upon examination of the time of symptom occurrence and ChE data provided. The
15 WOE document states “No compound effects regarding hematology, clinical chemistries and
16 urinalysis were reported”, except the atypical lymphocytes of some.

17
18 It was concluded by the investigators of this study that the daily dose of 1.8 mg/kg/day is
19 the LOAEL for the oral ingestion of ethephon in human subjects. However, that is quite
20 uncertain since lower doses were not given.

21 22 16 Day Oral Toxicity Study

23
24 In the other non-guideline oral toxicity human study (Weir 1977), ethephon was
25 administered by capsule in a powdered formulation (2.5% a.i. weight/weight) of hydrated silica
26 and starch to human adults (10/sex) at an average dose of 0.5 mg a.i./kg/day or placebo 0
27 mg/kg/day (6 males and 4 females) for 16 consecutive days followed by a 29 day recovery
28 period (phase three). The dosing phase (phase two) was preceded with a 6 day period (phase
29 one) where all subjects received daily placebos. Administration of the test material or the
30 placebo was given orally by capsule divided into three daily doses. Each subject received two
31 capsules postprandially for the first two dosing periods; the third dose was given at the end of the
32 work day in order to simulate as closely as possible ingestion of the material as a crop residue.
33 The capsules were dispensed daily (5 days/week) by an assistant who observed the subjects
34 while they took each capsule and recorded solicited comments related to the ingestion of the
35 material for 8 hours. The subjects and the assistant did not know which capsules were placebo or
36 the test material in of the study phases. Each Friday, the subjects received a supply of capsules
37 to be taken during the two day weekend. All subjects were monitored regularly for adverse
38 effects. Hematology measurements and clinical chemistry were conducted on blood samples
39 collected during phases I, II and III while urinalysis was conducted on samples collected during
40 phase II. ChE (plasma and RBC) determinations were conducted using the pH/hr method
41 (modified Michel method) on blood collected during phases I, II, and III.

42 43 Critique of 16 day Oral Toxicity Study

44
45 No clinical symptoms were reported, but it is not known if this was an omission as the
46 report does not state that there were no symptoms or signs. Hematological, clinical chemistry

1 values and urinalysis parameters were comparable to the placebo controls. The updated
2 executive summary of the Agency Data Evaluation Record (Khasawinah) for the 16-day human
3 study stated: “Plasma and RBC ChE activities were significantly inhibited in both placebo and
4 test groups. Therefore, this finding makes any conclusions regarding the effects attributable to
5 ethephon administration impossible. This is in contradiction to the study authors overall
6 conclusion that a daily ethephon oral dosage of 0.5 mg/kg/day produces significant plasma ChE
7 inhibition, which is reversible in 15 days.” The study report states “Plasma cholinesterase was
8 significantly inhibited for the placebo group at 4, 8, 12, and 16 days during the dosage period. In
9 the test group, plasma cholinesterase was also depressed (greater than the placebo group) at 4, 8,
10 12, and 16 days during the dosage period. Recovery does occur as indicated by the 15 and 29
11 post dosage values. RBC cholinesterase inhibition was similar for both placebo and test groups
12 (slightly depressed in both groups) on days 4, 12, and 16 of the dosage period and day 15 of the
13 recovery period.” Investigators also say that the test group plasma values were significantly
14 different than the placebo group values.

15
16 It appears that either the method of testing cholinesterase was poor, or, less likely,
17 something else was depressing plasma cholinesterase in this latter study while increasing it in the
18 former study. Possibly, as the ethephon risk assessment team concluded, measures of blood
19 cholinesterase, either plasma or RBC, were not appropriate for endpoint selection for ethephon.

20
21 According to the WOE document (Kent 2006) “The OPP/HED RfD Peer Review
22 Committee determined on February 10, 1994 that the reference dose (RfD) should be based on
23 the 28-day study in human subjects (MRID 00036510). Clinical signs of toxicity were observed
24 at 1.8 mg/kg/day (only dose tested) and included diarrhea, urgency of bowel movements, urinary
25 urgency and stomach cramps.”

26
27 A Report from EPA, Office of Pesticide Programs, Health Effects Division, Hazard
28 Identification and Assessment Review Committee (HIARC) dated 4/1/98 states in reference to
29 the 28-day human study: “This human study is considered to be of better quality and the findings
30 more reliable than the 16-day human study previously used to define the RfD.” Since 1994, the
31 HIARC has repeatedly (8/9/94, 4/1/98, 7/15/02 and 11/5/02) selected the 28-day human study to
32 assess all relevant exposure scenarios, and at the last HIARC meeting, a weight -of -evidence
33 comment regarding endpoint selection was included: “HIARC had considered experimental
34 animal studies on ethephon and concluded this human study was most appropriate for selection
35 of the endpoint of toxicity for risk assessment. The clinical signs of toxicity in this human study
36 occurred at dose levels that were much lower than seen in experimental animals.” “Despite the
37 limitations of the study, and the equivocal nature of the reported findings, the HED ethephon risk
38 assessment team believes that, in the absence of data ruling out the observed effects as
39 inconsequential, the 28-day human study is the most appropriate study to use for assessing both
40 the acute and chronic dietary risks of ethephon.”

41
42 The WOE document stated: “A number of conclusions relevant to endpoint selection can
43 be drawn from the database of ethephon oral toxicity studies in humans and animals:
44 (1) ethephon is neurotoxic in animal studies only at high doses (300 mg/kg and above), (2)
45 ethephon inhibits blood cholinesterases, in particular plasma cholinesterase in animals and
46 humans at relatively low doses, (3) brain acetylcholinesterase is insensitive to ethephon in adult

1 animals. Accordingly, the team then debated whether to use effects in animals (LOAEL
2 approximately 80 mg/kg/day) or in humans (LOAEL of 1.8 mg/kg/day) for endpoint selection,
3 and opted as a matter of prudence to base endpoint selection on effects observed in humans.”
4 The Board concluded that this is much lower than that expected from the animal studies,
5 however, the second study informs and provides the level of uncertainty to accept this LOAEL
6 and use it as a POD.
7

8 The WOE document concluded by stating: “The point of departure for the dietary
9 assessments use of a LOAEL was applied to the POD to obtain a reference dose of 0.06
10 mg/kg/day for assessment of both acute and chronic dietary risks was 1.8 mg/kg/day (with an
11 uncertainty factor of 10x for intraspecies variability and a 3x factor for use of a LOAEL ...)”
12

13 HSRB Consensus and Rationale

14

15 The ethephon 16 day human oral toxicity study (second study) had no value on its own,
16 but it did inform the ethephon 28 day human oral toxicity study (first study); it allows one to
17 consider the results of the first study in light of the information gained there from. The Board
18 approved the first study for use in EPA risk assessments, emphasizing that the scientific quality
19 is not adequate on its own, and that the dose level administered was almost certainly not the
20 lowest dose at which adverse effects are likely to be observed. However, its use in lieu of the
21 animal studies, will result in greater protection for exposed human populations.
22

23 **6.2. Ethical considerations**

24

25 **Charge to the Board**

26

27 In its ethics review of this research, EPA documented that the study reports contained
28 very little information concerning the ethical conduct of the research and that the available
29 information raised no ethical concerns. The Agency asks that the Board provide comment on the
30 following, taking into account all that is known about the ethical conduct of each study:
31

32 OPP’s conclusion that there is not clear and convincing evidence that the conduct of the
33 research was fundamentally unethical; and
34

35 whether there is clear and convincing evidence that the conduct of the study was significantly
36 deficient relative to the ethical standards prevailing when the study was conducted.
37

38 **Board Response to the Charge**

39

40 Brief Overview of the Study

41

42 Two studies involving repeat high- and low-dose oral exposure of human subjects to
43 ethephon (ethrel) were evaluated. Both studies were performed by Litton Bionetics, Inc. of
44 Bethesda, Maryland. The studies were sponsored by Amchem Products, Inc., of Ambler,
45 Pennsylvania. Both studies were conducted prior to publication of U.S. regulations (drafted
46 1974, adopted by DHHS and FDA in 1981) or the Common Rule (adopted by EPA in 1991), so

1 the regulatory standards established by 45 CFR 46 with respect to conduct of research involving
2 human subjects cannot be applied in this retrospective evaluation. The low-dose study, however,
3 was performed after the 1972 Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) went
4 into effect, and is subject to the regulations contained therein. In particular, FIFRA Section
5 12(a)(2)(P) states that “[i]t shall be unlawful for any person ... to use any pesticide in tests on
6 human beings unless such human beings (i) are fully informed of the nature and purposes of the
7 test and of any physical and mental health consequences which are reasonably foreseeable
8 therefrom, and (ii) freely volunteer to participate in the test.” Both studies were also expected
9 to be in compliance with the ethical standards for research involving human subjects established
10 by the 1964 version of the Declaration of Helsinki.

11 Critique of Study

12
13
14 The Board concurred with the factual observations of the strengths and weaknesses of the
15 study, as detailed in the EPA’s Initial Ethics Review of Ethephon Human Study, dated March 17,
16 2006. However, further comments are warranted regarding: 1) whether the repeat high-dose
17 oral-exposure protocols used were designed to minimize risks to study participants; and 2)
18 whether the documentation and process of study subject enrollment was sufficient to meet
19 prevailing standards of voluntary informed consent.

20 21 1) Minimization of Risks to Study Participants

22
23 The two repeat high- and low-dose oral-exposure studies were performed in 1971 and
24 1973, respectively. The high-dose study involved repeated dosing, over a 28-day period, of 12
25 subjects (six men and six women) with 1.8 mg/kg body weight of ethephon per day by oral
26 ingestion. Four subjects (two men and two women) received placebo control. The study
27 documents provide no clear justification for the selection of the 1.8 mg/kg body weight dose,
28 other than passing reference to “a preliminary dose range study ... carried out in two human
29 subjects. (Litton 1972).
30

31 On weekdays, clinical symptoms were monitored continuously for eight hours following
32 ingestion of the test compound; after hours and on weekends, subjects were requested to self-
33 report any adverse clinical symptoms. There was no mention in the documents reviewed by the
34 Board that the investigators made provision for observation or rescue during these off hours.
35 Laboratory analyses, including analysis of blood and urine samples, were performed weekly
36 during the study and also two weeks after last exposure. A large number of study subjects
37 receiving the active compound exhibited clinical symptoms clearly associated with ethephon-
38 exposure, including diarrhea and other gastrointestinal disorders (4 participants), increased
39 frequency and urgency of urination (5 participants), and persistent abdominal pain (1
40 participant). Laboratory analyses also revealed transient elevations in blood glucose and
41 increased inhibition of plasma cholinesterase in subjects receiving ethephon. No stopping criteria
42 were listed in the final report, nor was there evidence that the investigators considered halting the
43 trial even after large numbers of study participants began to report adverse clinical symptoms.
44

45 The low-dose study involved repeated dosing, over a 16-day period, of 20 subjects (10
46 men and 10 women) with 0.5 mg/kg body weight of ethephon per day by oral ingestion. Ten

1 subjects (six men and four women) received placebo control. The study documents again provide
2 no clear justification for the selection of the 0.5 mg/kg body weight dose, other than indirect
3 reference to the high-dose study described above.
4

5 As before, clinical symptoms were monitored continuously for eight hours and self-
6 reported after hours and on weekends. Laboratory analyses were performed on days 1, 9, and 16,
7 and two weeks following last dose. No adverse clinical events were reported. Surprisingly, the
8 report even fails to mention that no adverse clinical events were seen, leading several Board
9 members to suspect that adverse event data was not rigorously collected.
10

11 2) Voluntary Informed Consent
12

13 Neither study contained sufficient information for the Board to adequately determine
14 whether or not the informed consent process used to enroll study participants met the standards
15 outlined in FIFRA Section 12(a)(2)(P) and the 1964 version of the Declaration of Helsinki. Each
16 of the final study reports provided to the Board contained only but the briefest of statement as to
17 the nature of the voluntary informed consent process:
18

19 “Sixteen human volunteers judged to be in good health as ascertained by pre-physical
20 interview, physical examination and selected clinical laboratory evaluations were chosen for the
21 study. All subjects were thoroughly briefed on the nature and present use of the test product, the
22 animal toxicity data, and the potential pharmacological side effects.”
23

24 “All subjects were thoroughly informed on the nature and present use of the test
25 substance, the animal toxicity data, potential pharmacologic side effects, and the results of the
26 prior study in human volunteers. Based on this briefing, consent documents were obtained from
27 all subjects.”
28

29 Although the 1973 low-dose study does contain reference to written consent documents,
30 these were unavailable for review.
31

32 Despite the paucity of documentation, several Board members expressed concern over
33 what they consider to be potential violations of accepted standards of informed consent:
34

- 35 • Circumstantial evidence suggests that employees of Litton Bionetics, Inc., may
36 have been enrolled as study participants. For example, there was mention of four
37 study participants “working in the same building.” The possible participation of
38 employees in a laboratory-directed research project raised issues of coercion and
39 voluntariness.
- 40 • In the high-dose study, a large number of study subjects receiving active
41 compound exhibited clinical symptoms clearly associated with ethephon-
42 exposure, including diarrhea and other gastrointestinal disorders, increased
43 frequency and urgency of urination, and persistent abdominal pain. Not a single
44 study participant, however, withdrew from the study over the 28-day monitoring
45 period, regardless of the severity of illness. This again raised questions about the
46 voluntary nature of participation in research.

1
2 HSRB Consensus and Rationale

3
4 The Board concluded that:

5
6 a) The both the 28 and 16 day oral human toxicity studies failed to fully meet the specific
7 ethical standards prevalent at the time the research was conducted.

8
9 b) There was no clear and convincing evidence that the research is fundamentally
10 unethical--intended to seriously harm participants or that informed consent was not obtained.

11
12 c) There was no clear and convincing evidence of significant deficiencies in the ethical
13 procedures could that have resulted in serious harm (based on the knowledge available at the
14 time the study was conducted), nor that information provided to participants seriously impaired
15 their informed consent.

16
17 Although these two studies appear to lack justification for the dose selection, as well as
18 clear stopping criteria to prevent serious harm to research participants, the Board does not
19 believe that there was clear and convincing evidence that these studies could have resulted in
20 serious harm based on the knowledge available to the investigators at the time.

21
22 There were indications of informed consent deficiencies. Nevertheless, given the paucity
23 of information provided there was no evidence that the voluntary informed consent process used
24 in these two studies failed to meet the few regulatory and ethical standards applicable to research
25 conducted in 1971-1973.

26
27 The Board concluded that there was no obvious reason why the Agency cannot rely on
28 the results of this study, as appropriate under current pesticide laws, given the absence of clear
29 and convincing evidence that the research was fundamentally unethical or significantly deficient
30 relative to the ethical standards prevailing when the study was conducted.

31
32 **7. Hydrogen Cyanide**

33
34 **Charge to the Board**

35
36 When sodium cyanide is used as a fumigant, hydrogen cyanide is generated by
37 acidification. Because residues of HCN may remain on fumigated citrus, the Agency is
38 conducting an acute dietary risk assessment of hydrogen cyanide.

39
40 **7.1. Scientific considerations**

41
42 The Agency's WOE document describes a lack of data appropriate for developing an
43 acute dietary risk assessment for hydrogen cyanide. The WOE and DER present the results from
44 a clinical trial with amygdalin and the usefulness of this clinical trial in the acute dietary risk
45 assessment for hydrogen cyanide. The Agency has concluded that the clinical trial is appropriate
46 for establishing a point of departure in the acute dietary risk assessment for hydrogen cyanide.

1
2 Please comment on the scientific evidence that supports this conclusion.
3

4 **Board Response to the Charge**

5 Brief Overview of the Study 6 7

8 The clinical trial of amygdalin for treatment of cancer (Moertel et al., 1982), which
9 included the preliminary pharmacology and toxicology study (Moertel et al., 1981), involved the
10 administration, under controlled circumstances, of known amounts of amygdalin to patients with
11 terminal cancers for which no known treatments were available. Amygdalin was administered
12 intravenously for 21 days (4.5 g/m²), followed by oral maintenance therapy of 500 mg (three
13 times per day), plus a “metabolic therapy” consisting of diet, pancreatic enzymes, and vitamins.
14 Most patients were administered the standard dose regimen, but a small number received a high
15 dose regimen.
16

17 Clinical signs and symptoms consistent with cyanide toxicity (nausea, vomiting,
18 headache, dizziness, mental obtundation) were observed in some patients following a single 500
19 mg oral dose of amygdalin. The conclusions regarding toxicity were based largely on several
20 patients who developed such symptoms when they took two 500 mg doses too close together or
21 ate foods such as raw almonds that are rich in β -glucosidase, an enzyme that causes the release of
22 cyanide from amygdalin. In such patients, the symptoms typically resolved when oral amygdalin
23 was discontinued, the dose was reduced, or the patients stopped eating the critical foods. In the
24 pharmacology and toxicology study, the responses of two patients challenged with almonds
25 provided further support for the conclusion that the symptoms reflected cyanide toxicity. In
26 some patients, the appearance of symptoms was associated with relatively high blood cyanide
27 levels, which in some instances approached levels observed in human fatalities (>3 μ g/mL). A
28 dose of 500 mg was interpreted as a minimal effect level but not as a NOAEL.
29

30 Critique of Study 31

32 The trial had several strengths. The sample size was large, including 178 patients. Blood-
33 cyanide levels were assessed several times, at completion of intravenous (i.v.) treatment, 48
34 hours after initiation of oral treatment, and at every subsequent evaluation. Measurements were
35 made two hours after the first morning dose of oral amygdalin, when the maximum elevation in
36 cyanide was anticipated. The trial employed a good operational definition of the clinical
37 outcome of interest in the trial, which was tumor progression.
38

39 The trial also had several weaknesses. First, it was not blinded and did not include a
40 placebo group. Second, insofar as the primary aim was to assess the effectiveness of amygdalin
41 as a treatment for advanced cancer, the assessment of potential cyanide toxicities was not as
42 systematic as the assessment of tumor progression. As in any clinical trial of drug efficacy, side
43 effects were recorded, however, these side effects were examined in relation to whole-blood
44 cyanide levels. Oral dosing with amygdalin did not begin until 21 days of i.v. treatment had been
45 completed. It is unlikely that this introduced a bias, however. In contrast to delivery by oral
46 route, amygdalin delivered i.v. is largely excreted unchanged in urine without conversion to

1 cyanide. Whole blood cyanide levels were undetectable following i.v. dosing, making it likely
2 that the rise observed following oral dosing was not confounded by the prior i.v. dosing. Third,
3 patients who participated in the study were terminally ill with cancer. Although the inclusion
4 criteria required that patients be in "good general condition" and ambulatory, they are,
5 nevertheless, likely to represent a particularly sensitive subgroup, thus providing a conservative
6 estimate of the hydrogen cyanide dose at which toxicities occur. Fourth, all clinical signs and
7 symptoms of toxicity (nausea, vomiting, headache, dizziness, mental obtundation, and
8 dermatitis) were observed with nearly equivalent frequencies during i.v. and oral dosing. Many
9 of these signs and symptoms might be increased in terminal cancer patients anyway, and thus
10 unrelated to amygdalin or cyanide. Moreover, if these were indications of toxicity related solely
11 to elevations in blood cyanide, they would not be expected to have occurred with equal
12 frequency during i.v. and oral dosing. The data from the natural experiments within the trial,
13 such as when the doses of individual patients who developed symptoms were changed or the
14 patients were challenged, provides the most persuasive evidence of an increased risk of cyanide
15 toxicity in patients administered amygdalin.

16 HSRB Consensus and Rationale

17
18
19 The Board concluded that data from the amygdalin trial could be used for establishing a
20 point of departure in the acute dietary risk assessment for hydrogen cyanide.

21
22 Despite its limitations, this study provides the best data we are likely to ever have on
23 which to establish a POD for this purpose. Given the severity of the effect, the steepness of the
24 dose-response relationship, and the apparent inter-individual differences in response to a given
25 dose, it would be imprudent to undertake an intentional dosing study of healthy humans in order
26 to establish a LOAEL and NOAEL for hydrogen cyanide.

27 28 **7.2 Ethical considerations**

29 30 **Charge to the Board**

31
32 In its ethics review of this research, EPA did not identify any deficiencies with respect to
33 the ethical conduct of this research. The Agency asks that the Board provide comment on the
34 following, taking into account all that is known about the ethical conduct of this study:

35
36 OPP's conclusion that there is not clear and convincing evidence that the conduct of the
37 research was fundamentally unethical; and

38
39 whether there is clear and convincing evidence that the conduct of the study was
40 significantly deficient relative to the ethical standards prevailing when the study was conducted.

41 42 **Board Response to the Charge**

43 44 Brief Overview of the Study

1 Two related studies are being evaluated. Both were sponsored by the National Cancer
2 Institute. The first study was a pharmacologic and toxicological study involving six subjects
3 which took place at the Mayo Clinic (Rochester, MN), with the results being published in the
4 Journal of the American Medical Association in 1981 (the Board assumed the study was
5 conducted 1979-1980). Although the published report does not describe the ethical standards to
6 which the researchers adhered, the Board assumed that as a federally-funded study, the research
7 would have been subject to the then-current version of the federal DHHS [DHEW] regulations
8 (45 CFR Part 46) that would later become the Common Rule. The second study was essentially a
9 Phase II follow-up to the first study, involving 178 subjects who were enrolled at four sites
10 (Mayo Comprehensive Cancer Center, Mayo Clinic, Rochester, MN; UCLA Johnson
11 Comprehensive Cancer Center, Los Angeles, CA; Memorial Sloan-Kettering Cancer Center,
12 New York, NY; and University of Arizona Cancer Center, Tucson, AZ). The results of that
13 second study were published in the New England Journal of Medicine in 1982. That article also
14 does not specify the specific ethical rules under which the study was conducted, and this Board
15 again assumes that it would have been subject to the then-current version of the predecessor
16 regulations (DHHS [DHEW], 45 CFR Part 46) to the current Common Rule.

17 18 **Critique of Study**

19
20 The Board concurred with the factual observations about the study, as detailed in Carley
21 (2006h).

22
23 In particular, the Board concurred with the key observation regarding the difference
24 between this study and the other studies that the Board has heretofore been asked to review. This
25 study involved testing a compound for its safety and efficacy in treating a medical problem. As
26 such, the ethical evaluation of this study was significantly different from that of a study
27 involving exposing subjects to a pesticide. A major consideration in evaluating the ethics of such
28 a study involved determining whether the two arms of the study were in clinical equipoise, a
29 concept that was only in its earliest stages at the time of this study. Nonetheless, it does appear
30 that this study would meet even the modern-day understanding of that concept. All of the
31 subjects were persons with histologically-proven cancer for which no standard treatment was
32 known to be available. The subjects received either a standard dose or a high dose of amygdalin
33 (Laetrile). Prior to this study being conducted, there was substantial uncertainty regarding
34 whether this compound might indeed be a safe and effective treatment for many forms of cancer.
35 As stated in the New England Journal of Medicine article, it had “eclipsed any other unorthodox
36 therapy ever used for any disease in our time.” Thus, conducting this study served to answer an
37 important medical question of that time period.

38
39 Moreover, the published report of the second study indicated compliance with all of the
40 procedural protections that would be required today. It notes that all “patients were fully
41 informed about the experimental and unorthodox nature of the treatment program as well as any
42 possible alternative treatment available to them. A signed form giving informed consent,
43 approved by the Human Subjects Committee at each of the four participating centers, was
44 obtained from each patient.” (The actual forms were not available for review by the Board.) The
45 article further noted that “the methods of this trial were entirely comparable to those employed in

1 studies of any new agent being developed and tested for cancer treatment through more
2 traditional channels.”

3 4 HSRB Consensus and Rationale

5
6 The Board concluded that:

7 a) The hydrogen cyanide human oral toxicity study appeared to meet the specific ethical
8 standards prevalent at the time the research was conducted.

9
10 b) There was no evidence that the research was fundamentally unethical--intended to
11 seriously harm participants or that informed consent was not obtained.

12
13 c) There was no evidence of significant deficiencies in the ethical procedures that could
14 have resulted in serious harm (based on the knowledge available at the time the study was
15 conducted) nor that information provided to participants seriously impaired their informed
16 consent.

17
18 Although the Board based its determinations only on the two published articles, it did not
19 find any ethical flaws in the conduct of these two studies. The article described appropriate
20 informed consent procedures. The risks and potential benefits of the dosing were appropriate for
21 a medical treatment study for this particular cancer population.

22
23 The Board concluded that there was no obvious reason why the Agency cannot rely on
24 the results of this study, as appropriate under current pesticide laws, given the absence of clear
25 and convincing evidence that the research was fundamentally unethical or significantly deficient
26 relative to the ethical standards prevailing when the study was conducted.

27 28 **8. Amitraz**

29 30 **Charge to the Board**

31
32 Exposure to amitraz can result in neurotoxicity as evidenced by clinical signs such as
33 ataxia, ptosis, emesis, labored respiration, muscular weakness, tremors, hypothermia and
34 bradycardia. The Agency is conducting an aggregate (single chemical, multi-route) risk
35 assessment of amitraz.

36 37 **8.1. Scientific considerations**

38
39 The Agency's WOE document and DERs for amitraz describe the study design and
40 results of the amitraz acute oral and dermal toxicity human studies and the human metabolism
41 study. The WOE document also discusses the Agency's conclusions regarding the usefulness of
42 the human studies in the single chemical risk assessment for acute and chronic oral exposures in
43 addition to dermal and inhalation exposures of various durations. For oral exposure, the Agency
44 has concluded that the combined results from the single oral dose study and human metabolism
45 study establishes a dose response relationship in human subjects and that the single oral dose
46 study is appropriate for developing a point of departure for acute and chronic dietary risk, short-

1 term oral exposure, and inhalation exposures of various durations. The Agency has further
2 concluded that the human dermal study is appropriate for developing a point of departure for
3 dermal exposures of various durations.

4
5 Please comment on the scientific evidence that supports these conclusions.

6 7 **Board Response to the Charge**

8 9 Overview

10
11 The HSRB was asked to comment on the scientific evidence that supports the following
12 conclusions:

13
14 1. For oral exposure, the Agency concluded that the combined results from the
15 single oral dose study and human metabolism study establish a dose-response relationship in
16 human subjects, and that the single oral dose study is appropriate for developing a point of
17 departure for acute and chronic dietary risk, short-term oral exposure, and inhalation exposures
18 of various durations.

19
20 2. The Agency further concluded that the human dermal study is appropriate for
21 developing a point of departure for dermal exposures of various durations.

22
23 Amitraz [N'-(2,4-dimethylphenyl)-N-[(2,4-dimethylphenyl)imino]methyl]-N-
24 methylmethanimidamide] is an insecticide/acaricide with wide range of registered uses in the
25 United States. The toxicity profile for amitraz has not been characterized completely, especially
26 developmental and reproductive effects. Neurotoxicity is considered the most sensitive effect
27 resulting from exposure to amitraz. Neurotoxicity has been demonstrated in multiple species
28 (baboon, cat, dog, mouse, rat and rabbit). Clinical signs include central nervous system
29 depression, ataxia (loss of coordination), ptosis (droopy eyelids), emesis (vomiting), labored
30 respiration, muscular weakness, tremors, hypothermia and bradycardia (slow heartbeat). Similar
31 signs are seen in humans. The Agency's evaluation of amitraz included three human studies: a
32 1984 oral dose metabolism study (Campbell 1984), a 1992 oral dose tolerance study (Cass 1992)
33 and a 1997 dermal dose tolerance study (Langford 1997).

34 35 Critique of Oral Dose Metabolism Study (1984)

36
37 Radio-labeled amitraz was orally administered to rats, mice, baboons and humans. The
38 two human volunteers received a single 0.25 mg/kg dose of amitraz by capsule. This dose caused
39 dry mouth, drowsiness, disorientation, decreased temperature, bradycardia, and slightly pale
40 appearance in both subjects, persisting up to 12 hours after dosing. One subject fell asleep for 6
41 hours following dosing. Psychomotor testing was not included in the protocol. Approximately
42 44% of the dose was excreted within 12 hours, with 63% excreted within 24 hours. Study
43 deficiencies included the small number of subjects (N=2), testing of males only, and no control
44 group. Sex differences in the repeat dose animal studies have shown that females are more
45 sensitive than males. The study investigators concluded that humans were more sensitive to
46 amitraz than were other species included in the metabolism study.

1
2 The occurrence of multiple signs and symptoms in both subjects and the continued
3 bradycardia up to 12 hours post-dosing suggest that the 0.25 mg/kg dose was not the lowest
4 adverse effect level for amitraz in humans. Because there was only one dose level, it was not
5 possible to glean any information regarding dose-response from this study. Incomplete excretion
6 of the amitraz metabolites at 24 hours post-dosing suggests that some portion of the dose was
7 present on the day following dosing, albeit not at levels sufficient to produce the frank signs and
8 symptoms observed on the day of dosing.

9
10 Critique of Single Oral Dose Study (1992)

11
12 The design was a single oral dose double-blind tolerance study. The primary objective of
13 this study was to determine the tolerance of male volunteers to a single dose of amitraz at two
14 different dose levels (0.0625 and 0.125 mg/kg), with a placebo control. Six subjects completed
15 the study. Each subject received each of the doses and the placebo. Dosing events were separated
16 by at least 14 days for each subject. The dosing regimen was thoughtfully constructed, beginning
17 with the lower dose, and moving to the higher dose when it was observed that the lower dose
18 was well tolerated. The subjects were admitted to a clinical pharmacology unit the evening
19 before each dosing and remained there for 36 hours. Complete physical examinations were
20 conducted before and after the study. The subjects were followed for at least three weeks
21 following the study. The most sensitive neurologic endpoints measured in this study were two
22 psychomotor performance tests: choice reaction time and critical flicker fusion threshold. The
23 psychomotor performance tests were administered pre-dose, 2.5 hours, and 8 hours following
24 dosing. The report provides tabular and graphic representation of the results of these tests, but no
25 statistical analyses were conducted. Study deficiencies included the small number of subjects
26 (N=6), and the testing of males only. The study investigators concluded that no effects were
27 observed from the single oral dose in any of the subjects.

28
29 The major concern with this study was whether “no effect” was truly observed. The study
30 report was deficient in its description of the psychomotor performance testing, and in the
31 rationale for selecting these tests. There were no descriptions of the specific procedures followed
32 for the psychomotor tests, no standard operating procedures, and no quality assurance
33 documentation. Also, there was no evidence provided in the report to indicate that these tests
34 were the most sensitive available. A review of the current scientific literature, and discussions
35 with leading scientists in this area of expertise would permit the Agency to determine if these
36 psychomotor test results are adequate to characterize a “no effect” level in regard to
37 neurotoxicity for amitraz.

38
39 Critique of Dermal Toxicity Study (1997)

40
41 This study was a double-blind sequential dosing study with a randomized crossover and a
42 placebo group. The study aims were to 1) establish a no effect level for acute effects following
43 repeated dermal doses of amitraz, 2) produce data that would permit calculation of margins of
44 safety for agricultural workers. Eight male volunteers were given a total dose of either 0, 8, 16 or
45 24 mg/kg amitraz, applied dermally as four equal doses of 0, 2, 4 or 6 mg/kg in an aqueous 1:1
46 (w/w) slurry every 2.5 hours over 10 hours. Starch, 1:1 (w/w) in water, was used as a placebo.

1 Each of these amitraz experiments involved a timed series of dosing events (0, 2.5, 5.0, and 7.5
2 hours), followed by a washing event at 10 hours. Each dose within an experiment was applied to
3 a different 20 cm² skin surface area, so the total skin surface treated in each experiment was 80
4 cm². Thus, skin loadings for the three experiments were 7,000, 14,000, and 21,000 µg/cm²,
5 corresponding to the three dose levels (8 mg/kg, 16 mg/kg, 24 mg/kg), assuming a body weight
6 of 70 kg. No differences were observed between treatment and placebo for any of the measured
7 endpoints. No urinary metabolite monitoring was conducted to confirm that amitraz had been
8 absorbed. The study report is deficient in its description of the psychomotor performance testing,
9 and in the rationale for selecting these tests. There is no description of the specific procedures
10 followed, no standard operating procedures, and no quality assurance documentation. Also, there
11 is no evidence provided in the report to indicate that these tests were the most sensitive available.
12 A review of the current scientific literature, and discussions with leading scientists in this area of
13 expertise would permit the Agency to determine if these psychomotor test results are adequate to
14 characterize a “no effect” level in regard to neurotoxicity for amitraz.

15
16 The primary deficiency of this study is that it does not provide a realistic worker
17 exposure scenario; that is, the exposures of the subjects in these experiments did not correspond
18 to exposures likely to be seen among workers. Large amounts of amitraz (140-420 mg) were
19 applied to a relatively small skin surface area (80 cm²) in the experiments, whereas we typically
20 see much larger skin surface areas exposed to smaller amounts among workers. For example, the
21 hands, a skin surface commonly exposed to pesticides, have a total surface area of 990 cm², or 12
22 times greater than the exposed skin surface area in this experiment (USEPA 1997). If both the
23 hands and forearms were exposed, the total exposed surface area would be 3,900 cm², or 49
24 times the surface area exposed in this study. This discrepancy is important, as it has a major
25 impact on the amount of compound that can be absorbed within a defined time period. Dermal
26 dosing studies require careful consideration of three factors: mass applied to the skin, surface
27 area treated, and the duration of exposure. These three factors are typically used to determine the
28 flux of a chemical through the skin (e.g., mg/cm²/hr). Percent absorbed is highly dependent on
29 the skin loading, or mass applied per unit area (e.g., mg/cm²). The relationship between skin
30 loading and percent absorbed was made clear in a review of studies in the rat by Zendzian
31 (2000). In the case of azinphos-methyl, for example, a loading of 3 nanomole/cm² for 10 hours
32 resulted in 23% absorption, a loading 10-fold higher (29 nmolesM/cm²) resulted in 15%
33 absorption, and a further 10-fold increase in loading (293 nmoles/cm²) resulted in only 2.9%
34 absorption. These azinphos-methyl loadings, expressed as mass per unit area, were 0.95, 9.2, and
35 93 µg/cm², respectively. In earlier dermal dosing studies involving humans, radio-labelled
36 pesticides were applied at loadings of 4 and 40 µg/cm² (Feldmann and Maibach 1974). It is
37 evident that the skin loadings in the amitraz study were several orders of magnitude higher than
38 those used in the above cited studies. The key point is that chemical loadings on small areas of
39 skin result in relatively small amounts of chemical absorbed. In this dermal toxicity study, the
40 loadings were so high that the three “dose” levels were essentially equivalent in terms of dermal
41 absorption potential.

42
43 If it is assumed that the flux of amitraz in these experiments was approximately 2
44 µg/cm²/hr, as has been estimated from a study of chlorpyrifos, a lipophilic insecticide with a
45 similar molecular weight (Nolan et al. 1984), then the total mass absorbed in the amitraz
46 experiments over 10 hours would have been approximately 1,000 µg (1 mg), or an absorbed dose

1 of 0.014 mg/kg. If this same flux were applied to an exposure to the hands and forearms of a
2 worker, the total mass absorbed would be approximately 50,000 µg (50 mg), or an absorbed dose
3 of 0.71 mg/kg, well above both the 0.125 mg/kg single oral dose “no effect” level, and the 0.25
4 mg/kg single oral dose that elicited multiple signs and symptoms of neurotoxicity. It is for this
5 reason that the amitraz dermal toxicity study cannot be considered a “no effect” study for risk
6 assessment purposes.

7
8 HSRB Consensus and Rationale
9

10 a. The Board concluded that the results from the single oral dose study are informed by
11 the human metabolism study such that the single oral dose study is appropriate for developing a
12 point of departure for acute and chronic dietary risk and short-term oral exposure.
13

14 The appearance of overt signs and symptom of toxicity in the 1984 oral metabolism study
15 at a dose of 0.25 mg/kg provides some confidence that the highest dose (0.125 mg/kg) in the
16 1992 single oral dose study represent a no observable adverse effect level (NOAEL) for an acute
17 dietary exposure to amitraz. No effects of neurologic toxicity were observed in any of the study
18 subjects at 0.125 mg/kg. This conclusion rests on the assumption that the psychomotor tests
19 applied to the subjects are the most sensitive endpoints for neurotoxicity, and that they were
20 conducted properly. There was considerable uncertainty regarding these psychomotor tests, as
21 the procedures used were not described in the study, and on quality assurance information was
22 provided. Thus, the findings from this study should be used with caution for risk assessment
23 purposes.
24

25 b. The Board concluded that the combined results from the single oral dose study and the human
26 metabolism study were not appropriate for developing a point of departure for chronic dietary
27 risk, short-term oral exposure, or inhalation risk.
28

29 A single oral dose of 0.25 mg/kg elicited frank symptoms of toxicity in both subjects in
30 the 1984 metabolism study. Multiple signs and symptoms were observed in the subjects for
31 several hours post-dosing, and at least one sign (bradycardia) was still present at 12 hours post-
32 dose. Excretion of radio-labeled metabolites was incomplete (62%) at 24 hours. The proposed
33 NOAEL is only one-half the dose used in the 1984 study. Thus, subjects exposed to repeated
34 daily doses, as would occur in short-term or chronic oral dose studies, would achieve a
35 cumulative dose of 0.25 mg/kg within 48 hours. It seems quite possible that repeated short-term
36 oral exposures or chronic oral exposures would elicit at least some effects in humans within a
37 very few days. Thus, the available data do not demonstrate a no-effect level for multi-day oral
38 exposures. In regard to inhalation exposures, absorption through the respiratory tract is generally
39 more efficient than absorption through the gastro-intestinal tract. It is quite possible that adverse
40 effects would have been observed in an inhalation exposure study that delivered 0.125 mg/kg as
41 a single dose. Also, an oral dose study would not capture adverse effects unique to exposure by
42 the inhalation route. Thus, the available data do not demonstrate that the oral NOAEL and the
43 inhalation NOAEL are equivalent.
44

45 c. The majority of the Board concluded that the human dermal study was not appropriate
46 for developing a point of departure for dermal exposures of various durations.

1
2 The HSRB members did not consider the dermal toxicity study to be a valid scientific
3 study for risk assessment because: 1) the study involved extremely high loadings (mass per unit
4 surface area) of amitraz on the skin, making the three dose levels used in the study very nearly
5 equivalent in regard to dermal absorption potential, and not equivalent to worker exposure
6 scenarios; 2) the study did not demonstrate an effect (i.e., the study was equivalent to a
7 “NOAEL-only” study); and 3) there was no corroborating evidence to demonstrate that an
8 internal dose had been delivered to the subjects by the dermal route. One member of the HSRB,
9 while agreeing with these scientific criticisms, considered that the study might still be of use to
10 the Agency.

11 **8.2. Ethical considerations**

13 **Charge to the Board**

14 a. The Agency requests that the Board provide comment on the following:
15
16

17
18 With respect to the Campbell (1984) research, whether the lack of medical surveillance of
19 subjects, following the termination of dosing, to establish that subjects’ signs of adverse effects
20 had returned to normal should be considered significantly deficient relative to the ethical
21 standards prevailing when the study was conducted; and
22

23 With respect to the Cass (1992) and the Langford (1998) studies, whether references to
24 the test material as a drug and other statements that could indicate the study constituted medical
25 research, that appear in the materials used to obtain informed should be considered significantly
26 deficient relative to the ethical standards prevailing when the study was conducted; and
27

28 b. The Agency asks that the Board provide comment on the following, taking into account all
29 that is known about the ethical conduct of each study:
30

31 OPP’s conclusion that there is not clear and convincing evidence that the conduct of the
32 research was fundamentally unethical.
33

34 Whether there is clear and convincing evidence that the conduct of the study was
35 significantly deficient relative to the ethical standards prevailing when the study was conducted.
36

37 **Board Response to the Charge**

38
39 The compound amitraz is an alpha-two-adrenergic agonist that produces sympatholytic
40 effects leading to sedation and other neurological signs and symptoms, as well as cardiovascular
41 depression with a drop in blood pressure and heart rate. The studies under ethical review include
42 two metabolic studies in 1984 using C¹⁴ radio-labeled isotope technology, a 1992 oral dosing
43 study, and a 1998 dermal absorption study.
44

45 The two metabolic studies were performed in 1984 and involved two volunteer subjects.
46 The oral dosing study was performed in 1992 and involved six research subjects (with an

1 additional (1) subject withdrawn for an unrelated rash). The oral dosing study was a double-
2 blind, randomized, placebo-controlled, crossover study involving two different doses. The oral
3 dosing study progressed in three phases, escalating from the lower dose to the higher dose. The
4 protocol stated that the research was in compliance with the 1989 version of the Declaration of
5 Helsinki, an early version of good clinical practice (GCP) guidelines promulgated by the EMEA
6 (111-3976-88-EN), the ABPI Guidelines for Medical Experiments in Non-Patient Human
7 Volunteers (1988, 1990), the 1986 RCP Guidelines on Research Using Healthy Volunteers, the
8 FDA Compliance Program for Drugs and Biologics and the GCP guidelines from Japan that
9 were current in 1992 (Notification No. 874 from the Ministry of Health and Welfare). The
10 approval of the research ethics committee was given on November 20, 1991, but no information
11 was provided about the substance and process of that review.
12

13 The dermal absorption study was performed in 1998 and involved eight research subjects.
14 The study was a double-blind, randomized, placebo-controlled, crossover study involving three
15 different doses (in a four phase dose escalation design). The protocol stated that the research was
16 in compliance with the above standards, with the updated GCP guidelines promulgated in 1996.
17 The approval of the research ethics committee was given on December 22, 1997, although no
18 information is provided about the substance and process of that review.
19

20 Critique of Study

21

22 The Board concurred with the factual observations of the strengths and weaknesses of the
23 two 1984 metabolic studies, as detailed in Carley (2006g). Specifically, there is no justification
24 for the dose selection, no record of independent committee review, and no record of informed
25 consent (other than the description of the participants as volunteers. The dose selected did result
26 in adverse events as might be expected from the administration of a product with this
27 pharmacological profile. The period of recorded observations was eight hours, and stated
28 observation time was 12 hours. Although the period of observation was insufficient to assure
29 that the physiologic condition of the volunteers had returned to baseline, the known physiologic
30 effects of the compound are such that the period of observation was sufficient to assure the lack
31 of significant adverse effects.
32

33 The Board concurred with the factual observations of the strengths and weaknesses of the
34 1992 oral dosing study as documented in Carley (2006h). Of note was that the volunteer
35 information included information about the risks of the administration of the study compound
36 which were relatively complete in the scope of the risks described (yet absent a few details
37 within each organ system).
38

39 The Board concurred with the factual observations of the strengths and weaknesses of the
40 1998 dermal absorption study, as documented in Carley (2006i). Although concern has been
41 expressed about the choice of dose, the study design involved a dose escalation such that adverse
42 events at lower doses could have been anticipated.
43

44 HSRB Consensus and Rationale

45

46 The Board concluded that:

- 1
2 a) The amitraz acute oral and dermal human toxicity studies failed to fully meet the specific
3 ethical standards prevalent at the time the research was conducted.
4
5 b) There was no clear and convincing evidence that the research was fundamentally unethical--
6 intended to seriously harm participants or that informed consent was not obtained.
7
8 c) There was no clear and convincing evidence of significant deficiencies in the ethical
9 procedures that could have resulted in serious harm (based on the knowledge available at the
10 time the study was conducted), nor that information provided to participants seriously impaired
11 their informed consent.
12

13 Although the Board concurred in the noted deficiencies with the 1984 metabolic studies,
14 the Board did not consider the lack of documentation of medical supervision following the
15 termination of dosing in the 1984 metabolic studies to be significantly deficient relative to the
16 ethical standards prevailing when the study was conducted.
17

18 The Board also acknowledged the observations that the 1992 oral dosing study and the
19 1998 dermal absorption study includes potentially misleading references in the informed consent
20 documents. However, the Board does not believe that these deficiencies could have resulted in
21 serious harm based on the knowledge available to the investigators at the time, nor seriously
22 impaired the informed consent of the research subjects. As such, the Board did not consider these
23 findings significantly deficient relative to the ethical standards prevailing when the study was
24 conducted.
25

26 The Board concurred with the assessment of the Agency that there was not clear and
27 convincing evidence that the conduct of the research was fundamentally unethical. The research
28 as designed and conducted was not intended to seriously harm participants nor failed to obtain
29 informed consent. Thus, the Board concluded that there was no ethical objection to the
30 appropriate use of the data from these studies (as discussed in the scientific assessment) given
31 the absence of clear and convincing evidence that the research was fundamentally unethical or
32 significantly deficient relative to the ethical standards prevailing at the time.
33

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