# Chapter 14
# Non-clinical Safety Studies for the Conduct of Human Clinical Trials for Pharmaceuticals: ICH M3 and M3(R2)

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**Abstract** The extent of non-clinical safety testing to support clinical trials at different stages of development differed greatly between the EU, Japan and the USA prior to the adoption of the original ICH M3 guidance in 1997. The guideline achieved some notable harmonizations, but there was still significant disharmony, especially around the duration of dosing for non-rodents and the timing and extent of reproductive toxicology studies to support trials in women of childbearing potential. The inability to harmonise on these particular issues led to a reluctant acceptance of finalising the M3 guidance.

In 2006, a revision of ICH M3 commenced with an aim to remove the un-harmonised components. Although the M3 guideline is essentially concerned with the timing of non-clinical studies in relation to clinical development, further topics were also introduced by the Expert Working Group during the discussions. The ICH M3(R2) document was signed off by the regulators in June 2009. While the 2000 version of the guideline had 6 pages of text, the revision had 27. All the objectives had been largely met and with only one minor difference still in place.

## 14.1 Introductory Comments

The extent of non-clinical safety testing to support clinical trials at different stages of development differed largely between the EU, Japan and the USA prior to the adoption of the ICH M3 guidance in 1997. These regional differences had been

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highlighted and discussed at one of the safety workshops held at the First International Conference of Harmonization in Brussels in October, 1991 (Scales 1991). Although regional differences were apparent to international pharmaceutical companies making clinical trial submission in several regions, the exact differences were difficult to pinpoint as there was no clear guidance published on the timing of non-clinical safety studies in relation to stage of clinical development in either Japan or the USA, and in the EU community, there was only a draft guidance available with no formal status.

Although the ICH process early identified the need for the establishment of clear and internationally harmonised recommendations on the extent of non-clinical safety studies needed to support clinical trials of different phases, it was not until 1994 the "M3 project" was formally adopted as an ICH topic and an Expert Working Group (EWG) was formed. This delay in the initiation of the more formal work with the M3 guidance was logical considering that several important ICH safety topics related to the scope of the M3 guidance were at the initial stage of development, and thus the timing question which could not be addressed prior to these safety topics had become more mature in terms of regional harmonization. For example, until there had been a position on the type of genotoxicity studies that would be acceptable to support a marketing authorisation (ICH S2B), it would be impossible to adopt a clear and understandable guidance what type of data would normally be needed to support clinical trials of different stages/durations. Other examples of specific ICH safety topics worked on were the guidance on Duration of Non-Rodent Toxicity Testing (S4), Reproduction Toxicity (S3) and Carcinogenicity [particularly S1A (Need for Carcinogenicity Testing of Pharmaceuticals)].

The object of the ICH Steering Committee to initiate work on a regulatory guidance on the timing of non-clinical safety studies was obviously to create international harmonization, i.e. harmonization of the recommendations/requirements from the regulatory authorities in EU, Japan and the USA. However, the lack of clear regional positions on many timing aspects prior to the initiation of this work pushed the regional authority groups and the regional industry groups to formulate updated positions on these aspects. This meant that the discussions and negotiations were made from an essentially equal playing field which fostered an open and constructive dialogue. It is perhaps unknown to many that the M3 guidance that was adopted by the three regional authorities in 1997 had, from a formal point of view, a weak position in the EU. At this time, there was no European Clinical Trial Legislation (the EU Clinical Trials Directive did not come into force until May 2004), and thus the guidance was not binding to the EU member states even though the CPMP had adopted the guidance. However, in view of the lack of guidance relating to the extent of non-clinical safety testing to support clinical trials and divergent regulatory scrutiny of clinical trial applications within the EU member states, the development of the ICH M3 guidance was perhaps of particular significance for EU in that it both catalysed necessary harmonization within EU and provided an important basic element for the forthcoming EU Clinical Trial Directive.

The EU, Japanese and US pharmaceutical companies were understandably eager proponents of the M3 guidance. Since a large proportion of the companies were working on the international market, they were keen to obtain harmonization on this important subject particularly as inconsistent regulatory request for non-clinical safety studies to support either national or multinational clinical trials slowed clinical development and thereby incurred additional costs. As indicated above, the regulatory participants of the EWG from the three regions were also eager to work towards a harmonised guidance in part because of the awareness that the lack of clear regional guidance was very unsatisfactory for drug developers and that without some type of harmonization of the timing issues, the achievements made in harmonising the technical standards for non-clinical safety testing (the other ICH topics) would not be fully appreciated.

## 14.2 Overall Content of the M3 Guidance

Once the ICH Steering Committee in 1994 approved a proposal that work should commence on a guidance addressing timing of safety studies in relation to clinical trials, the appointed Expert Working Group rapidly came to an agreement that the guidance should focus on the following principle areas of toxicity testing supporting clinical development:

- Safety pharmacology studies (effects on vital organ systems)
- Single- and repeat-dose toxicity studies
- Genotoxicity and carcinogenicity studies
- Toxicity to reproduction

Moreover, it was agreed that the principles of toxicokinetics needed a prominent place in the guidance since this was becoming more generally recognised as a fundamental part of non-clinical safety assessment. Other safety areas that were included without any controversy were local tolerance data and data to support clinical trials in paediatric populations.

## 14.3 Safety Pharmacology Studies

It is interesting to note that in the initial review of the timing issue presented to the participants of ICH I in Brussels in 1991 (Scales 1991), safety pharmacology was not included as one of the areas that needed to be addressed from a timing perspective. However, the request for an assessment of effects on vital functions such as cardiovascular, central nervous and respiratory system was soon incorporated in the guidance by the EWG and without much controversy. The wording “assessment of effects on vital functions” was carefully chosen to imply that such information could be

obtained in conjunction with single dose or more likely repeat-dose toxicity studies. This was also mentioned in the guidance.

The ICH M3 guidance from 1998 (ICH M3 1997) contains no reference to whether the non-clinical safety studies should be conducted according to GLP. Since this was already a requirement for all toxicity studies, it was argued by many EWG members (particularly those from the FDA and US Pharma) that this did not need to be included in the document. However, as there at the time was no specific guidance on safety pharmacology, studies conducted to assess the effect on vital organ system could, at least in some regions, be conducted without GLP compliance. In Europe, there was already a CPMP guidance clarifying the 91/507/EEC Directive regarding GLP and safety tests stating that "pharmacodynamic studies designed to test potential for adverse effects" must conform to GLP (CPMP III 3824 92 Rev). Thus, even though, as mentioned above, there was no harmonization of the legislation relating to applications and conduct of clinical trials within the EU, safety pharmacology data, when submitted for a marketing authorisation application, was expected to be derived from GLP-compliant studies. It should be noted that the ICH S7A guidance from 2001 (ICH S7A 2000) that addresses the specifics of safety pharmacology testing does address the GLP issue of safety pharmacology data. It is encouraging that this guidance gives some flexibility with regard to GLP compliance.

## 14.4 Single- and Repeat-Dose Toxicity Studies

The timing of single-dose toxicity studies was obviously of no controversy as such data should logically be available prior to first dose in humans. At the time when the work with M3 was initiated, international harmonization had already been achieved with regard to the number and of type of single-dose studies needed to support human clinical trials (Ohno 1991). Although several M3 EWG members likely felt that specific acute toxicity studies had limited value for human risk assessment, it was not possible to reopen an issue that was just recently harmonised and promoted as a major achievement of the ICH process. It is therefore of great satisfaction that the revised M3 document from 2009 (ICH M3(R2) 2009) abandons the request for specific GLP-compliant single-dose studies with two routes of administration and instead recommends that acute or single-dose toxicity information may be derived from dose-finding studies to support dose setting of repeat-dose studies.

One of the most difficult areas to harmonise during the entire ICH process has been the extent of repeat-dose toxicity data needed to support clinical trials of different stages and durations. This difficulty surfaced already at the beginning of the work with the M3 guidance as reported at the Third International Conference of Harmonization in Yokohama in 1995 (Hayashi 1995; Sjöberg 1995). Consensus could not be reached in the following areas:

(a) Duration of rodent toxicity studies to support single-dose and repeat-dose trials of up to 14 days (Japan requested 4-week rodent studies, while EU and USA accepted 2-week rodent studies)
(b) Duration of rodent and non-rodent studies to support phase III trials (EU requested longer study durations than Japan and USA)
(c) Duration of non-rodent studies needed to support clinical trials with a duration of more than 6 months (USA needed studies of 12-month duration, while EU was content with a 6-month study duration)

These regional differences were essentially based on what was considered a safe approach for clinical trial testing, i.e. no solid data was presented by either region to support its specific position. The Japanese argued that a 4-week rodent toxicity study was needed to assess the potential of the drug in question to interfere with male fertility and that such an assessment was needed for even the shortest clinical trial in humans. The EU regulators on the other hand argued that for confirmatory clinical trials, i.e. phase III trials, more solid toxicity data (longer term studies) was needed to establish the true toxicity profile and assess patient safety compared to the exploratory trial situation where homogeneity of patient population and patient numbers were different. Finally, the USFDA argued that in their experience, there was additional value of having non-rodent toxicity data from 9- or 12-month exposure duration as compared to the 6 advocated particularly by the EU. Owing to the failure of the EWG in reaching consensus on the maximum duration of non-rodent repeat-dose testing, a specific EWG was set up, and the ICH S4 topic was created in 1997, i.e. just prior to the finalisation of the M3 guidance. Based on an assessment of a limited data set of non-rodent repeat-dose toxicity studies that covered both 6- and 12-month exposure, the S4 EWG was also unable to come to a consensus on the maximal duration of repeat-dose studies (ICH S4 1998). Although the ICH Steering Committee was not pleased with the inability of the EWG to harmonise the timing and duration of repeat-dose toxicity studies, the EWG members, particularly those from the regulatory side, were less concerned with this inability. Their views were incorporated in the guidance, and it was felt that the differences did not overshadow the overall achievements in harmonising timing issues.

## 14.5 Genotoxicity and Carcinogenicity Studies

In co-operation with the EWG on genotoxicity, the M3 EWG rapidly came to a consensus that phase I clinical trials should be amply supported by the so-called ICH standard battery of in vitro studies while phase II trials should be supported by the complete set of ICH-compliant in vitro and in vivo studies (Mayahara 1995). In hindsight, one may question the total logic in this rigid separation between data request for phase I and those for phase II trials. The exposure duration may certainly

be longer in some phase I trials than in some phase II trials, and a recommendation based on exposure duration rather than what stage of development the drug is at would be a more logical approach considering that a genotoxic liability would likely be strongly linked to exposure duration and total dose. It should be noted that the updated M3 guidance (ICH M3(R2)) states that single-dose trials are generally supported by an assay for gene mutation, and thus this new guidance has adopted, from this perspective, a more logical approach to request for genotoxicity data. However, multiple-dose phase I studies, regardless of exposure duration, are seemingly still supported by the in vitro studies, while the complete set of in vitro and in vivo studies are needed prior to the conduct of phase II trials, again regardless of exposure duration.

The issue of timing of carcinogenicity studies was not primarily dealt with by the M3 EWG since a specific guidance relating to the "Need for Carcinogenicity Studies" (ICH S1A) was developed and adopted as a step 4 document by the ICH Steering Committee already in November 1995, i.e. shortly after the initiation of the work on the M3 guidance. In relation to the timing issue, the S1A guidance concluded that "when carcinogenicity studies are required they usually need to be completed before application for marketing approval", and unless there is specific concern, carcinogenicity data would not be needed prior to the conduct of large clinical trials. An example of where regulators subsequently have recommended carcinogenicity testing prior to large-scale clinical trials/patient treatment duration longer than 6 months is with the peroxisome proliferator-activated receptor (PPAR) agonists (CDER 2008; EMEA/341972/2006).

## 14.6 Toxicity to Reproduction

Similar to the situation for the timing of repeat-dose toxicity testing, the three regions, EU, Japan and USA, could not reach complete consensus of what toxicity to reproduction studies was needed to support the inclusion of men and women of childbearing potential in clinical trials of different development stage. Consensus could not be reached in the following two areas:

(a) Extent of repeat-dose data to make an assessment of potential to interfere with male fertility (in Japan, a 4-week repeat-dose toxicity study was considered essential to assess toxicity to the male reproductive system, whereas in EU and USA, 2-week toxicity studies were considered sufficient for an overall assessment of potential toxicity).
(b) Type of data needed to include women of childbearing potential, using highly effective birth control, in shorter-term clinical trials (assessment of female fertility and embryo–foetal development is needed in Japan, and embryo–foetal development studies are needed in EU, whereas in USA, women of childbearing potential could be included in "early, carefully monitored" clinical studies provided that "adequate precautions were taken to minimise risk").

It may be difficult to appreciate the original Japanese position that a 4-week toxicity study was needed to assess potential effect on male fertility even for a drug that is to be given for a single dose, but then one need to consider that in Japan, male fertility studies were needed to support inclusion of men in any clinical trial prior to the adoption of the M3 guidance in 1997. The Japanese should also be credited for performing experimental studies to support their new position that a 4-week rat toxicity study was sensitive to pick up potential effects on male reproductive organs (Takayama et al. 1995). In an additional collaborative study in Japan, rodent data were obtained supporting the position that 2-week toxicity studies in rats were as sufficient as 4-week studies to identify male reproductive toxicants (Sakai et al. 2000). The M3 guidance was therefore updated to include this new position of the MHW (ICH M3(R1)) and has been kept in the most updated version of ICH M3, i.e. ICH M3(R2).

The EWG discussions on the acceptability of including women of childbearing potential with adequate contraception in early clinical trials were fairly straightforward with no real attempt by either the EU or Japanese regulators to convince the USA that the more stringent position was correct. The EU regulators of the EWG had clear sympathy for the US position as their approach seemingly had been shown to work safely. When this position was forwarded to the CPMP, strong oppositions were given from a couple of the leading members, and there was thus no way a change in attitude could come about from EU regulators. The inability to harmonise on this particular issue and on the maximum duration of non-rodent toxicity studies was almost sufficient to stop the work on the M3 guidance by EU regulators not close to the actual work. When the overall benefits of all the harmonization that were achieved were enforced, there was a reluctant acceptance of finalising the M3 guidance. It is noteworthy that the Japanese and EU regulators have moved its position on this topic to the position USFDA was in 1998 (ICH M3(R2)).

## 14.7 ICH M3(R2)

At the ICH Steering Committee in early 2006, it was agreed that the ICH M3 guideline required further revision to try and achieve closer harmonization in non-clinical testing of pharmaceuticals. The issues to be discussed in the revision process were agreed and included the nature and timing of reproductive toxicity studies to support the conduct of different phases of clinical trials, the duration of repeated-dose toxicity studies to support the conduct of different phases of clinical trials, the duration of chronic toxicity studies in non-rodents, the requirement of the toxicity package to support first entry into human and the definition of the role of the M3 guideline in the development of biotechnology derived.

Although the M3 guideline is essentially concerned with the timing of non-clinical studies in relation to clinical development, further topics were introduced by the Expert Working Group during the discussions and included the removal of the need to keep single-dose toxicity studies as a fixed requirement prior to first human exposure.

In addition to regulatory and industry representatives for the three ICH regions, the working group also included observers from the European Free Trade Association (EFTA), Health Canada and the interested parties, the International Generic Pharmaceutical Alliance (IGPA) and the Biotechnology Industry Organisation (BIO).

The discussions surrounding the scope of the revised guideline centred on achieving a document that should facilitate the timely conduct of clinical trials, reduce the use of animals in accordance with the 3R (reduce/refine/replace) principles and reduce the use of other drug development resources.

The discussions surrounding whether single-dose toxicology studies were needed were supported by a publication on a European pharmaceutical company initiative challenging the regulatory requirement for acute toxicity studies in pharmaceutical drug development (Robinson et al. 2008) that followed on from work in 2007 by the UK's National Centre for the 3Rs (NC3Rs).

A considerable amount of time was taken to try and harmonise the nature and timing of reproductive toxicity studies to support the conduct of different phases of clinical trials. Industry associations from all three ICH regions provided vast databases on reproductive toxicology studies and publications on work to evaluate toxicity on male reproductive organs by 2-week repeated-dose toxicity studies in rats (Sakai et al. 2000) and on the evaluation of ovarian toxicity by repeated-dose and fertility studies in female rats (Sanbuissho et al. 2009). The final document almost achieved complete harmonization, with only one minor difference still in place. In the United States, assessment of embryo–foetal development could be deferred until before phase III for women of childbearing potential (WOCBP) using precautions to prevent pregnancy in clinical trials. In the EU and Japan, small numbers (about 150) of WOCBP could now be included in clinical trials of short duration, but definitive non-clinical developmental toxicity studies were still needed to be completed before exposure of large numbers. This represented a significant shift in opinion from the EU as previously most member states would not allow clinical trials in WOCBP without the results from reproductive toxicology studies.

The ICH M3(R2) document was signed off by the regulators in June 2009. The title had changed and was now "Guidance on the Non-Clinical Safety Studies for the Conduct of Human Clinical Trials and Marketing Authorisations for Pharmaceuticals". While the 2000 version of the guideline had 6 pages of text, the revision had 27. All the objectives had been met, and, in addition, the new guideline had sections on High Dose Selection for General Toxicity Studies, i.e. Safety Margins, Metabolites in Safety Testing, Estimation of the First Dose in Humans, Exploratory Clinical Trials, Immunotoxicology, Phototoxicity, Non-Clinical Abuse Liability and Combination Drug Non-Clinical Testing. These sections were added as it was agreed that there was a lack of regulatory guidance in these areas. The new guideline also included reference to juvenile animal toxicology studies to support clinical trials in paediatric populations.

The new section on Exploratory Clinical Trials was extremely interesting. It was recognised that in some cases earlier access to human data can provide improved insight into human physiology/pharmacology, knowledge of drug candidate characteristics and therapeutic target relevance to disease. The USFDA had

already published guidance on this subject, and the EU's Safety Working Party had published a concept paper and was drafting their guideline. Once it was decided to include a section within ICH M3, the EU stopped their independent work. While ICH M3 is essentially a timing document, i.e. it advises when studies are required during clinical development; this new section included definitive advice on the type and design of studies.

Exploratory clinical studies for the purpose of this guidance were considered to be those intended to be conducted early in phase I, involve limited human exposure, have no therapeutic intent and were not intended to examine clinical tolerability. Recommended starting doses and maximal doses for the five approaches were also included. Five study types were included, two micro-dose designs, one single-dose design and two repeated-dose designs, one based on the FDA guidance and one based on the "EU approach".

Another notable achievement was the addition of the new section "High Dose Selection for General Toxicology Studies". Generally, in toxicity studies, effects that are potentially clinically relevant can be adequately characterised using doses up to the maximum tolerated dose (MTD). Other equally appropriate limiting doses include those that achieve large exposure multiples or saturation of exposure or use the maximum feasible dose (MFD). This section was added to prevent the use of doses in animals that would not add value to predicting clinical safety, and the recommendations were consistent with those for ICH reproduction and carcinogenicity study designs that already had defined limit doses and/or exposures.

While still in its early phases of the implementation, and even though the document had been released for public comments on two occasions, the complexity of the guidance, its broader scope and the numerous changes in recommendations from the ICH M3(R1) guidance generated questions that could have impacted on its successful implementation.

Several of these questions and issues were considered to be outside the scope of the guideline, while others were addressed by question and answer (Q&A) documents that were released in 2011 and 2012. The issues covered in the Q&A documents were limit doses, exploratory clinical trials, reversibility of findings, metabolite testing, juvenile animal toxicology studies, reproductive toxicology studies and safety pharmacology.

## 14.8 Concluding Remarks

The first international guidance document addressing the aspect of timing on non-clinical safety studies in relation to clinical trials should in hindsight be viewed as a success for regulators and pharmaceutical companies alike although complete harmonization could not be reached on all timing issues. It was particularly useful for the member states of the EU that from 2004 were obliged to follow the EU Clinical Trials Directive (2001/20/EC). The M3 guidance document was obviously an important component of the implementation of this guidance.

The inability of the three regions to harmonise on all areas of non-clinical safety testing to support clinical trials should be judged from the fact that an exact value of non-clinical safety testing in predicting human safety cannot be given, and therefore there will always be an element of personal/regulatory agency judgment in defining what type of studies is necessary to safeguard patients in a particular clinical trial situation. The significant expansion of the ICH M3(R2) document and the subsequent issuing of explanatory Q&A documents are interesting and are probably further reflections on this point.

Lastly, it should be emphasised that the ICH process has overall made very significant contributions in underpinning the scientific basis for various standards and recommendations by encouraging retrospective analysis of non-clinical safety data and the initiation of prospective studies. Many of these contributions have had a direct impact on recommendations made in the M3 document. If such efforts continue, the M3 document will maintain its status as one of the most important non-clinical regulatory documents.

## References

2001/20/EC. Directive 2001/20/EC of the European Parliament and of the Council of 4 April 2001on the approximation of the laws, regulations and administrative provisions of the Member States relating to the implementation of good clinical practice in the conduct of clinical trials on medicinal products for human use. Official Journal of the European Communities 1.5.2001. http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2001:121:0034:0044:en:PDF

CDER (2008) Guidance for industry. Diabetes Mellitus: developing drugs and therapeutic biologics for treatment and prevention. Center for Drug Evaluation and Research, Feb 2008. http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM071624.pdf?utm_campaign=Google2&utm_source=fdaSearch&utm_medium=website&utm_term=PPARguidance2008&utm_content=2

CPMP III/3824/92 Rev.1. Committee for Proprietary Medicinal Products. Applicability of good laboratory practice. Commission of the European Communities

EMEA/341972/2006. CHMP SWP reflection paper on PPAR's (Peroxisome Proliferator Activated Receptors), Sept 2006. http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2009/11/WC500015479.pdf

Hayashi (1995) Duration of repeated dose studies to support first entry into humans. In: D'Arcy and Harron (eds) Proceedings of the third international conference on harmonization, Yokohama, pp 342–343

ICH M3 (1997) Nonclinical safety testing for the conduct of human for pharmaceuticals. ICH, July 1997. http://ocw.jhsph.edu/courses/drugdevelopment/PDFs/FDA_Guidance_on_NonCLinical_Safety_Studeis.pdf

ICH M3(R2) (2009) Guidance on nonclinical safety studies for the conduct of human clinical trials of and marketing authorization for pharmaceuticals. 11 June 2009. http://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Multidisciplinary/M3_R2/Step4/M3_R2__Guideline.pdf

ICH S4 (1998) Duration of chronic toxicity testing in animals (rodents and non-rodents). 2 Sept 1998. http://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Safety/S4/Step4/S4_Guideline.pdf

ICH S7A (2000) Safety pharmacology studies for human pharmaceuticals. 8 Nov 2000. http://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Safety/S7A/Step4/S7A_Guideline.pdf

Mayahara (1995) Progress of the ICH guideline. In: D'Arcy and Harron (eds) Proceedings of the third international conference on harmonization, Yokohama, pp 339–342

National Centre for the Replacement, Refinement and Reduction of Animals in Research (2007) Challenging requirements for acute toxicity studies: Workshop report

Ohno (1991) Toxicity testing: regulatory perspectives. In: D'Arcy and Harron (eds) Proceedings of the first international conference on harmonization, Brussels, pp 186–188

Robinson S, Delongeas JL, Donald E, Dreher D, Festag M, Kervyn S, Lampo A, Nahas K, Nogues V, Ockert D, Quinn K, Old S, Pickersgill N, Somers K, Stark C, Stei P, Waterson L, Chapman K (2008) A European pharmaceutical company initiative challenging the regulatory requirement for acute toxicity studies in pharmaceutical drug development. Regul Toxicol Pharmacol 50:345–352

Sakai et al (2000) Collaborative work to evaluate toxicity in male reproductive organs by repeated dose studies in rats – overview of the studies. J Toxicol Sci 25:1–21

Sanbuissho A, Yoshida M, Hisada S, Sagami F, Kudo S, Kumazawa T, Ube M, Komatsu S, Ohno Y (2009) Collaborative work on evaluation of ovarian toxicity by repeated-dose and fertility studies in female rats. J Toxicol Sci 34:SP1–SP22

Scales (1992) Timing of toxicity studies vs conduct of clinical trials – existing requirements. In: D'Arcy and Harron (eds) Proceedings of the first international conference on harmonization, Brussels, pp 322–329

Sjöberg (1995) Data to support the extension of clinical trials. In: D'Arcy and Harron (eds) Proceedings of the third international conference on harmonization, Yokohama, pp 343–345

Takayama et al (1995) A collaborative study in Japan on optimal treatment period and parameters for detection of male fertility disorders induced by drugs in rats. J Am Coll Toxicol 14:266–292