European Food Safety Authority

EFSA Scientific Report (2009) 282, 1-46

SCIENTIFIC REPORT OF EFSA

Outcome of the public consultation on the revision of the existing guidance document on dermal absorption (SANCO/222/2000 rev. 7) under Council Directive 91/414/EEC.

Prepared by the PPR Unit

(Question No EFSA-Q-2008-755)

Issued on 25 March 2009

BACKGROUND

In 2006, EFSA consulted Member States on their priorities for development and revision of guidance documents and in response to that some Member States expressed a wish for an update of the guidance document on dermal absorption.

The Pesticide Risk Assessment Peer Review Unit (PRAPeR Unit) reported that within the frame of the peer review of pesticides a number of questions regarding the assessment of dermal absorption of plant protection products are recurring, since they are not, or are insufficiently covered by the current guidance document. In addition to that, the level of knowledge about dermal penetration of chemicals has advanced and comprehensive evaluations of methodological aspects in this field have recently been published or are currently being carried out.

Therefore the Executive Director of EFSA has formally requested an opinion of the PPR Panel on the revision of the current guidance document.

The Plant Protection Products and their Residues Unit (PPR Unit) has carried out a public consultation on the revision of the existing Guidance Document on Dermal Absorption (SANCO/222/2000 rev.7) under Council Directive 91/414/EEC.

The public consultation, which was open from 15th November 2008 to 15th January 2009 was launched with the objective of involving interested stakeholders at a very early stage of the work on the revision of the guidance document and to gain further information on which issues should be considered and how they might be addressed.

COMMENTS RECEIVED

By the end of the deadline EFSA had received in total 83 comments from 14 different stakeholders. 49 comments were submitted by Member States Authorities, two by the Commission (JRC) and 32 comments by Industry/Industry associations. Notably, many comments addressed more than one issue.

Table 1 Comments received from the different stakeholders

Stakeholder	No of comments*
Institute of Public Health, **Slovenia**	7
Federal Office of Consumer Protection and Food Safety, **Germany**	7
Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of **The Netherlands**	13
Swedish Chemicals Agency (KemI), **Sweden**	5
Scientific Institute of Public Health, **Belgium**	3
French Food Safety Agency, AFSSA, **France**	5
Estonian Research Institute of Agriculture, **Estonia**	1
Ministry of Health and Consumer Affairs, **Spain**	3
Hellenic Ministry of Rural Development and Food, **Greece**	1
Pesticides Safety Directorate, **United Kingdom**	4
ECVAM, DG JRC, **European Commission**	2
Charles River Laboratories, UK	14
Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	7
Technology Sciences (Europe) Limited (TSGE), UK	11

*A comment can also consist of more than one general/specific comment

The comments had to be given to the individual chapters of the current guidance document. In addition to that it was also possible to make general comments and comments/suggestions for additional chapters to be inserted in the revised guidance document.

Table 2 gives the distribution of the 83 comments over the different chapters and subchapters.

Table 2 Comments per chapter

Chapter/subchapter of the current guidance document	No of comments
0. General comments	9
1. Introduction	6
2. An overview of dermal absorption 2.1. Introduction	3
2. An overview of dermal absorption 2.2. The studied tissue	6
2. An overview of dermal absorption 2.3. Active substance properties affecting penetration	2
2. An overview of dermal absorption 2.4. Experimental conditions	7
3. Studies on dermal absorption 3.1. I*n vitro* studies	6
3. Studies on dermal absorption 3.2. *In vivo* studies 3.2.1 Animal studies	4
3. Studies on dermal absorption 3.2. *In vivo* studies 3.2.2. Human volunteer studies	3
4. Decision making process for setting dermal absorption percentages General comments	4
4. Decision making process for setting dermal absorption percentages 4.1. Dermal absorption based on default values	4
4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on *in vitro* human and rat studies	6
4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on *in vivo* data	8
4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on in vivo rat studies in combination with *in vitro* data	5
5. Proposal for a tiered approach to risk assessment for operator exposure, using default dermal absorption percentage or dermal absorption percentage determined experimentally	3
6. Comments for insertion of additional chapters	7

SUMMARY EVALUATION OF THE CONTENT OF THE COMMENTS

The comments that are summarised below are grouped together based on the issues they addressed. Numbers in brackets [] refer to the comment number given in the first column of the table in the Annex, where all the comments with the full text as submitted by the stakeholders are listed.

Each comment submitted will be critically considered within the frame of the work on the revision of the guidance document.

1. Improving usability of the guidance

It was noted that more practical examples should be inserted, methodology for testing should be streamlined to improve comparability of results. Knowledge gaps should be documented. Overall, the strengths and weaknesses of the different assessment methods should be discussed. Guidance on assessment of exposure for re-entry workers should be given.

Comments: [1], [2], [81];

2. Updating text and references and scope of the guidance

A number of comments were received requesting revision of text on the scope of the guidance, taking into account also the current state of the peer review of plant protection products under Council Directive 91/414/EEC. Outdated references should be deleted. New relevant references should be inserted.

Comments: [11], [12], [14], [17], [18], [24], [35], [37], [38], [40], [42], [64];

3. Consideration of work being carried out by other regulatory bodies

It was noted that guidance documents and on-going work of other regulatory bodies, in particular OECD, should be taken into account and contradictory statements should be avoided. Contacts with other relevant bodies, in particular OECD, should be established.

Comments: [3], [11];

4. Read across for active substances in different formulations

Guidance/recommendations for the application of read-across between formulations or related active substances in formulations should be given. General principles for read across should be discussed. It was also suggested to add a list with co-formulants to which special attention should be given. Guidance on the acceptability of studies carried out with the active substance only was requested.

Comments: [2], [7], [8], [13], [28]; [31], [79], [82];

5. Use of human data

It was proposed that human (*in vivo*) data should be used only when the determined absorption values are higher than in animal studies. Text on the use of human data should be updated taking into account the new draft regulation on PPPs.

Comments: [4], [44], [45], [46];

6. Stratum corneum and tape stripping

It was noted that more explanations on the function of the stratum corneum as a reservoir should be given (possibly in a separate chapter). Ficks's Second Law of Diffusion should be presented in this context. Profiling of the stratum corneum was also advocated in order to decide on inclusion/non inclusion of retained substance in the fraction absorbed. Description and most of all clear guidance on inclusion/exclusion of tape strips was repeatedly requested for *in vivo* and *in vitro* tests. It was recommended also to consider the current practice of PRAPeR to exclude two tapes by default as not absorbed.

Comments: [6], [9], [36], [39], [47], [49], [56], [58], [59], [63], [65], [66], [68], [77], [78], [80], [83];

7. Definition and description of skin

A clear definition of what was meant by the term "skin" was requested. In addition, it was suggested to introduce text on the anatomy and physiology of the skin (explaining also Fick's First Law of Diffusion).

Comments: [5], [16], [25], [57];

8. Application of default values

The application of default values for dermal absorption based on physical-chemical properties in the absence of valid test data, as described in the current guidance, should be re-examined. Relevant text in the guidance should be modified accordingly.

Comments: [7], [15], [51], [52], [53], [54];

9. Use of full-thickness skin vs. split-thickness skin

The use of split-thickness skin was advocated (as also recommended by OECD). In addition measurements of skin integrity were proposed to avoid using damaged skin.

Comments: [19], [22], [23];

10. Exposure time, end of absorption, "serial non-detects" approach

It was pointed out that clear guidance on exposure time in *in vitro* and *in vivo* tests should be given. Guidance on derivation of dermal absorption reflecting re-entry worker exposure should also be provided. In one comment it was proposed to recommend only an exposure time of 8 hrs for consistency reasons. The term "non significant absorption" should be clearly defined. The "serial non-detects approach" should be revisited. The end of absorption/the

most relevant time point in *in vivo* studies should be clearly defined for deriving dermal absorption values.

Comments: [7], [34], [43], [61], [62], [64], [67];

11. Further guidance on studies, harmonisation between *in vitro* and *in vivo* studies, "triple pack approach"

It was suggested to clearly support the "triple pack approach" in the revised guidance. Reference to metabolic competence of test system should be made since it had an impact on absorption. More guidance on how to carry out *in vivo* and *in vitro* studies in practice should be given. The protocols for studies should be harmonised (tape stripping, exposure time, etc.) between *in vivo* and *in vitro* rat/human studies to increase comparability. Guidance on how to deal with studies where poor recovery is observed was also necessary. Clear definitions of terms "dermal absorption rate", "amount absorbed","fraction absorbed" and "fraction bioavailable systemically" should be provided. Suggestions for changes in Fig. 4 (describing the triple pack approach) were also submitted. It was proposed to use the term "skin depot" or "skin residue" instead of "skin dose".

Comments: [29], [33], [50], [59], [64], [70], [71] [72], [73], [79];

12. Alternative assessment methods

It was suggested to give a recommendation to assess dermal absorption with appropriate tests instead of applying alternative methods (phys-chem properties or read a cross). The use of acute toxicity studies should not be recommended. The possibility of using subacute studies for derivation of dermal absorption values, however, should be described in the revised guidance. The kinetic basis for assuming that dermal absorption does not exceed oral absorption should also be clarified. The application of QSARs for the assessment of dermal absorption should be discussed.

Comments: [2], [51], [53];

13. Use of flux vs. mass balance in *in vitro* studies

It was noted that clearer guidance on whether to use flux or mass balance for the assessment of dermal absorption should be given (OECD advocates mass balance). Addition of further text on the use of maximum flux for comparison through species was also proposed.

Comments: [36], [60];

14. *In vitro* test with human skin as stand alone method

The approach already currently applied in practice, to derive absorption values solely on the basis of human *in vitro* data should be recommended together with a scientific justification. The tiered approach should be rearranged accordingly.

Comments: [55], [74];

15. Tiered approach for assessment of dermal absorption

In one comment it was recommended to delete the entire chapter 5 since the guidance document on dermal absorption was not intended to give guidance on risk assessment. In another comment refinement of the tiered approach was proposed.

Comments: [75], [76];

FOLLOW UP

All comments, as listed in the Annex to this report, will be critically evaluated by the PPR Panel for further consideration for the work on the revision of the guidance document on dermal absorption. Adoption of the revised guidance is planned for the first half of 2011. Before adoption of the revised guidance document another public consultation, on the final text of the document is foreseen.

1 ANNEX: TABLE LISTING ALL COMMENTS

No	Institution	Chapter	Comment
1	Institute of Public Health, Slovenia	0. General comments	The guidelines should be optimised in order to give fewer options for choice to the notifier and consequently allow better comparability of data. This would also allow the creation of a database of comparable results which should give essential information for further developing of the second tier evaluation (cut/off criteria) in the future. It will also facilitate read-across approach. Where expert judgement is mentioned examples would be very useful.
2	Federal Office of Consumer Protection and Food Safety, Germany	0. General comments	The guidance document on dermal absorption has proven to be very useful. However, some revisions and amendments are in fact needed. Because the Federal Institute for Risk Assessment (BfR) will actively participate in the next stages of the revision process and will certainly provide much more detailed comments, the focus of this contribution is on identification of those parts of the document that should be substantially amended and of additional issues that are not addressed so far but should be included in the revised version. General comments: The possible sources of information on dermal absorption should be mentioned, per-haps in a summary table, which can be used instead of the (unrealistic) assumption of a 100 % default value. The advantages and limitations of the different methods and approaches and their very different reliability for prediction of the dermal absorption rate should be highlighted. A clear recommendation should be given to use experimental data obtained in valid dermal absorption studies if available because of their much higher reliability as compared to, e.g., the comparison between oral and dermal short-term toxicity studies or the various non-experimental methods (consideration of physico-chemical properties, QSARs, “Read-across” etc.). All the latter information, at best, can be accepted to provide rough estimates of dermal absorption that will allow refinement (i.e., reduction) of the default value to rounded figures such as 50 %, 25 %, 10 %, 5 % or 1 %. Even results from properly conducted dermal absorption studies should be considered with care and some reservation. In any case, results should be preferably rounded to whole numbers. It might be recommended to use 1 % as a “worst

No	Institution	Chapter	Comment
			case" for substances for which "very low" dermal absorption has been experimentally proven. Formulations of active compounds are authorised by the MS but not the actives themselves. Naturally, for technical and ethical reasons and because resources are limited, it is not possible to test all formulations. Accordingly, guidance should be given under which conditions data from one formulation can be applied to another containing the same active ingredient(s). i.e., how to define a "similar" formulation or how to define significant differences. For this purpose, the possible impact of co-formulants, mixtures of compounds and dilution factors must be addressed. Guidance should also be given regarding the transfer of absorption data based on the active substance alone. Gaps in our knowledge should be clearly stated as such. References should be checked to find out whether in fact the best and most reliable of the available scientific information has been included.
3	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	0. General comments	It is essential that the next version of Sanco/222/2000 does not conflict with the OECD Guidance notes for the estimation of dermal absorption values (the draft of 26 May 08 is now being revised by an expert group lead by Australia). Collaboration between the two activities should therefore be initiated.
4	Swedish Chemicals Agency (KemI)	0. General comments	We are concerned about testing of chemicals on humans, particularly pesticides, as these chemicals are toxic per definition. However, if such dermal absorption studies are already available, they should be used only if they show a higher dermal absorption than already obtained in the animal studies.
5	Charles River Laboratories, UK	0. General comments	This is a general comment that in sections 4.2 and 4.4 there is no mention of stratum corneum for calculation of results (only "skin" mentioned). Therefore, especially for in vitro studies, there should be further clarification on if, when referring the "skin" this document means: 1. viable skin (epidermis and dermis) + all stratum corneum. 2. viable skin (epidermis and dermis) - all stratum corneum. 3. viable skin (epidermis and dermis) + lower layers of stratum corneum.

No	Institution	Chapter	Comment
			If (3.) is the most accurate description then further clarification is needed on what constitutes "lower layer" (i.e. how many tape strips). If this is dependant upon results (flux data etc) then this should also be clarified. Whilst I have focused on in vitro, I believe this may also be applicable for in vivo studies where there are no serial non-detects in urine or faeces. One other minor point is OECD references need updated to 2004 in section 3.1.
6	Charles River Laboratories, UK	0. General comments	<u>General suggestions:</u> There is very little discussion on the issue of the stratum corneum. There is no discussion on Ficks 2nd Law of diffusion. Since absorption is Ficks 2nd Law of diffusion, it is important to define what is the semi-permeable membrane. It is generally accepted that the membrane is actually the stratum corneum. This makes it essential to include the "viable" skin, but not the stratum corneum in the risk assessment (in line with the citation for EPA, 1992). However, it does not take into account the potential stratum corneum reservoir. With stratum corneum profiling detailed, it is much simpler to decide if all/ some/ none of the stratum corneum should be included in the risk assessment especially if compared with the absorption profile. Should you require any further comment or require my input in person, then I will be available.
7	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	0. General comments	Unfortunately, due to time constraints, ECPA Members* were unable to provide comments on all of the concerns held with the existing guidance document. In addition, some detail has been lost due to the limitations set by EFSA on the length of individual comments. ECPA Members hope to be able to rectify these omissions during future steps of the review process. ECPA Members believe that the critical points that require revision include: • New guidance on default absorption values based on database of studies conducted in accordance with OECD 427 and 428 • New guidance on tape stripping to cover both in vitro and in vivo studies • New guidance on the use of read across between formulation type and/or related active substances • Revision on guidance for the definition of end of in vivo absorption *EFSA note: Individual names are not disclosed.

No	Institution	Chapter	Comment
8	Technology Sciences (Europe) Limited (TSGE), UK	0. General comments	Discussion of the general principles of read-across between tested formulations may be useful. This may include the following points: It may be possible to extrapolate a dermal absorption value generated using a solvent-based formulation to a water-based or solid formulation as dermal absorption from the solvent-based formulation is likely to represent a worst case. Similarly it may be possible to extrapolate from a water-based formulation to a solid formulation. However extrapolation in the other direction is not possible. It may be possible to extrapolate a dermal absorption value generated using a product dilution to the product conventrate, as this is likely to be a worst case. Similarly it may be possible to extrapolate a derrmal absorption value from a more dilute product to a less dilute product, however extrapolation from a less dilute product to a more dilute product should be addressed on a case by case basis and is unlikely to be acceptable for a significant difference in concentration (e.g. 2-3 times ?) unless exisitng data indicate that absorption is not strongly influenced by concentration.
9	Technology Sciences (Europe) Limited (TSGE), UK	0. General comments	Inclusion of a discussion of tape-stripping is needed. This should include its potential role in demonstrating the likely systemic avaialability or non-availability of the skin residue and the identification of surface residue (1st 2 tape strips).
10	Institute of Public Health, Slovenia	1. Introduction	The second paragraph (indented) should preferably be moved out from the Introduction into a separate chapter, maybe placed before the Introduction. Maybe it should be rewritten to clearly separate what is the rule and what are exceptions in specific situations.
11	Federal Office of Consumer Protection and Food Safety, Germany	1. Introduction	The limitation of the scope of the document to the 3d stage of the evaluation programme must be deleted. It should not be allowed to use 10 % default value for ongoing evaluation without being justified by physico-chemical properties etc. In addition, the revised version should be placed properly into the context of other regulatory documents on dermal absorption such as the OECD guidelines 427 and 428, the OECD guidance document, or the comprehensive IPCS (EHC) paper on dermal absorption. The “OECD guidance notes” on harmonisation of study interpretation and the approaches to be taken if there is no specific data available, which are currently being drafted by an expert group, should also be taken into consideration.

No	Institution	Chapter	Comment
12	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	1. Introduction	Page 2, Introduction, first paragraph: It should be more explicitely stated that this Guidance Document is used for the risk assessment of operators, workers and bystanders and that the whole document is applicable to these groups. Page 2, third paragraph: Some exposure models for the operator are mentioned, but not for the bystander and worker. Since this GD does not cover exposure models, it is advised to remove these models from the text.
13	Swedish Chemicals Agency (KemI)	1. Introduction	It should be emphasized in the introduction that for product authorisation, dermal absorption studies on the product are required. If studies are not available on the product applied for, studies on related products can be used if the chemical compositions of the two are similar. A suggestion is to list formulants that should be paid special attention to, e.g. ethanol (as in Appendix III of the OECD draft document Guidance Notes for the Estimation of Dermal Absorption Values, 26 May, 2008).
14	Charles River Laboratories, UK	1. Introduction	Paragraph 3, last line: OECD, 2004a, b, c (draft 2000 replaced by final 2004). This needs to be checked throughout the entire document.
15	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	1. Introduction	Comment 1: The following text *"To provide a reliable framework … when applying the Uniform principles."* should be replaced by: *"Since the release of the initial Guidance Document (Rev.7), dermal absorption studies conducted in accordance with OECD 427 and 428 test guidelines have been conducted and submitted as part of the EU review process. Therefore, now it is possible to determine realistic default dermal absorption values based on this extensive empirical database, which covers the impact of formulation type on absorption of an active substances from a PPP and its spray dilutions."* This revision is required because the use of physical-chemical properties of an active substance has been shown to be an inadequate predictor of dermal absorption of an active substance from a formulation and its spray dilutions. This can be demonstrated by a comparison of the physical-chemical properties and dermal absorption values of an active substance published as CEP's in the EU review process and therefore this comparison should be a part of the EFSA review procedure for this Guidance Document. Comment 2: The following text *"This document provides … guidance on how to conduct relevant dermal absorption studies"* should be deleted.

No	Institution	Chapter	Comment
			This revision is required because guidance on how to conduct studies is contained in the OECD test guidelines (427 and 428) and also in the specific OECD Guidance Document (#28) for conduct of these studies. Additional guidance on conduct is not required, or provided, in this guidance document.
16	Scientific Institute of Public Health, Belgium	2. An overview of dermal absorption 2.1. Introduction	Line 12 up to line 24 (...1993): we propose a modification of the paragraph as following: *"upon contact with the skin, a compound penetrates the outer layer, the stratum corneum (keratinocytes), which is basically impermeable to water and hence to water soluble polar chemicals. The stratum corneum also prevents evaporation of water from the underlying cell layers. Since in normal conditions the stratum corneum is highly hydrated, the skin can still take up polar substances, albeit slowly, through passive diffusion as governed by Fick's First law. The hair follicle shafts are thought to provide a further route of entry. This route is considered to be most important during early stages of absorption process, especially for lipophilic compounds. The sweat and sebaceous glands are also thought to confer permeation routes. Skin permeability is enhanced by hydration of the skin.* *Other factors affect penetrability such as changes in ambient temperature, damage to the horny layer as well as edema, solar irradiance, exposure to organic solvents.* *The second phase is diffusion od the chemical through the lower layers of epidermis and dermis. These cell layers are far inferior to the stratum corneum as barriers. They contain porous, nonselective aqueous diffusion medium and chemicals pass through this area also by diffusion. The chemicals diffuse to the dermis where a microcapillary bed of blood vessels serves to carry the molecules away from the dermis. Once the permeating molecules have entered the blood, they are considered to be bioavailable. These different aspects explain that skin structure differs from one species to another."*
17	French Food Safety Agency, AFSSA	2. An overview of dermal absorption 2.1. Introduction	The following documents/references could be mentioned in this introduction: OECD, Guidance notes for the estimation of dermal absorption values, draft may 2008WHO, IPCS/EHS 235, Dermal absorption, 2006.
18	Charles River Laboratories, UK	2. An overview of dermal absorption 2.1. Introduction	Paragraph 2, last sentence: The stratum corneum is the barrier to absorption. As stated above, once in the viable epidermis it is in the living tissue. The Flynn reference should be deleted and replaced with something more suitable and far less abstract in thinking. Paragraph 3, line 4. Suggest adding SCCP Guidance document (cosmetic products), although this is under review currently.

No	Institution	Chapter	Comment
19	Institute of Public Health, Slovenia	2. An overview of dermal absorption 2.2. The studied tissue	Three types of skin membranes can be used for in vitro experiments although for two of them disadvantages are mentioned in the same paragraph. I would prefer to see the split-thickness skin as the main choice and any deviations should be justified. That would reduce variability in testing which is important in creating a good database.
20	Scientific Institute of Public Health, Belgium	2. An overview of dermal absorption 2.2. The studied tissue	If the previous comments are taken into account then the first 6 lines of point 2.2 could be deleted. The paragraph could start with "In case of in vitro studies..."
21	Estonian Research Institute of Agriculture, Estonia	2. An overview of dermal absorption 2.2. The studied tissue	2.2.- page 5, line 1. Scott(1991) - skin permeability could be related to species differences in skin structure..... -skin structure is depending on age of rats and maybe on strains of rats ? How to avoid this differences and mistakes?
22	Charles River Laboratories, UK	2. An overview of dermal absorption 2.2. The studied tissue	Paragraph 1, line 10: full thickness skin is not recommended cf OECD TG 428. The lower dermis will act as a significant sink, so partitioning to the receptor fluid and thus prediction of systemic availability will be far lower.This does not impact on the overall risk assessment because the “viable” skin should always be included as well.
23	Technology Sciences (Europe) Limited (TSGE), UK	2. An overview of dermal absorption 2.2. The studied tissue	Lines 14-15: *"epidermal membranes are more fragile and sometimes overestimate human in vivo skin absorption"* Some clarification of this statement is required: It is generally accepted that in vitro studies overestimate dermal absorption in human skin in vivo - this is the basis for the use of in vitro data outlined later in the document. However the statement above indicates that damaged membranes may be used, which is not the case if appropriate measurements of integrity are made.
24	Technology Sciences (Europe) Limited (TSGE), UK	2. An overview of dermal absorption 2.2. The studied	Line 1 page 5 should read *"Scott et al (1991)"*.

No	Institution	Chapter	Comment
		tissue	
25	Scientific Institute of Public Health, Belgium	2. An overview of dermal absorption 2.3. Active substance properties	Point 2.3 line 1-11: We propose to include in the text somewhat more details such as: *"Two intrisinc factors contribute to the absorption rate of a given compound:* *-hydrophobicity: this parameter is measured by the octanol/water partition coefficient (Kow) partitioning of an agent into the skin is greatly affected by its solubility in or adhesion to the medium in which it is applied. the physicochemical properties of the vehicle are very important in influencing the rate of percutaneous absoprtion, since they will regulate the vehicle/stratum corneum partition coefficient.* *-rate of diffusion through this barrier: this property is an inverse function of molecular weight or more accurately, of molecualr volume.* *Ionic state plays an important role as non ionized molecules penetrate easily. however, substances which are in the ionized state at pH 7.4 may have a higher permetation coefficient than would be expected from Poct or by molecular weight."*
26	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	2. An overview of dermal absorption 2.3. Active substance properties affecting penetration	Please include "*penetration*" after the first parenthesis after the first bullet, otherwise the impression is created that liposolubility is at its maximum between log Pow +1 and +2.
27	Technology Sciences (Europe) Limited (TSGE), UK	2. An overview of dermal absorption 2.4. Experimental conditions	Line 6: For compounds binding to the skin, dermal penetration at low concentrations is actually lower than at high concentrations. Similarly ionisation of compounds may increase with dilution, resulting in lower dermal absorption. Line 14: The term "strongest dilution" can be misinterpreted. Suggest using "greatest dilution" (lowest concentration).

No	Institution	Chapter	Comment
28	Institute of Public Health, Slovenia	2. An overview of dermal absorption 2.4. Experimental conditions	The influence of the solvent on skin penetration of a substance may be different for different substances therefore studies should be performed for the preparation, which is not always the case in practice. It is difficult to judge the argumentations of notifiers if we do not have clear rules in which situation a study with active substance would be accepted. Studies performed with three concentrations (product, mix/load dilution and spray dilution) would add a lot to better understanding of dependence of skin penetration on a substance and the solvent present.
29	ECVAM, DG JRC, European Commission	2. An overview of dermal absorption 2.4. Experimental conditions	Paragraph 2.4, page 5, line 4 of paragraph, and further down in document: SANCO Document 222/2000 states: "At low concentrations the absorbed test substance expressed as percent of applied dose per time interval is in general higher than the percentage absorption at high concentrations. As a consequence, there is no standard absorption percentage for a given substance." It is correctly stated that the bioavailability by dermal route is not a single percent figure, since it depends on the concentration applied, but also on the vehicle, on the surface of contact, on the part of the body which is exposed, on the time of contact, etc. This should be more clearly emphasised. Further in the text the expression "absorption percentage" should be avoided because it intuitively implies a constant ratio, and it is often misunderstood as such. More precise and less concise wording is necessary to avoid this ambiguity. A possible way around this difficulty might be to give a few initial definitions, and refer to them further in the document. For instance, the following concepts (or other as necessary) could be defined (to be discussed by experts): - Dermal absorption rate (how much goes through and into the skin per unit of time and surface), - Amount absorbed (how much has gone through and into the skin after a given time), - Fraction absorbed (amount absorbed, relative to the amount of substance applied on the skin surface), - Fraction bioavailable systemically: (amount of substance which passes into the general circulation, relative to the amount of substance applied on the skin surface).

No	Institution	Chapter	Comment
30	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	2. An overview of dermal absorption 2.4. Experimental conditions	Please consider the following reference: Buist, H.E., Schaafsma, G. van de Sandt, H., 2009. Relative absorption and dermal loading of chemical substances: Consequences for risk assessment. Manuscript submitted to Regulatory Toxicology and Pharmacology. This document confirms the inverse relation between area dose and percentage of absorption, based on an up-to-date and extensive database.
31	French Food Safety Agency, AFSSA	2. An overview of dermal absorption 2.4. Experimental conditions	At the end of § 2.4: In vitro dermal absorption studies performed on the active substance (without commercial vehicle/formulants) should not be used to predict dermal absorption from a formulated product, unless the vehicle used in the dermal absorption study is comparable to the preparation' s vehicle
32	Charles River Laboratories, UK	2. An overview of dermal absorption 2.4. Experimental conditions	Paragraph 2 line 5: Add: Therefore results should be expressed as both % applied dose and µg/ cm2. Paragraph 2, line 12: strongest dilution (I assume this means most concentrated) + last paragraph: "diluted to minimum use concentration". I think this is misleading. Since absorption follows Ficks 2nd Law of diffusion, we should test at least the most concentrated in-use or a range.The most diluted concentration would not cover higher concentrations in line with this Law.
33	Technology Sciences (Europe) Limited (TSGE), UK	2. An overview of dermal absorption 2.4. Experimental conditions	Some reference is required either in this section or elsewhere to the metabolic capacity of the test system in vitro. For compounds where dermal absorption is influenced by metabolism, adequate metabolic capacity in the test system is required, otherwise absorption may be significantly underestimated.
34	Institute of Public Health, Slovenia	3. Studies on dermal absorption 3.1. In vitro studies	For the exposure time one value only should be given, preferably 8 hours as the worst scenario of the values suggested now. This is another step to reduce variability in the test procedure. The same holds for point 3.2.

No	Institution	Chapter	Comment
35	Ministry of Health and Consumer Affairs, Spain	3. Studies on dermal absorption 3.1. In vitro studies	Update the first paragraph as follows: The test should be carried out in accordance with "OECD Guideline for the Testing of Chemicals. Guideline 428: Skin Absorption: in vitro method" (OECD, 2004) and the OECD Guidance Document for the Conduct of Skin Absorption Studies (OECD, 2004).
36	Federal Office of Consumer Protection and Food Safety, Germany	3. Studies on dermal absorption 3.1. In vitro studies	In most studies of this type that we receive, exposure time was 24 hours but not 6 – 8 al-though this would better reflect a normal working day in Europe. Some guidance should be given because the revised guidance document will have an impact on the study design. The question of tape stripping that is sometimes applied also in vitro, should be addressed. There is an ongoing discussion whether the flux or the percentage of absorbed radioactivity (including the skin bound portion) should preferably be used. A clearer guidance would be desirable. At least in the OECD expert group, a majority of participants was in favour of the absorbed radioactivity and this approach is also supported by the BfR.
37	Charles River Laboratories, UK	3. Studies on dermal absorption 3.1. In vitro studies	One other minor point is OECD references need updated to 2004 in section 3.1
38	Charles River Laboratories, UK	3. Studies on dermal absorption 3.1. In vitro studies	Paragraph 1: OECD, 2004
39	Hellenic Ministry of Rural Development and Food, Greece	3. Studies on dermal absorption 3.1. In vitro studies	The revised version (rev. 7) of Guidance Document on Dermal Absorption is very informative and helpful in the derivation of the dermal absorption values in relation to the risk assessment of plant protection products. According to our opinion, the interpretation of data concerning tape stripping in the in vitro studies with human skin should be further clarified As it is mentioned in the "EFSA Handbook for the experts' meetings, Section 2: Mammalian toxicology", available in CIRCA, the SCP opinion (Doc. SANCO/222/2000 – rev4 dated 11 April 2001) has stated not to include the amount of substance bound in the stratum corneum to the amount systemically absorbed. This issue has been widely discussed during the PRAPeR meeting leading to the following approach: In the in vitro dermal absorption test, the amounts detected on the first two tape strips can be considered as not absorbed (because the substance apparently remains in the stratum corneum) while the amounts on the other tape strips are considered as absorbed. EL agrees to this approach.

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40	Ministry of Health and Consumer Affairs, Spain	3. Studies on dermal absorption 3.2. In vivo studies 3.2.1 Animal studies	Update the first paragraph as follows (adding the highlighted text): The test should be carried out in accordance with "OECD Guideline for the Testing of Chemicals. Guideline 427: Skin Absorption: in vivo method" (OECD, 2004) and the OECD Guidance Document for the Conduct of Skin Absorption Studies (OECD, 2004).
41	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	3. Studies on dermal absorption 3.2. In vivo studies 3.2.1 Animal studies	line 8:replace faith with fate.
42	Charles River Laboratories, UK	3. Studies on dermal absorption 3.2. In vivo studies 3.2.1 Animal studies	Paragraph 1: OECD, 2004
43	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA)*, Belgium	3. Studies on dermal absorption 3.2. In vivo studies 3.2.1 Animal studies	Comment 1: The following text "In order to get insight in the fate of the amount located in the skin, the sampling time should be long enough to determine that absorption from the application site is no longer significant, e.g. until serial non-detects in excreta." should be clarified. Although the original guidance was adequate, it was incorrectly interpreted by some Member States such that the absolute requirement for serial non-detects was the sole criterion for determining whether absorption had ceased. This was never the intention of the original guidance as it was clearly stated that absorption from the application site should in fact "no longer be significant". This is a reasonable requirement. Therefore, the principle should not be lost but the wording needs to be clarified such that the new guidance is totally unambiguous. All that is required is a simple

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			definition of "no longer be significant". ECPA propose the following text: *"In order to get insight in the fate of the amount of active substance located in the skin at the application site, the duration of the study should be long enough to determine that absorption is no longer significant. This can be achieved by determination of the amount of active substance excreted per day until the amount eliminated becomes insignificant in relation to the total amount excreted over the duration of the study. In this case, all of the material remaining at the application site should be excluded from the absorbed dose. This approach can be underpinned by consideration of the absorption profile with time, to confirm that absorption had effectively reached a plateau level at the end of the study.* *If it is not possible to establish that absorption is insignificant, the relationship between the absorbed dose and the residue at the application site during the study should be used to estimate the fraction of the material remaining at the application site that should be used to determine the total absorbed dose."* *Comment 2:* A specific case worthy of mention in the guidance document relates to bound skin residue (BSR) in the stratum corneum at the application site. "Skin contact with crop protection chemicals can occur during mixing and/or loading and spraying operations. However, systemic uptake of chemicals in contact with the skin is attenuated by the stratum corneum, the principal barrier to dermal penetration and absorption. The stratum corneum is the non-viable outer layer of the skin and is composed of anucleated cells (corneocytes) floating in a lipid matrix. In absence of damage, penetration of chemicals through the stratum corneum is passive and generally via the intercellular lipid lamellae. In general, corneocytes of the stratum corneum are constantly shed and renewed in a process known as desquamation, which lasts approximately 14-21 days (Milstone, 2004; Roberts and Marks, 1980). On occasion, chemicals that contact the skin can become bound to the stratum corneum, the bound skin residue (BSR), which may be lost during the normal process of desquamation or taken up by the capillaries in the viable epidermis and distributed systemically. Generally, if the rate of desquamation is faster than the rate of passive diffusion, the systemic availability of the BSR will likely be negligible. One option to unequivocally resolve the disposition of the BSR, animals, typically rats, should be held post-exposure for 21 days or longer, if required. Evidence of a decline in the total amount of radioactivity eliminated in excreta (urine and/or faces) and expired air (if applicable) must be demonstrated. Milstone, L.M. (2004). Epidermal desquamation. Journal of Dermatological Science 36, 131-140.

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			Roberts, D., and Marks, R. (1980). The determination of regional and age variations in the rate of desquamation: a comparison of four techniques. Journal of Investigative Dermatology 74, 13-16.
44	Ministry of Health and Consumer Affairs, Spain	3. Studies on dermal absorption 3.2. In vivo studies 3.2.2. Human volunteer studies	We consider that the last paragraph should be completed as follows (adding the following text): *"Results from field studies, if well conducted, and especially biomonitoring data may be helpful to confirm results obtained from experimental dermal in vivo and in vitro testing, and should be considered when it comes to estimating the dermal penetration of a substance. Additionally, even if there is not a field study carried out with the substance of interest, but there is one conducted with a chemically related substance that has similar physicochemical properties, the obtained results should be taken into account".* For example, a recent study has evaluated exposure and occupational risk during manual operations with ornamental plants treated with omethoate in intensive cultivation tunnels (Aprea, 2005). The plants had been treated 37 h before reentry with 220 ml of a commercial product (565 g/l pure omethoate) dispersed in 200 l water (equivalent to 0.62 g/l of active ingredient).The urinary excretion of alkylphosphate allowed for estimating that the fraction of omethoate absorbed through the skin during work was about 16.5%. Omethoate (O,O-dimethyl S-methylcarbamoylmethyl phosphorothioate) is a metabolite (the oxygen analogue) of dimethoate: O,O-dimethyl S-methylcarbamoylmethyl phosphorodithioate; both compounds have very similar physico-chemical properties (omethoate: mw 213 and logPow -0.7; dimethoate: mw 229 and logPow 0.7). In fact, the octanol–water partition coefficient of omethoate would suggest a more unfavourable absorption compared to dimethoate. However, in the conclusion regarding the peer review of the pesticide risk assessment of the active substance dimethoate (EFSA, 2006) the dermal absorption established is 0.15% for the concentrate and 2.0% for the dilution. These values come from the combination of rat in vitro and in vivo data and human in vitro data (Davies, D. J., 1999; Heylings, 2000; Leibold, E., 2001), but interestingly, in the dermal penetration in vitro study using human membranes, at the end of an 8 hour exposure with a spray-strength dilution of 2 g dimethoate/l, the absorption of dimethoate was 14,5%. This outcome suggests that the conclusion laid down in EFSA's document could represent an underestimation. Literature references: OECD Environmental health and safety publications. Series on testing and assessment n 28. Guidance Document for the Conduct of Skin Absorption Studies; March 2004.

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			OECD Guideline for the testing of chemicals: 428. Skin absorption: in vitro method. Adopted : 13 April 2004. OECD Guideline for the testing of chemicals: 427. Skin absorption: in vitro method. Adopted : 13 April 2004 C. Aprea, L. Centi, S. Santini, L. Lunghini, B. Banchi, G. Sciarra. Exposure to Omethoate During Stapling of Ornamental Plants in Intensive Cultivation Tunnels: Influence of Environmental Conditions on Absorption of the Pesticide. Arch. Environ. Contam. Toxicol. 49, 577–588 (2005). EFSA Scientific Report (2006) 84, 1-102, Conclusion on the peer review of dimethoate. Davies, D.J., 1999. Dimethoate: in vitro absorption from a 400 g/l EC formulation through human and rat epidermis. Company Report No. JV1591/REG/REPT. DTF Doc No. 460-003. Unpublished. Heylings, J.R., 2000. Dimethoate: In vitro absorption from a 400 g/l EC formulation through human and rat epidermis. Statement regarding project no.: 104-065 contract CO9027 - JV1591. Company Report No. CO9027-JV1591. DTF Doc No. 481-036. Unpublished. Leibold, E., Hoffmann, H.D., 2001. Study on the dermal penetration of 14C-Dimethoate in rats. Company Report No. 01B0418/006016. DTF Doc No. 654-002. Unpublished.
45	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	3. Studies on dermal absorption 3.2. In vivo studies 3.2.2. Human volunteer studies	This text needs to be updated.
46	Technology Sciences (Europe) Limited (TSGE), UK	3. Studies on dermal absorption 3.2. In vivo studies 3.2.2. Human volunteer studies	The section needs to be modified and clarified, in line with the position on the use of human volunteer studies.

No	Institution	Chapter	Comment
47	ECVAM, DG JRC, European Commission	4. Decision making process for setting dermal absorption percentages General comments	General comment on Chapter 4: fate of compound/stratum corneum, skin reservoir: There is a need for a standard (tiered?) procedure to interpret the possible role of reservoir of the stratum corneum. A recurrent discussion point in regulatory meetings is the number of strips which should be considered as non-absorbed when doing a tape strip procedure at the end of a dermal absorption study. There is also some disagreement in the available guidance or literature: For in vitro tests, the SCCP statement in their 2006 opinion (SCCP/0970/06) is that *"the amount present in the stratum corneum at the time of sampling is considered as not contributing to the systemic dose."* Yourick et al. (2004) on the same topic seem to have an opposite view (p. 318): *"The above examples demonstrate that the amount of material remaining in the skin at the end of a study should be included as part of the total dose absorbed (as recommended by Bronaugh and Collier, 1991), unless the fate of the chemical in skin is investigated and it is shown not to be available for systemic absorption."* Possibly the approach could be linked to the lipophilicity of the substance in question? It is usually considered that the stratum corneum is the most effective barrier against hydrophilic compounds, and the living layers of the epidermis and dermis against lipophilic compounds. References: 1. Jeffrey J. Yourick, Michael L. Koenig, Debra L. Yourick, and Robert L. Bronaugh. Fate of chemicals in skin after dermal application: does the in vitro skin reservoir affect the estimate of systemic absorption? Toxicology and Applied Pharmacology 195 (2004) 309–320. 2. SCCP/0970/06. Scientific Committee on Consumer Products Opinion on basic criteria for the in vitro assessment of dermal absorption of cosmetic ingredients - updated March 2006. Adopted by the SCCP during the 7th plenary of 28 March 2006.

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48	Swedish Chemicals Agency (KemI)	4. Decision making process for setting dermal absorption percentages General comments	Dermal absorption values should be given as integers and numbers below 1% should be rounded to 1%. This would compensate for scratches and small wounds on the farmers skin, which would represent a larger percentage of a low figure than a high figure. Furthermore, the accuracy of the studies does not allow setting dermal absorption figures with decimal precision.
49	Swedish Chemicals Agency (KemI)	4. Decision making process for setting dermal absorption percentages General comments	Clear instructions on how to deal with substances found in stratum corneum are needed. When should tape strips be used (in vivo/in vitro), how many strips should be excluded etc. In this respect, as the number of tapes stripped seem to vary between studies, it could be a point to add that regardless of the number of tapes stripped, the outer numbers excluded should be the same, if equal conditions are assumed, and so it is better to suggest a number of tape strips to exclude instead of a number to include.
50	Technology Sciences (Europe) Limited (TSGE), UK	4. Decision making process for setting dermal absorption percentages General comments	Figure I (page 10) consistency is required in the use of "rat" or "animal" Two boxes "in vivo studies" are not required. The two options under the second "in vivo studies" box should be consistent: (1) no serial non-detects and no strong decline (2) serial non-detects or strong decline Box at bottom right [dermal absorption percentages following in vitro studies] indicates that the value should be calculated using the skin plus receptor medium. There is no option here for refining the value by discounting all or some of the skin residue using expert judgement, as stated earlier in the document
51	Federal Office of Consumer Protection and Food Safety, Germany	4. Decision making process for setting dermal absorption percentages 4.1. Dermal	It is further supported that dermal absorption must not be derived from a comparison between oral and dermal acute studies as outlined in the current document. However, the situation may be different when subacute studies are considered. This principal opportunity should be mentioned and conditions should be described under which a comparison might contribute at least to a rough estimate. The opinion that dermal absorption, usually, will not exceed the oral one may be right but it should be clarified what

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		absorption based on default values	the scientific basis for this assumption actually was. In the OECD expert group, this argument was not accepted and dermal absorption should not be esti-mated on the basis of oral data. Rather than only providing a reference to literature, the experimental evidence for choosing either 10 or 100 % default values based on molecular mass and log POW should be better represented and more extensively discussed along with the uncertainties associated with this approach.
52	Pesticides Safety Directorate, United Kingdom	4. Decision making process for setting dermal absorption percentages 4.1. Dermal absorption based on default values	Page 7 - lines 8 -15: The scientific basis for the log Pow and molecular weight criteria is questionable. There is also a large margin from 100% to 10% it is possible that some intermediate value e.g. 30% might be justifiable based on alternative criteria. The choice of these default values should be reviewed to see if they are justified scientifically. A basis for any investigation could be the studies submitted during the 91/414 review process.
53	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	4. Decision making process for setting dermal absorption percentages 4.1. Dermal absorption based on default values	first paragraph: It should be realized that only few compounds (evaluated within the framework of 91/414/EC) exist with the physico-chemical properties leading to the default of 10% absorption. Since 1999, considerable progress has been made in the area of QSARs and it would be useful to provide some up to date information (see e.g. T. Bouwman, M.T.D. Cronin, J.G.M. Bessems, J.J.M. van de Sandt (2007). Improving the applicability of (Q)SARs for percutaneous penetration in regulatory risk assessment. Human and Experimental Toxicology 27. 269-276, 2007). second paragraph (oral absorption/ADME study): The assumption that dermal absorption is not likely to exceed oral absorption is indeed applied in current risk assessments. However, this paragraph refers only to oral absorption determined in bile cannulation studies. Oral absorption can also be determined based on an absorption study in non-cannulated animals. In case it is clear that a substance is almost exclusively excreted via urine and excretion via bile is relatively small, and the oral absorption has been determined in non-cannulated animals, the assumption that dermal absorption is not likely to exceed oral absorption can still be made. Please add this to the Guidance Document.

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54	Charles River Laboratories, UK	4. Decision making process for setting dermal absorption percentages 4.1. Dermal absorption based on default values	Paragraph 1, line 6-10: In practice, I have rarely seen values greater than 50% for absorption, perhaps a slightly less conservative value could be suggested eg 75% -80%.
55	Federal Office of Consumer Protection and Food Safety, Germany	4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on in vitro human and rat studies	There are not so few examples of the EU or individual MS having established the dermal absorption rate only on the basis of results obtained with human skin samples. It should be emphasised that this is considered usual practice in the EU and scientific evidence to sup-port this approach should be given, as well as possible limitations
56	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on in vitro human and rat studies	In dermal absorption studies (in vitro and in vivo) tape stripping can be performed. It would be useful to add a paragraph to provide some more tools for the risk assessors how to deal with tape stripping data. What number of tape strips can be regarded as non-absorbed in in vivo and in vitro studies? It has to be noted that the time of tape stripping is important. For instance, tape stripping immediately following washing of the skin may remove test substance that might have been absorbed through the skin, thus possibly leading to an underestimation of the dermal absorption. Test substance removed from the skin by tape stripping 24h after washing is less likely to have been absorbed. However, also a general warning should be given regarding the use of tape stripping data, given its limitations such as variability and the absence of general guidance. Since this issue will also dealt within the OECD guidance document (mentioned above; comments on the first concept have been received and the document is currently under revision) a reference to this document may suffice. In the current EU risk assessments of Plant Protection Products, it has been decided in the PRAPeR meetings to use the

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			pragmatic approach that the first two tape strips in an in vitro study can be regarded as not absorbed (See the EFSA List of decisions for mammalian toxicology). (The problem however is that in many study reports the results of the individual tape strips are not reported)
57	Swedish Chemicals Agency (KemI)	4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on in vitro human and rat studies	The word 'skin' is frequently used in the document. A recurring problem is whether stratum corneum, or parts of it, should be included or not. It should therefore be clearly stated which layers of the skin are referred to when the word 'skin' is used. (Example, top paragraph, page 8, line 5: 'By including the amount retained in the skin in vitro, a more acceptable estimation of skin absorption can be obtained.')
58	French Food Safety Agency, AFSSA	4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on in vitro human and rat studies	1st paragraph: *"There is an increase in the number…If refinement is needed, it should be convincingly demonstrated that the skin dose does not become absorbed at a later stage."* Comment to add: Continuous desquamation makes the first layers of the stratum corneum unlike to be part of this reservoir. Therefore, it is generally admitted that the 2-3 first strips are not included in the absorbable dose. The deeper layers and the remaining skin are included, unless it is demonstrated that they should not. 2nd paragraph: *"The maximum flux at relevant……within one species (provided they are tested under otherwise identical and relevant test conditions)".* This paragraph could perhaps be more detailed, to allow a pertinent use of the maximum flux value.
59	Charles River Laboratories, UK	4. Decision making process for setting dermal absorption percentages 4.2. Dermal	Paragraph 1, line 11: The major issue identified here is as a result of the way that data is generated in the two test systems (in vitro versus in vivo). Historically, skin was removed from the rat and analysed. There was no consideration taken as to the stratum corneum/ epidermis/ dermis distribution. The in vitro studies always have at least separated the "dead" stratum corneum from the "living" skin and often have separated the layers of stratum corneum and the epidermis/ dermis.

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		absorption based on in vitro human and rat studies	Indeed, we always take individual tape strips (1-20) to provide a stratum corneum profile. This adds information on whether the penetrant is on or near the surface of the stratum corneum and hence will be sloughed off or if it is uniformly distributed throughout the stratum corneum so the stratum corneum should then be added to the skin values as this material is clearly labile. The latter example then brings the in vivo/ in vitro studies closer together. We also perform our in vivo studies now in line with the in vitro studies, i.e. we remove the separate layers of the stratum corneum by tape stripping rather than taking the skin as a single sample. A closer correlation is then possible. Last line: the stratum corneum profiling will allow demonstration that the "skin" dose does not become absorbed. The material in the "living" skin tissue (viable epidermis and dermis" should always be included in the risk assessment cf para 2, line 1, section 2.1, dermal absorption definition (EPA, 1992). Paragraph 2: The maximum flux comparison is of course useful, but there are other important parameters to compare eg total absorption and skin distribution (as µg/cm2), and the shape of the absorption/ stratum corneum profiles. This is especially important in comparing species where the kinetics may be different.
60	Technology Sciences (Europe) Limited (TSGE), UK	4. Decision making process for setting dermal absorption percentages 4.2. Dermal absorption based on in vitro human and rat studies	Page 8, 2nd paragraph, line 2. It should be clarified that the maximum flux can ALSO be used for comparison of dermal absorption between species - a comparison can also be given using amounts in the receptor fluid (with or without skin) as described in the previous paragraph.
61	Federal Office of Consumer Protection and Food Safety, Germany	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	In this section, the most difficult question is on the termination of absorption that continues over the post-observation time. The "serial non-detects approach" should be re-examined because it failed to find world-wide acceptance so far. Furthermore, the most relevant time points should be defined that should be used for deriving dermal absorption values to be used in risk assessment (e.g., 8 – 10 hr measurements vs. determination after 24 hrs). Or, as an alternative, should the numerically highest value be used in any case?

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62	Pesticides Safety Directorate, United Kingdom	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	Page 8 - line 6 - 12 of section 4.3. The text regarding completion of excretion and serial non-detects should be deleted as it can be an invitation to do poor studies. At present this text is being interpreted very strictly by some MS (ignoring the subsequent comment about clear decrease in excretion) such that a study with excretion levels at about the LoQ (limit of quantification) at the end of the study has not shown complete excretion and thus the skin depot is deemed as being included. One way round this is to do the study with radio-label of a low specific activity and hence a high LoQ. A preferred approach would be to stipulate that the duration of an in vivo study should be such that it is the longer of 96 hours or four times (??) the terminal excretory half life from the oral ADME study. If at the end of such a dermal study there is a clear demonstration that excretion has shown a clear decline (or some arbitrary value e.g. 90% of the absorbed dose has been excreted) then the residue at the application site can be excluded from the bioavailable fraction.
63	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	See identical comment under Section 4.2. (Comment 53) In dermal absorption studies (in vitro and in vivo) tape stripping can be performed. It would be useful to add a paragraph to provide some more tools for the risk assessors how to deal with tape stripping data. What number of tape strips can be regarded as non-absorbed in in vivo and in vitro studies? It has to be noted that the time of tape stripping is important. For instance, tape stripping immediately following washing of the skin may remove test substance that might have been absorbed through the skin, thus possibly leading to an underestimation of the dermal absorption. Test substance removed from the skin by tape stripping 24h after washing is less likely to have been absorbed. However, also a general warning should be given regarding the use of tape stripping data, given its limitations such as variability and the absence of general guidance. Since this issue will also dealt within the OECD guidance document (mentioned above; comments on the first concept have been received and the document is currently under revision) a reference to this document may suffice. In the current EU risk assessments of Plant Protection Products, it has been decided in the PRAPeR meetings to use the pragmatic approach that the first two tape strips in an in vitro study can be regarded as not absorbed (See the EFSA List of decisions for mammalian toxicology). (The problem however is that in many study reports the results of the individual tape strips are not reported)

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64	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	first sentence: OECD 428 should be OECD 427 In an in vivo study, measurements are performed at different time points (e.g. after 24 h, 48 h, 168 h). Currently, different approaches are taken by Member States to evaluate these in vivo results: some Member States calculated dermal absorption based on the results after 24 h, while others calculate the worst-case values (e.g. based on results after 168 h). Is it possible to provide guidance on the time point which should be used to determine dermal absorption: the values after 24 h, 48 h, 72 h, 168 h, or the worst-case values (taking into account that the AOEL is expressed as acceptable exposure per 24 hours)? Guidance on normalization should be included (for both in vitro and in vivo studies) in case the recovery is <90% (with an acceptable justification for the relatively low recovery). Should the results be corrected for the lower recovery? And if yes, how (e.g. 'normalise' the results to 100%: 8% absorption with a recovery of 85% would be 9.4%)? This can probably only be resolved on a case-by-case base, depending on the cause of the low recovery, but some examples could be illustrative. This issue will also be dealt with within the OECD guidance document (mentioned above).
65	French Food Safety Agency, AFSSA	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	This comment can also be found in section 4.2 (comment 55) Continuous desquamation makes the first layers of the stratum corneum unlike to be part of this reservoir. Therefore, it is generally admitted that the 2 first strips are not included in the absorbable dose. The deeper layers and the remaining skin are included, unless it is demonstrated that they should not.
66	Charles River Laboratories, UK	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	First paragraph line 1: Replace OECD 428 (OECD 200b) with OECD 427 (2004b): First paragraph line 8 and last sentence:: Having information on the stratum corneum profile will allow a simpler interpretation of the data leading to a rational data based argument for including/ rejecting the stratum corneum in the risk assessment. The living epidermis and dermis must always be included.

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67	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	Comment 1: Replace *"If sampling is done over a sufficiently long period of time (e.g. until serial non-detects in excreta) the amount detected in the application site after washing should not be included in the amount absorbed."* with text based on the rationale for determining the end of absorption contained in ECPA Comment 1 in Section 3.2.1: *"In order to get insight in the fate of the amount of active substance located in the skin at the application site, the duration of the study should be long enough to determine that absorption is no longer significant. This can be achieved by determination of the amount of active substance excreted per day until the amount eliminated becomes insignificant in relation to the total amount excreted over the duration of the study. In this case, all of the material remaining at the application site should be excluded from the absorbed dose. This approach can be underpinned by consideration of the absorption profile with time, to confirm that absorption had effectively reached a plateau level at the end of the study.* *If it is not possible to establish that absorption is insignificant, the relationship between the absorbed dose and the residue at the application site during the study should be used to estimate the fraction of the material remaining at the application site that should be used to determine the total absorbed dose."* *Comment 2:* Delete the paragraph: *"In case excretion of the substance and/or its metabolites has not come to an end within the sampling period, but there are indications of a clear decrease in excretion, only a part of the skin bound dose may be included in the absorption by expert judgement (Thongsinthusak, 1999 ; De Heer, 1999). In case the experiment is terminated before serial non-detects in excreta are observed and/or no clear decline in excreta is measured, the amount located in the skin should be considered as being absorbed (Chu, 1996) (see Figure 1)"* based on the rationale for determining the end of absorption contained in ECPA Comment #1 in Section 3.2.1, as shown above. Comment 3: New text: In the absence of sufficient experimental data to adequately define the end of absorption the following approach may be considered. *"Dermal absorption is defined as the amount of applied dose that is ultimately taken up systemically and eliminated via urine, faeces and/or expired air, and that includes the amount recovered in tissue and carcass at termination.*

No	Institution	Chapter	Comment
			However, systemic uptake of chemicals in contact with the skin is attenuated by the stratum corneum, the principal barrier to dermal penetration and absorption. On occasion, chemicals can become bound to the stratum corneum, the bound skin residue (BSR), and unless its disposition is resolved, it must be assumed to be absorbed. The BSR may be lost during the normal process of desquamation or taken up by the capillaries in the viable epidermis and distributed systemically. Generally, if the rate of desquamation is faster than the rate of passive diffusion, the systemic availability of the BSR will likely be negligible. Therefore, sampling of excreta (urine, faeces, expired air) must be done over a sufficiently long period of time to resolve disposition of the BSR (i.e., up to 21 days post exposure, which encompasses the period of desquamation); a plot of cumulative radioactivity eliminated (via excreta) in maximized aliquots of excreta from analysis by liquid scintillation counting will provide the best practice for demonstration of a decline in absorption (from the BSR). However, for rat in vivo dermal studies terminated before a sufficient period of time post-exposure needed to resolve disposition of the BSR, a modeling technique can be employed to estimate the maximum absorption, and therefore, the systemic availability of the BSR (Thongsinthusak et al., 1999)." The Thongsinthusak et al. (1999) exponential saturation model was validated against a published dermal absorption study in human volunteers of 12 pesticides (Feldmann and Maibach, 1974). Overall, the model-derived dermal absorption estimates determined by the model were consistent with the reported values in Feldmann and Maibach (1974) and provided more realistic (yet conservative) estimates for those pesticides where the terminal elimination half-life ($t_{1/2}$) for urinary excretion was greater than the $t_{1/2}$ for dermal absorption. Accuracy of the maximum absorption prediction by the model is dependent on the elimination rate (via excreta) being faster than the absorption rate from the BSR to the systemic circulation. When the only source of available dose is that contained in the stratum corneum as the input (the BSR), and the elimination rate or output (via excreta) of the systemic dose is faster than the absorption rate or the input from the BSR, and there's no evidence of binding of the chemical or its metabolites to tissues once the chemical enters the body, then BSR depletion, either into the body or its loss via desquamation, will be directly reflected in the excreta output. However, if the chemical binds to tissues, as either the parent chemical or a metabolite, which in turn slows the overall rate of elimination from the body (output<input), the model losses the ability to resolve elimination from the BSR alone. In this case, the model will predict maximum absorption to occur long after the BSR has been lost by the normal process of desquamation (14-21 days) and the extent of absorption at the asymptote is suspect. Reason and justification for new text, if necessary: Removed concept of "serial non-detects" and provide additional guidance and clarification on resolution of BSR. References: Feldmann, R. J., and Maibach, H. I. (1974). Percutaneous penetration of some pesticides and herbicides in man.

No	Institution	Chapter	Comment
			Toxicology and Applied Pharmacololgy 28, 126-132. Thongsinthusak T., Ross J.H., Saiz S.G., and Krieger R.I. (1999). Estimation of Dermal Absorption Using the Exponential Saturation Model. Regulatory Toxicology and Pharmacology 29, 37-43.
68	Institute of Public Health, Slovenia	4. Decision making process for setting dermal absorption percentages 4.3. Dermal absorption based on in vivo data	There is great variability in expert judgments on the active substance in stratum corneum layers to be included or excluded as the amount absorbed. More guidance would be recommended in this document
69	Institute of Public Health, Slovenia	4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on in vivo rat studies in combination with in vitro data	If the usefulness of the Kp for dermal risk assessment is limited, this should not be included in predictions of absorbed dose. The equation would be preferred also for additional options of use (different formulations).
70	Federal Office of Consumer Protection and Food Safety, Germany	4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on in vivo rat studies in	A clear recommendation to use this so-called triple pack, if valid studies are available, should be given.

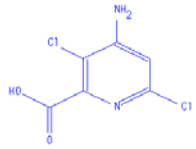
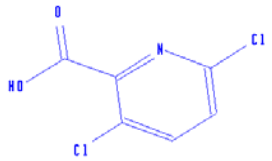
No	Institution	Chapter	Comment
		combination with in vitro data	
71	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on in vivo rat studies in combination with in vitro data	This paragraph describes that in vivo dermal absorption data in the rat may be adjusted in the light of the relative absorption through rat and human skin in vitro. It should be clearly stated here that the conditions under which the in vitro studies with human and rat skin were performed should be similar, for instance with respect to concentration of the test substance, vehicle used, receptor fluid, duration of test/ stripping etc. Page 9, last sentence: This should be rephrased: 'Similar adjustments can be made for differences between the dermal absorption of an active substance and the dermal absorption of an active substance in combination with (a) formulant(s) (e.g. in vivo active substance in rat and in vitro rat data on a formulant+active substance and active substance).
72	Charles River Laboratories, UK	4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on in vivo rat studies in combination with in vitro data	This method is certainly a useful approach to predict absorption in the human in vivo situation. However, this approach needs to be carefully considered. For example, the study design, formulation, test item concentration etc must be identical in the 3 tests used to generate in vivo absorption. Also, a realistic absorption value is needed and not an over prediction. Ethanol: water receptor fluids frequently generate an over prediction of absorption and provided an acceptable MoS is obtained, this does not matter. However, the overprediction is accentuated in the rat in vitro compared to the human in vitro and as such will result in a smaller calculated in vivo human value. Figure 1.: Dermal absorption percentages: must always include viable skin, but not stratum corneum (ie the barrier), some or all of the stratum corneum may be added where absorption is in steady state (absorption profile) or if the stratum corneum profile indicates test item is unlikely to be sloughed off.
73	Technology Sciences (Europe) Limited (TSGE), UK	4. Decision making process for setting dermal absorption percentages 4.4. Dermal absorption percentage based on	Page 9 line 9 It is stated that the "dermal absorption percentage" (receptor medium plus skin dose)" can be used to determine a dermal value. However it is stated in section 4.2 that the skin dose need not be included if it can be demonstrated that this does not become absorbed at a later stage. The term "skin dose" is misleading, suggest using "skin depot" or "skin residue" for clarification.

No	Institution	Chapter	Comment
		in vivo rat studies in combination with in vitro data	
74	Pesticides Safety Directorate, United Kingdom	5. Proposal for a tiered approach to risk assessment for operator exposure, using default dermal absorption percentage or dermal absorption percentage determined experimentally	Figure 2: In order to reduce the use of animals without compromising the overall evaluation, the first stage in Tier 3 should be to take the values from well performed studies using human skin in vitro. If these give an acceptable outcome then there is no need for any further studies. Rat studies in vitro over estimate human values so there is no need to do rat in vitro studies unless there is a need to do a rat to human comparison using in vivo & in vitro data.
75	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	5. Proposal for a tiered approach to risk assessment for operator exposure, using default dermal absorption percentage or dermal absorption percentage determined experimentally	Since this document does not cover the risk assessment process, it is advised to remove chapter 5 and Figure 2. Furthermore, Figure 2 is not complete with regard to the risk assessment process and does not add relevant information to the guidance on dermal absorption.
76	Technology Sciences (Europe) Limited (TSGE), UK	5. Proposal for a tiered approach to risk assessment for operator exposure, using default dermal absorption	Figure 2 page 12 Tier II indicates that a dermal absorption value based on in vitro data should be based on "receptor medium plus skin dose". This excludes the possibility of expert judgement regarding the likely systemic availability of the skin dose, as outlined earlier in the document. The figure also needs to include an option for refining the default dermal absorption values based on the extenet of oral

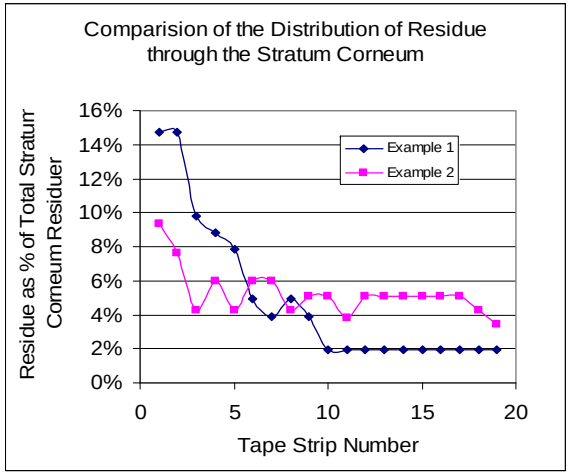
No	Institution	Chapter	Comment
		percentage or dermal absorption percentage determined experimentally	absorption, as detailed in Section 4.1
77	Board for the Authorisation of Plant Protection Products and Biocides (CTBG), on behalf of the Netherlands	6. Comments for additional chapters	Please add an additional chapter on tape-stripping (see also NL comment on 4.2 and 4.3).
78	Pesticides Safety Directorate, United Kingdom	6. Comments for additional chapters	Area where there is a need for additional guidance include: 1.Extrapolating dermal absorption of an active substance from one formulation type with data to another with no data. Some formulation types give lower dermal absorption values than others e.g. granule less than an EC. It is therefore feasible to take the data from the EC and use the value for a granule. However the reverse is not necessarily applicable. PSD has a simple matrix that it uses (see http://www.pesticides.gov.uk/uploadedfiles/Web_Assets/PSD/Regulatory_Update_15-2005_AnnexA_Tables_Flow-chart(1).pdf) but note the caveats that apply due to the uncertainties of such an approach. It is possible that such an approach might not be acceptable to all MS in which case some alternate guidance e.g. "no extrapolation is possible" would make it clear to applicants that formulation specific data are required if default values are not to apply. In order to achieve annex 1 listing under 91/414, only one lead product needs to be considered. Providing guidance on how to use data on the dermal absorption of the lead product for other products could increase consistency of assessments for registration across member states. 2.Even though it is clearly mentioned in the OECD test guideline, in vitro dermal absorption studies are still being performed without adequate confirmation of the active substance solubility in the receptor fluid. SOme text addressing this should be provided along the lines of: - " for active substances with low water solubility (e.g. <100mg/L) solubility in the receptor fluid should be demonstrated to be greater than 10 times the amount diffusing into the receptor fluid

No	Institution	Chapter	Comment
			during the study.". If the active substance is poorly soluble in the receptor fluid, the results of the study can be significnaly influenced by the limiation on the solubility. 3. The current text has no guidance on tape stripping. Clear guidance should be developed on the performance of tape stripping e.g. strips should be analysed individually so that the profile within the stratum corneum can be determined. Also on which tape strips can be considered to represent non-available material under all circumstances e.g. the first 3 strips and those which might be excluded if there is clear evidence of absorption and excretion declining (see previous comments on section 4). 4. It could be useful to provide guidance on how to deal with poor recoveries under different conditions. For example if all animals / wells have a recovery of <92% and the dermal absorption value is 3% is it valid to assume all the missing material could have been absorbed - this could make a 3 fold difference to the result? Instead of adding in all the missing material to the absorbed fraction is it better to multiply all values up ass by 100/92? If one or two samples have low recoveries but others are ca 100% it is possible to compare the high and low recovery samples to see where the losses might be occurring. Under what circumstances can recoveries be considered to be so poor that the results are meaningless and the study should be discarded?
79	French Food Safety Agency, AFSSA	6. Comments for additional chapters	We propose to add a section to Point 4 "4.5 Dermal absorption based on studies conducted on other formulations" When specific experimental data are missing, available information from other products can be used to predict the dermal absorption rate in the evaluated product. We suggest a possible guidance to help the assessor to decide the basic rules where the extrapolation is acceptable. Examples are given below: - differences on the a.s. concentration (same type of formulation and same vehicle)Extrapolations are possible for the diluted (if similar dilutions) and for the concentrate (if similar concentrations). - vehicle is different (same a.s concentration and same type of formulation)For liquids, if the (main) solvents belong to the same chemical family, extrapolation are acceptable for both diluted (same in-use concentrations) and concentrate.Concerning solid formulations with different inerts and formulants of same chemical family, extrapolation are possible for both diluted (same in-use concentrations) and concentrate. different type of formulations (same a.s concentration and similar vehicle/inerts) WP WDG yes, for both concentrate and diluted, SC WP, WDG yes for diluted EC, SC, SL, WP, WDG, (concentrate)GR yes = worse case

No	Institution	Chapter	Comment
80	Charles River Laboratories, UK	6. Comments for additional chapters	General suggestions: There is very little discussion on the issue of the stratum corneum. There is no discussion on Ficks 2nd Law of diffusion. Since absorption is Ficks 2nd Law of diffusion, it is important to define what is the semi-permeable membrane. It is generally accepted that the membrane is actually the stratum corneum. This makes it essential to include the "viable" skin, but not the stratum corneum in the risk assessment (in line with the citation for EPA, 1992). However, it does not take into account the potential stratum corneum reservoir. With stratum corneum profiling detailed, it is much simpler to decide if all/ some/ none of the stratum corneum should be included in the risk assessment especially if compared with the absorption profile.
81	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	6. Comments for additional chapters	Re-entry worker exposure: Under conditions of Good Agricultural Practice, workers re-entering a treated field may be exposed to treated plants and thus to dried residues of the active ingredient present on the plant surface. In dermal absorption studies, normally two or three concentrations are tested, i.e. - the undiluted formulation to mimic exposure during mixing/loading (high skin area concentration), and -1 or 2 aqueous spray-strength dilutions to mimic exposure during spray application of the diluted product (low skin area concentration). Guidance is required on how to derive dermal absorption estimates to adequately reflect the exposure scenario for re-entry workers. This will require guidance on experimental testing to cover re-entry worker exposure.
82	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	6. Comments for additional chapters	Comment - Read Across Guidance on the concept of '*Read Across'* is required as under 91/414 dermal absorption values are normally generated for an active substance in a single formulation type. Registration of the active substance under Annex III may require modified or different formulation to fulfil specific Member State agricultural requirements to meet the needs of the different application systems, crop or target pest. *Read Across* was developed under the REACH directive for chemical categories or analogues based on the similarities of physico-chemical properties and structural similarities. However, for PPPs, unlike chemicals assessed under REACH, the impact of formulation type on the dermal absorption of the active substance must be taken into consideration. *Read Across* can be applied to: 1) new active ingredient or 2) modified or new formulation type for an existing registered active substance.

No	Institution	Chapter	Comment
			1. New Active Ingredient In the absence of experimental data on a new active, a Tier One assessment can be made based on the similarity of an existing registered active(s), to indicate whether an active substance can be assigned default dermal absorption values of 1%, 5%, 10%, 25% or 50%. An example of the applicability of this approach is presented in Table 1 where the dermal absorption of either active can be predicted from the other. **Table 1 Potential for Read Across based on Physico-chemical Properties** **Structure**   **Compound** 1 2 **Molecular weight** 207.16 192 **Water Solubility** 200 g/L 142 g/L **Log K_{ow}** -2.87 -2.63 **Formulation Type** Oil in Water* Emulsion Soluble Liquid **Concentration** 30 g/L 97.5 g/L **Absorption Formulation** 1.70% (3% **) 2.36% (3.23%**) **Spray Concentration** 0.446 g/L 0.237 g/L **Absorption Spray Dilution** 2.91% (4.8%**) 1.48% (3.2%**) *active in water phase ** including Stratum Corneum *EFSA Scientific Report (2009) 282, 39-43* 2. Modified or New Formulation Type

No	Institution	Chapter	Comment
83	Dow Agrosciences, UK on behalf of the European Crop Protection Association (ECPA), Belgium	6. Comments for additional chapters	Stratum corneum- Tape stripping *in vivo* and *in vitro* studies OECD Guidance Document No. 28 describes how tapping stripping of the application site can be used to fractionated the amount of applied test substance residues in the different layers of the skin, specifically, the stratum corneum. This procedure is applicable to both *in viv*o and *in vitro* studies. The OECD guidance document mentions 15-20 tape strips for human skin. The number of tape strips taken determines the amount of detail provided on the distribution of the residue through the stratum corneum. Washing of the skin surface, both *in vivo* and *in vitro*, as required by the OECD test guidelines, leaves a proportion of the applied dose at the surface of the *stratum corneum*. This surface dose is not available for absorption and is readily removed by tape stripping. In effect, removal of this surface layer is equivalent to a washing procedure that mimics the efficiency of normal washing by an operator/worker. When 15 or more tape strips can be taken, at least the first two tape strips should be included with wash as non-absorbed dose. This has become an accepted procedure by many Member States. *In vivo* there are other options to determine if absorption from the application site (including the *stratum corneum*) has ceased (Section 3.2.1), but tape stripping can be useful to demonstrate potential movement of the residue within the *stratum corneum* over the duration of an *in vivo* study, which may provide additional information on the ultimate fate of the *stratum corneum* residue. *In vitro*, providing that the key aspects of OECD 428 have been followed, in particular, the ability of the test substance to partition freely into the receptor fluid, dermal absorption can be estimated from the receptor fluid alone. However, it is recognised that a proportion of the applied dose that remains within the skin sample following washing may have eventually diffused into the receptor fluid beyond the duration of the experiment. Although the normal process of desquamation removes chemicals lodged in the *stratum corneum* in vivo (Ramsey *et al*, 1992), in order to ensure the conservative nature of the *in vitro* approach, the proportion of the dose remaining in the skin preparation, in the absence of any additional information should be regarded as absorbed. Data on the potential absorption from the *stratum corneum* can be obtained by examining the profile of the residue with in the *stratum corneum* by plotting the amount present against the tape strip number (Figure 1):

No	Institution	Chapter	Comment
			 Example 1 demonstrates that movement through the stratum corneum has decreased to a minimum whilst Example 2 indicates that movement through the stratum corneum could be on going. The *stratum corneum* data should also be considered in conjunction with the residue in the epidermis/dermis and the receptor fluid. The relationship between the stratum corneum residue and epidermis/dermis compared with the relationship between epidermis/dermis and receptor fluid can be used to determine if the *stratum corneum* will become available. Guidance on such relationships and the outcome are given by the theoretical data in the following table, which contains three examples of receptor fluid values and three differing potential scenarios for *stratum corneum* and epidermis/dermis residues.

No	Institution	Chapter	Comment

Example	SC	Epidermis/ dermis	Total skin	Receptor Fluid	Components in Absorbed Dose	Absorbed Dose
1a	>10	0.5	10.5	0.5	Epidermis/dermis plus receptor fluid	1
1b	5	5.5	10.5	0.5	Epidermis/dermis plus receptor fluid/part of the SC	6 +
1c	<2	8.5	10.5	0.5	Epidermis/dermis plus receptor fluid / SC excluding strips 1 and 2	9+
2a	>10	2.5	12.5	10	Epidermis/dermis plus receptor fluid	12.5
2b	5	5	10	10	Epidermis/dermis plus receptor fluid and part of the SC	15+
2c	<3	7	10	10	Epidermis/dermis plus receptor fluid / SC excluding strips 1 and 2	17+

No	Institution	Chapter	Comment

3a	>10	2	12	25	Epidermis/dermis plus receptor fluid	27
3b	7	8	15	25	Epidermis/dermis plus receptor and part of the SC	33+
3c	<5	10	15	25	Epidermis/dermis plus receptor fluid / SC excluding strips 1 and 2	35+

SC Stratum corneum

Examples 1a, 2a and 3a clearly indicate that movement from the *stratum corneum* is negligible because levels in the epidermis are relatively low. Example 1c, 2c and 3c indicate a likelihood that the *stratum corneum* residue becomes systemically available with the exception of the surface layers. Guidance on the fate of the remaining *stratum corneum* can be obtained from examining the distribution of the residue through the *stratum corneum* (Figure 1). Examples 1b, 2b and 3b require expert judgment. A conservative approach that could be applied would be to exclude in all cases a minimum of the next 5 tape strips (i.e., 2 to7) as non-absorbed due to potential desquamation. In order to ensure the conservative nature of the *in vitro* approach, the proportion of the dose remaining in the skin preparation, in the absence of any additional information should be regarded as absorbed, unless there is clear evidence from the tape strip profile and/or the absorption profile or flux that absorption from the *stratum corneum* is negligible.

Trebilcock KL, Heylings JR and Wilks MF. In vitro tape stripping as a model for in vivo skin stripping. Toxicology In Vitro, 8, 665-667, 1994.

Ramsey JD, Woollen BH, Auton TR, batten PL and Leeser JE. Pharmacokinetics of fluazifop butyl in human volunteers II: dermal dosing. Human and Experimental Toxicology, 11, 247-254, 1992.

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