Health Canada Santé Canada

Therapeutic Products Programme
Holland Cross, Tower "B"
1600, Scott Street
Address Locator # 3102D1
OTTAWA (Ontario)
K1A 1B6
February 14, 2000

00-002145

To: Associations

Subject: Stereochemical Issues in Chiral Drug Development

I am pleased to inform you of the release of Therapeutic Products Programme (TPP) guidance document entitled *Stereochemical Issues in Chiral Drug Development*. This Guidance for Industry replaces the draft of the same title posted on the TPP Website in July 1998.

This document is intended to provide drug submission sponsors with guidance on issues specific to chiral pharmaceuticals that should be addressed during drug development.

The comments that were received in response to my letter of July 9, 1998, have been reviewed and suggestions have been incorporated into the current guidance document, where appropriate. For your information, a summary of the more significant comments, together with our analysis, is attached to this covering letter (Attachment I). We hope that this information is useful to you.

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This Guidance for Industry is effective as of May 1, 2000. The document may be found on TPP's Website (www.hc-sc.gc.ca/hpb-dgps/therapeut/) under the path Guidelines / Chemistry and Manufacturing Guidance Documents. Should you have any questions or comments, please do not hesitate to contact:

Mr. Gary Condran
Division of Pharmaceutical Quality,
Bureau of Pharmaceutical Assessment,
Therapeutic Products Programme,
Health Canada
A.L. 0202A2
Finance Building, Tunney's Pasture
OTTAWA, Ontario
K1A 1B6

Telephone: (613) 957-3192
Facsimile: (613) 941-0571
E-mail: gary_condran@hc-sc.gc.ca

(Original Signed by)

Dann M. Michols
Director General

Enclosure

ATTACHMENT I

CONSULTATION FOR: Stereochemical Issues in Chiral Drug Development

Comments concerning the draft guidance were solicited from the stakeholders. The comments that were received have been reviewed and suggestions have been incorporated into the current guidance document, where appropriate. A summary of the more significant comments, together with our analysis, is outlined below. The comments have been organized according to the various sections of the guidance document.

Scope:

- The Scope could be interpreted to cover drugs with several chiral centres. For a number of examples, it will be virtually impossible to prepare the antipode and, in any case, conversion of the enantiomeric drug to its antipode may not be an issue.

  Response: It is recognized that scientific considerations and/or technical limitations may preclude the application of some of the concepts described in the guidance document (e.g., drugs with multiple chiral centres). A statement to this effect has been added to the section.

Preamble:

- The requirement for using enantioselective assays during drug development and subsequent studies was opposed. It was suggested that a non-enantioselective assay, coupled with a method for determining the opposite enantiomer as an impurity, should be sufficient.

  Response: The Programme considers that it would be necessary to employ enantioselective assays under certain situations.

Chemistry and Manufacturing:

- The stage at which conducting in-process testing for chiral intermediates should not be specified.

  Response: The in-process testing has been clarified to the effect that the testing for the identity and purity should be enantioselective for key intermediates in which additional chiral centre(s) have been introduced.

- The requirement for the control of enantiomers in a racemic mixture is excessive, and a specific test over and above optical rotation for ensuring that spontaneous resolution has not occurred during the purification of the bulk drug exceeds standard analytical requirements.

  Response: The guidance does not require the control of enantiomers in a racemate, whether the racemate is a racemic compound or a racemic mixture. The guidance document suggests that during the drug substance development, a test for the identity of the racemate should be performed to ensure that spontaneous resolution has not occurred during the purification of the bulk drug. Optical rotation may be considered adequate for this investigation.

- The requirement for the investigation of the enantiomeric purity of the drug substance in the finished product during the stability studies conducted to determine shelf life should be deleted. Studies conducted during drug development would be sufficient.

  Response: It is considered necessary to monitor the enantiomeric purity of the drug substance in the drug product during the stability studies conducted to determine shelf life. Results from primary stability studies may be considered sufficient. However, a test for enantiomeric purity should be incorporated into the drug product specification if results of these investigations warrant.

Bioequivalence of Solid Oral Dosage Forms:

- One respondent asks how a drug substance will be handled if the information on the *in vivo* disposition of each enantiomer of a marketed racemate is unknown. Further, the respondent suggests that the most important issue will be to require a baseline level of information on the racemate be available (and generated if not known) to allow a strategy to be developed based on the facts associated with the racemate and the proposed single enantiomer.

  Response: Information such as this would be particularly relevant to new drug development and as such, is addressed in earlier portions of the guidance document. In the case of comparative bioavailability studies conducted to compare products of differing type, e.g., a Group II product as defined in *Conduct and Analysis of Bioavailability and Bioequivalence Studies - Part B: Oral Modified Release Formulations* (Guideline B), information regarding the pharmacokinetic properties of the enantiomers would be necessary in order to justify the use of a non-stereoselective assay in studies involving a chiral drug. As outlined below, instances where the *in vivo* disposition of each enantiomer is pertinent to the bioequivalence assessment of two oral solid dosage forms of similar type containing a defined ratio of enantiomers would be rare. It is anticipated that, if such a situation were to arise, the enantiomeric behaviour of such a compound would be widely known.

- One respondent suggests that the issue at hand is whether two drug products containing a chiral drug, that have been shown to be bioequivalent with a non-stereoselective assay, can actually be shown to be non-bioequivalent with a stereoselective assay. The respondent goes on to provide an analysis of the literature to show that data demonstrating such an instance, that meets the rigour of existing bioequivalence standards, is not currently available in the literature.

  Response: Literature suggests there are a limited number of situations in which the use of a stereoselective assay might be argued to be necessary when assessing the bioequivalence of two formulations. Those are as follows: when changes in oral input cause changes in the *in vivo* ratio of enantiomers due to a phenomenon such as high first pass metabolism of the active enantiomer and secondly, when there is a relatively low first pass metabolism of the active enantiomer and a specific isomer ratio is important for optimal therapeutic effect. However, based on a review of the existing literature, data comparing similar types of dosage forms collected from appropriately designed (based on the principles in current TPP guidelines) comparative bioavailability studies, does not appear to be available to indicate that two products found to be bioequivalent based on the application of TPP assessment standards to total drug levels, would not also be found to be bioequivalent based on individual enantiomer levels. That is, it has not been demonstrated that two oral solid dosage forms of similar type, containing the same ratio of isomers, that are bioequivalent based on total drug levels in appropriately

designed studies satisfying current TPP requirements, would produce clinically significant differences in individual enantiomer levels. Should such data become available in the future, it will be taken into consideration in future revisions of the guidance document. The present guidance document has been revised to reflect the current status of the available information.

- One respondent suggests that the only case where enantiomeric assays may be required is when first market entry (Group II) MR products are compared to an IR formulation.

  Response: As revised, the current guidance document indicates that stereoselective assays are not normally required when comparing products of similar type e.g., two immediate-release formulations, that contain the same ratio of isomers. The situation noted by the respondent does not meet these criteria, and as noted in the revised guidance document, a stereoselective assay may be required in such a case.

Health Canada Santé Canada

GUIDANCE FOR INDUSTRY
Stereochemical Issues in Chiral Drug Development

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Therapeutic Products Programme
Guidance Document

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1. INTRODUCTION

1.1 PURPOSE

This document is intended to provide sponsors of New Drug Submissions (NDS's) and Abbreviated New Drug Submissions (ANDS's) with guidance on issues specific to chiral pharmaceuticals that should be addressed during drug development. It complements the Programme's existing guidelines on *Chemistry and Manufacturing: New Drugs* and *Toxicologic Evaluation*.

The Therapeutic Products Programme recognizes that this document cannot address every possible situation and each submission will be considered and judged on its own merits.

1.2 BACKGROUND

It should be emphasized that the scientific and regulatory principles that underlie chiral drugs are not fundamentally different from those of non-chiral drugs; however, it has been recognized for some time that the chirality of drugs poses unique problems. Progress in enantioselective synthesis and enantioselective separation, and better understanding of the *in vivo* behaviour of enantiomers now permit the stereochemical issues of chiral drug development to be addressed from a regulatory perspective.

A Glossary of terms related to stereochemistry may be found in Appendix 1. With respect to nomenclature, chemical names should be expressed according to the International Union of Pure and Applied Chemistry (IUPAC) or Chemical Abstract Service (CAS) rules.

Stereoisomers are compounds made up of the same atoms bonded in the same sequence but having different orientations in space. The term stereoisomer encompasses diastereoisomers (including *cis-trans* isomers) and enantiomers. Diastereoisomers are chemically distinct and often pharmacologically different compounds, and can generally be separated by achiral analytical methods. Therefore, diastereoisomers should be developed separately, rather than as a mixture, unless *in vivo* interconversion of the isomers occurs or the mixture fortuitously represents a reasonable fixed dose combination. This class of stereoisomers is not the focus of this document.

Enantiomers are stereoisomers whose mirror images cannot be superimposed. Enantiomers have identical physical and chemical properties except that they rotate the plane of polarized light in opposite directions and behave differently in a chiral environment. Thus, they interact at different rates with other chiral compounds including many biological macromolecules.

Mixtures of equimolar amounts of enantiomers are called racemates. They may exist as racemic mixtures (conglomerates) or racemic compounds (true racemates). In the solid state, the physical properties such as melting points, solubilities and heats of fusion of the racemates may differ from those of the individual enantiomers.

Many drugs are marketed as racemates. There are numerous examples in the literature where the enantiomers in a racemate differ substantially with respect to their pharmacokinetics, pharmacodynamics, toxicity, protein binding, etc. With some drugs, one of the enantiomers is mostly responsible for a given pharmacological activity. The other enantiomer may be less active, inactive, toxic, or may give rise to an entirely different pharmacological response. Interactions between enantiomers have been also described. Consequently, enantiomers should be recognized as distinct substances.

1.3 SCOPE

This guidance document specifically deals with issues related to the development of enantiomers and their mixtures. It applies to pharmaceutical drugs, including synthetic drugs, semi-synthetic drugs, and drugs produced from fermentation or derived from natural sources. It does not apply to biologics or radiopharmaceuticals.

It is recognized that scientific considerations and/or technical limitations may preclude the application of some of the concepts described in the guidance document (e.g., drugs with multiple chiral centres).

1.4 PREAMBLE

The decision whether to develop a single enantiomer, racemate, or non-racemic mixture (enantiomeric mixture other than racemate) rests with the sponsor and should be based on scientific data relating to quality, safety and efficacy and ultimately to the risk/benefit assessment of the drug under the proposed conditions of use. Cases where the development of a racemate may be justified include, but are not limited to, the following:

a) The enantiomers are configurationally unstable *in vitro* or undergo racemization *in vivo*.

b) The enantiomers have similar pharmacokinetic, pharmacodynamic and toxicological properties.

c) It is not technically feasible to separate the enantiomers in sufficient quantity and/or with sufficient quality.

The cases where development of a non-racemic mixture may be justified include those where a specific enantiomeric ratio is expected to improve the therapeutic profile.

The requirements for drug submissions concerning chiral drugs are outlined in the sections that follow. It is emphasized that these requirements are not all encompassing and depending on the drug, may vary as to specifics. If it is believed that a particular drug requires more definitive guidance, the sponsor is encouraged to discuss the issues with the Programme in advance.

Enantioselective assays should be developed and validated at an early stage of drug development, and used wherever relevant unless it has been clearly demonstrated that a non-enantioselective assay provides results equivalent to those obtained with the enantioselective assay. A list of analytical methods for chiral drugs may be found in Appendix 2.

2. CHEMISTRY AND MANUFACTURING

The requirements of the Programme's guideline on *Chemistry and Manufacturing: New Drugs* are applicable to both chiral and non-chiral drugs. This section discusses additional requirements that are specific to chiral drugs.

2.1 SINGLE ENANTIOMER

2.1.1 Drug Substance

A full description of the manufacturing process used to obtain the individual enantiomer should be given. The identity and enantiomeric purity of chiral starting materials and chiral reagents should be established. In-process testing for identity and purity should be enantioselective for key intermediates in which additional chiral centre(s) have been introduced. Whenever possible, absolute configuration should be determined as part of structural elucidation studies.

The specification for the drug substance should include enantioselective tests for identity and purity. Optical rotation may be used for both, but in addition, enantiomeric purity should be established by a second validated enantioselective analytical procedure. Reference standards should be available for the enantiomer and the antipode, and both should be of acceptable enantiomeric purity. A limit should be specified for the antipode and this limit should be qualified through the levels found in batches used in preclinical and clinical studies. The stability of the drug substance towards racemization under stress conditions (e.g., acid, base, etc.) and on long-term storage should be investigated.

Diastereoisomers are potential impurities in the drug substance and the drug product, based on a single enantiomer containing two or more chiral centres. Since these impurities are chemically distinct from the enantiomer, they should be investigated and limited as drug substance/drug product impurities in accordance with the Programme's guideline *Chemistry and Manufacturing: New Drugs*.

2.1.2 Drug Product

The enantiomeric purity of the drug substance in the drug product should be investigated using a validated enantioselective method prior to and during the stability studies conducted to determine the shelf life. Results from the primary stability studies may be considered sufficient. However, a test for enantiomeric purity should be incorporated into the drug product specification if results of these investigations warrant.

2.2 RACEMATE

The physicochemical properties, including the nature of the racemate, i.e., whether a racemic compound (true racemate) or racemic mixture (conglomerate), should be investigated. During the drug substance development, a test for the identity of the racemate should be performed to ensure that spontaneous resolution has not occurred during the purification of the bulk drug. Also, when technically feasible, basic physicochemical information on the individual enantiomers should be provided. Typically, this may include information on melting point, optical rotation, crystal form, and stability towards racemization.

2.3 NON-RACEMIC MIXTURE

2.3.1 Drug Substance

Two different types of mixtures are considered.

a) an enantiomerically enriched drug substance resulting from incomplete resolution or partial enantioselective synthesis;

b) a mixture of enantiomers in specified proportions.

Enantioselective tests for identity and composition are required in the specification. Optical rotation may be suitable for identity but a more specific and sensitive method is required for the quantitative determination of the enantiomers. Limits should be set for each component based on the composition of the batches used in preclinical and clinical studies. Reference standards of acceptable purity should be prepared for each component. The stability of the mixture under stress conditions (e.g., acid, base, etc.) and on long-term storage under ambient conditions should be investigated.

2.3.2 Drug Product

A test with limits on the composition of the mixture is required in the drug product specification and the relative proportions of the components should be monitored in the long-term stability studies.

3. PRECLINICAL AND CLINICAL

3.1 SINGLE ENANTIOMER

In addition to the same documentation as required for any new active substance, information on the following should be provided:

- The *in vivo* stability of the enantiomer must be established. If the antipode is formed *in vivo*, it should be considered to be a metabolite, and addressed as such during the drug development process. The metabolism and disposition of the enantiomer should be followed using enantioselective methods in each species used in preclinical studies, and in the Phase I studies in humans. If it is established that racemization or inversion does not occur, enantioselective methods may not be needed in all subsequent studies.

3.2 RACEMATE

In addition to the same documentation as required for any new active substance, information on the following should be provided:

a) Primary and secondary pharmacodynamic effects of each enantiomer in animals and *in vitro* systems with regard to potency, specificity, maximum effect, etc., where appropriate.

b) Pharmacokinetic studies in animals and in humans using enantioselective assays to validate the results of acute and multiple-dose toxicity studies performed with the racemate. Pharmacokinetic studies should be performed in animals with the same doses/routes/species as the toxicological studies and in humans at the proposed therapeutic doses, at steady state where applicable. Validated enantioselective analytical techniques should also be used in subsequent studies.

3.3 SWITCH FROM A RACEMATE TO AN ENANTIOMER

The documentation required for a single enantiomer of a marketed racemate is the same as that for any new active substance. However, many of the studies already performed with the racemate may not have to be repeated with the selected enantiomer, if adequate "bridging studies" have been performed. The purpose of the bridging studies is to validate the relevancy of the studies performed with the racemate, and therefore, the nature of the studies must be determined on a case-by-case basis. Examples of bridging studies are:

a) Comparison of the acute toxicity, the pharmacodynamic activity and the pharmacokinetics of the selected enantiomer and racemate.

b) Preclinical toxicology bridging studies could consist of repeat dose studies, the duration of which would depend upon the proposed duration of use in humans. In general, these studies should not be shorter than three months. Some exceptions exist (e.g., single dose drugs such as neuromuscular relaxants). In addition, the reproductive toxicology segment II study should be repeated in the most sensitive and relevant species, using the single enantiomer.

4. BIOEQUIVALENCE REQUIREMENTS FOR SOLID ORAL DOSAGE FORMS

The requirements outlined below apply to subsequent-market entries of solid oral dosage forms and variations of existing dosage forms that are to be evaluated based on comparative bioavailability studies.

Comparative bioavailability of products containing a single enantiomer or a mixture of enantiomers should be assessed against an acceptable Canadian Reference Product as defined in section C.08.002.1 of the *Food and Drug Regulations* (see the relevant Programme's guidance documents and policies, e.g., *Conduct and Analysis of Bioavailability and Bioequivalence Studies - Parts A and B* and *Canadian Reference Product*). Comparisons should be made between "pharmaceutically equivalent" products that meet standards for enantiomeric purity/composition acceptable to the Programme.

In general, when comparing solid oral dosage forms of similar type, e.g., two immediate-release formulations, that contain the same isomeric ratio of medicinal ingredient(s), the parameters to be determined and the standards to be met will be the same as those specified in the above mentioned guidance documents and will be based on the measurement of total drug concentrations.

When conducting comparative bioavailability studies to compare solid oral dosage forms of differing type, e.g., the comparison of a Group II modified-release drug product as defined in *Conduct and Analysis of Bioavailability and Bioequivalence Studies - Part B: Oral Modified Release Formulations* (Guideline B) to an immediate-release or a different kind of modified-release formulation, stereoselective analysis may be required. If the rate of release and/or absorption of the medicinal ingredient into the systemic circulation affects the *in vivo* enantiomeric ratio (e.g., drugs with enantioselective non-linear first pass metabolism), the comparative bioavailability requirements outlined in the appropriate Programme guidance document must be met on each enantiomer.

APPENDIX 1 - GLOSSARY

ABSOLUTE CONFIGURATION (or Absolute Stereochemistry) - The specific three-dimensional arrangement of substituents around a chiral element.

ANTIPODE - The mirror image isomer of a chiral molecule. A pair of enantiomers is known as optical antipodes.

CAHN-INGOLD-PRELOG CONVENTION - The assignment of configuration about a chiral atom as "R" or "S" by designation of the sequence according to a set of rules.

CHIRALITY - The property of non-superimposability of a molecule on its mirror image. A molecule that contains just one carbon atom connected to four different groups (called the chiral carbon) is chiral. Chirality in a molecule could be also induced, for example, by the presence of other quadrivalent chiral atoms, restricted rotation about single bonds, or by helicity.

CHIRAL INVERSION - Conversion of one enantiomer into its mirror image.

CONFIGURATIONS - If two different three-dimensional arrangements in space of the atoms in a molecule are not interconvertible by free rotation about bonds, they are called configurations.

CONFORMERS - If two different three-dimensional arrangements in space of the atoms in a molecule are interconvertible merely by free rotation about bonds, they are called conformers.

DIASTEREOISOMERS - IUPAC defines diastereoisomers as “Stereoisomers that are not enantiomeric”. Diastereoisomers may be chiral or achiral. However, the term diastereoisomers is used currently by many scientific publications to denote exclusively diastereoisomers that are chiral.

DISTOMER - Refers to the enantiomer with lower pharmacological affinity or activity. Note: A "distomer" for one particular activity can be the "eutomer" for another activity

ENANTIOMERS - Stereoisomers whose mirror images cannot be superimposed.

ENANTIOMERIC PURITY/ENANTIOMERIC EXCESS - The percentage of the enantiomer in excess of its antipode. For example, a drug substance containing 99% of an enantiomer and 1% of the antipode has an enantiomeric purity of 98%.

ENANTIOSELECTIVITY - Characteristic of a process whereby one enantiomer is favoured exclusively or predominantly over the other. In pharmacological terms, the extent to which a biological structure, i.e., an enzyme or any other macromolecular structure (e.g., antibody or receptor) exhibits affinity towards one enantiomer over the other.

ENANTIOSELECTIVE ASSAY - Analytical method capable of separating and quantifying enantiomers.

ENANTIOSELECTIVE SYNTHESIS - Any reaction in which one enantiomer is formed predominantly or exclusively.

EPIMERS - Two diastereoisomers that have a different configuration at only one chiral centre.

EPIMERIZATION - The change of configuration at a chiral centre in a molecule containing two or more chiral centres.

EUDISMIC RATIO - The pharmacological potency ratio of two enantiomers is called the eudismic ratio, and its logarithm, the eudismic index (EI).

EUTOMER - Refers to the enantiomer with higher pharmacological affinity or activity.

ISOMERS - Compounds that have identical molecular formulae but differ in the nature or sequence of bonding of their atoms or in the arrangement of their atoms in space.

***cis-trans* ISOMERS (Geometric Isomers) -** Stereoisomers that differ only in the arrangement of atoms relative to a specified plane in cases where these atoms are, or are considered as if they were parts of a rigid structure, e.g., a ring or a double bond.

MESO-COMPOUND - A compound containing two or more chiral centres, whose mirror images are superimposable because the molecule as a whole is symmetric.

OPTICAL ROTATION - The change of direction of the plane of polarized light to either the right or to the left as it passes through a molecule.

RACEMATE - A mixture of equimolar amounts of enantiomers.

RACEMIC COMPOUND - A homogeneous solid phase composed of equimolar amounts of enantiomeric molecules.

RACEMIC MIXTURE - A mixture of equimolar amounts of enantiomeric molecules present as separate solid phases.

RACEMIZATION - Conversion of an enantiomer to its racemate.

STEREOISOMERS - Isomers that differ in the arrangement of atoms in space.

STEREOSELECTIVITY - Characteristic of a process whereby one of a set of stereoisomers is favoured predominantly or exclusively over the others.

STEREOSPECIFICITY - Characteristic of a process whereby stereoisomers of a molecule induce stereoisomerically different effects. All stereospecific processes are stereoselective but the converse is not true. In a stereoselective synthesis, one of a set of isomers is predominantly or exclusively formed whereas in a stereospecific synthesis, one isomer leads to one product while another isomer leads to the opposite product.

APPENDIX 2 - ANALYTICAL METHODS FOR CHIRAL DRUGS

Physico-chemical methods that can be used to provide information about chiral drugs are listed below. This is by no means an exhaustive list, and any validated method that is demonstrated to be useful will be considered.

CHIRAL HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC) - Chiral HPLC may be used to separate mixtures of enantiomers directly without forming diastereoisomeric derivatives. Separations can be effected through the use of chiral stationary phases, or chiral mobile phase additives in conjunction with regular (achiral) columns.

CHIRAL GAS CHROMATOGRAPHY - Stationary phases modified with chiral agents are available for the separation of enantiomers.

MELTING POINT - The melting points may be used in distinguishing individual enantiomers from the racemate.

NUCLEAR MAGNETIC RESONANCE (NMR) - NMR is a useful tool for the determination of enantiomeric purity or enantiomeric composition. This is accomplished by making the NMR signals for the protons of the enantiomers non-equivalent by the use of chiral lanthanide shift reagents, chiral solvating agents or chiral derivatizing agents.

OPTICAL ROTATION - This method can be used to distinguish between enantiomers because they rotate the plane of polarized light in opposite directions but in equal amounts.

OPTICAL ROTATORY DISPERSION (ORD) AND CIRCULAR DICHROISM (CD) - ORD measures the change of specific rotation of an optically active compound with the wavelength of the light used. CD measures the differential absorption of left and right circularly polarized light by an optically active compound. These chiroptical methods can be used to identify and/ or quantitate enantiomers.

X-RAY CRYSTALLOGRAPHY - X-Ray crystallography in the solid state could be used to determine the absolute configuration of molecules and to distinguish conglomerates from racemic compounds.