



(72) AVERBACK, PAUL, CA

(72) GHANBARI, HOSSEIN, US

(72) BEHESHTI, IRAJ, US

(72) MORSE, DAVID, CA

(71) NYMOX CORPORATION, CA

(51) Int.Cl.⁷ C07C 43/164, C07D 211/90, A61K 31/52, A61K 31/00,
A61K 31/495, A61K 31/44, C07C 43/174, C07C 217/10, A61K 31/08,
C07D 295/08, C07D 473/04, C07D 295/02

(30) 1997/05/19 (08/858,343) US

(54) **AGENTS PHARMACEUTIQUES DESTINES AU TRAITEMENT
DE L'AMYLOIDOSE CEREBRALE**

(54) **PHARMACEUTICAL AGENTS FOR THE TREATMENT OF
CEREBRAL AMYLOIDOSIS**

(57) Cette invention concerne un procédé permettant de traiter ou de prévenir la formation d'amyloïde cérébrale associée à la maladie d'Alzheimer. Ce procédé fait appel à des médicaments qui permettent de prévenir efficacement et in vivo la rupture de microsphères denses (MSD), ou d'altérer cette rupture de manière à obtenir les effets suivants: (i) une réduction du volume des tissus occupés par la matière protéinique MSD transformée et de nouveau répartie, et par la zone de blessure qu'elle entraîne; (ii) une réduction de la persistance ou de la durée de la blessure et de l'inflammation associées aux MSD brisées et à la matière protéinique MSD transformée et de nouveau répartie; et/ou (iii) un accroissement de la digestion et de l'élimination de la matière MSD par le système réticulo-endothélial.

(57) Formation of cerebral amyloid associated with Alzheimer's disease can be treated or averted using medicaments that are effective in vivo in preventing disruption of dense microspheres (DMS) or in altering the disruption so as to (i) reduce the volume of tissue occupied by redistributed, transformed DMS protein material and its associated injury focus; (ii) reduce the persistence or duration of injury and inflammation associated with the disrupted DMS and redistributed transformed DMS protein material; and/or (iii) increase the digestion and removal of DMS material through the reticuloendothelial system.

PCTWORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶ : C07C 43/164, A61K 31/08, C07C 43/174, 217/10, C07D 211/90, 295/02, 295/08, 473/04, A61K 31/495, 31/645, 31/44, 31/52	A1	(11) International Publication Number: WO 98/52898 (43) International Publication Date: 26 November 1998 (26.11.98)
(21) International Application Number: PCT/US98/10218 (22) International Filing Date: 19 May 1998 (19.05.98) (30) Priority Data: 08/858,343 19 May 1997 (19.05.97) US (63) Related by Continuation (CON) or Continuation-in-Part (CIP) to Earlier Application US 08/858,343 (CON) Filed on 19 May 1998 (19.05.98) (71) Applicant (for all designated States except US): NYMOX CORPORATION [CA/CA]; 175 Boulevard Bouchard, Dorval, Quebec H9S 1B1 (CA). (72) Inventors; and (75) Inventors/Applicants (for US only): AVERBACK, Paul [CA/CA]; 383 Lakeshore Road, Beaconsfield, Quebec H9W 4J1 (CA). GHANBARI, Hossein [US/US]; 15537 Ambiance Drive, Gaithersburg, MD 20878 (US). BEHESHTI, Iraj [US/US]; 1146 Regal Oak Drive, Rockville, MD 20852		(US). MORSE, David [CA/CA]; 6170 Chemille, Anjou, Quebec H1M 1T2 (CA). (74) Agents: BENT, Stephen, A. et al.; Foley & Lardner, Suite 500, 3000 K Street, N.W., Washington, DC 20007-5109 (US). (81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i> <i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>
(54) Title: PHARMACEUTICAL AGENTS FOR THE TREATMENT OF CEREBRAL AMYLOIDOSIS (57) Abstract Formation of cerebral amyloid associated with Alzheimer's disease can be treated or averted using medicaments that are effective <i>in vivo</i> in preventing disruption of dense microspheres (DMS) or in altering the disruption so as to (i) reduce the volume of tissue occupied by redistributed, transformed DMS protein material and its associated injury focus; (ii) reduce the persistence or duration of injury and inflammation associated with the disrupted DMS and redistributed transformed DMS protein material; and/or (iii) increase the digestion and removal of DMS material through the reticuloendothelial system.		

PHARMACEUTICAL AGENTS FOR THE TREATMENT OF CEREBRAL AMYLOIDOSIS

BACKGROUND OF THE INVENTION**1. Field of the Invention**

The present invention relates to compounds that act, at physiologically-compatible levels, to impede the formation of amyloid in brain, including Alzheimer senile amyloid plaques *in vivo*. More specifically, the present invention relates to compounds that impede the transformation of dense microspheres (DMS) into amyloid. The invention also relates to methodology for the screening of compounds that possess this activity, and to treating cerebral amyloidosis by administering such compounds therapeutically.

2. Description of Related Art

There is no effective therapy for cerebral amyloidosis, which almost invariably has a fatal outcome following the onset of amyloid deposits. For example, Alzheimer's disease is estimated to be the fourth or fifth leading cause of death in North Americans.

A universally accepted indicator of cerebral amyloidosis is the accumulation of lesions, so-called "senile plaques," that are comprised in large part of amyloid fibrils. Senile plaques are spherical, ranging from 10 to 200 μm in diameter, and are found occasionally in aged cerebral cortex (see below) but in larger numbers in Alzheimer-affected cortex.

Most specialists agree that reproduction of amyloid fibrils experimentally from precursor materials which are extracted, activated, or otherwise derived from human brain constitutes the best available evidence linking an agent or precursor to the progression of cerebral amyloidosis. Indeed, the literature has documented the use of such materials from human brain, normal and Alzheimer-affected, that are not already amyloid, and their transformation into amyloid.

Accordingly, a reliable indicator is available for compounds that might be effective in treating cerebral amyloidosis. In particular, it is possible to determine whether a given compound prevents the structural transition of a brain-localized precursor to a β -pleated sheet conformation and thus prevent conversion to cerebral amyloid (*i.e.*, that displays "anti-amyloid activity") at physiologically acceptable levels of the active agent. See U.S. patent No. 5,231,170, the contents of which are incorporated by reference.

In a majority of cases, subjects with cerebral amyloidosis, including Alzheimer's patients, display symptoms on a quantitative basis. Blessed *et al.*, *Brit J. Psychiatry* 114: 797-811 (1968); Tomlinson *et al.*, *J. Neurol. Sci.* 7: 331-56 (1968); Tomlinson, B.E., *et al.*, *J. Neurol. Sci.* 11: 205-42 (1970); Corsellis, MENTAL ILLNESS AND THE AGEING BRAIN, Oxford University Press, London (1962); Corsellis, "Ageing and the Dementias," in GREENFIELD'S NEUROPATHOLOGY 796-848, Edward Arnold, London (1976). Elderly subjects who have a small number of senile plaques are asymptomatic and are categorized by some experts as preclinical, by others as presymptomatic, and by still other authorities as normal variants. In any event, the elderly brain apparently can have a low number of senile plaques and still be categorized as "normal." When the amyloid plaque number is high, however, symptoms of dementia appear. Thus, treatment regimens that prevent the formation of amyloid plaques or that reduce the number or rate of formation of amyloid plaques are useful.

A microscopic structure, the so-called "dense microsphere" (DMS), is found in normal brain and in brain affected by Alzheimer's disease. See Averbach, *Acta Neuropathol.* 61: 148-52 (1983); results confirmed by Hara, *J. Neuropath. Exp. Neurol.* (1986). Evidence for the existence of dense microspheres comes from microscopic histological section studies of fixed whole brain tissue, where the dense microspheres are seen to have a proteinaceous central region ("DMS protein") surrounded by continuous membrane ("DMS membrane").

The extraction, purification, and characterization of isolated samples of DMS and the use of DMS material have been documented. See, for example,

U.S. patents No. 4,919,915 and No. 4,816,416, the respective contents of which are incorporated by reference.

5 DMS disruption is believed to commence after individual DMS reach a threshold size in the elderly or Alzheimer group. Even DMS that have reached the threshold size are quite small, with diameters on the order of approximately 10 microns or less. When the DMS disrupt, constituent protein matter transforms and redistributes to occupy a tissue volume, (anywhere between 10 to 1,000 times larger than the precedent DMS), which comprises a much larger injury focus.

SUMMARY OF THE INVENTION

10 Preventing the disruption of DMS would prevent the formation of amyloid plaques. If DMS disruption can be curtailed, for example, by around 25% or more, cerebral amyloid formation, and its rate of formation, induced by DMS disruption also can be reduced or prevented. Thus, reducing the size of the injury foci associated with disrupted DMS would impede the formation of amyloid
15 plaques, by reducing the number of DMS disruptions that may be caused by an initial disruption. In addition, reducing the persistence of injury and inflammation associated with DMS disruption, including, for example, increasing the digestion and removal of DMS material through the reticuloendothelial system, would prevent the formation of amyloid plaques or, at least, reduce the number and/or
20 rate of formation of other, secondary DMS disruptions.

Accordingly, a need exists for an approach to preventing DMS disruption. A need also exists for a method to reduce the size of the tissue volume associated with disrupted DMS (injury foci). Furthermore, there is a need for a technique to reduce the persistence of inflammatory reaction brought about by DMS
25 disruption, as well as for methods to increase the digestion and removal of DMS material through the reticuloendothelial system.

It therefore is an object of the present invention to provide a composition and a method that are useful in impeding DMS disruption and, hence, in treating cerebral amyloidosis, a condition characterized by the presence of abnormal
30 amounts of amyloid β -protein associated plaques (senile plaques) and other

amyloid deposits. It is an additional object to provide a screening approach for identifying compounds that are useful in impeding DMS disruption.

It is also an object of the present invention to provide a treatment for cerebral amyloidosis by the administration of a compound selected from a class of pharmaceutically active agents that have in common an ability to impede the disruption of intact DMS.

In accomplishing the foregoing objects, a method has been provided, in accordance with one aspect of the present invention, for treating cerebral amyloidosis, comprising the step of administering to a subject in need thereof, a pharmaceutically effective amount of a compound that impedes disruption of intact DMS by: (i) reducing the mean tissue volume of disrupted DMS; (ii) reducing the ratio of the number of inflammatory cells per DMS; or (iii) increasing the ratio of the number of macrophages containing disrupted DMS material per DMS, each when compared to controls. The compound impedes the disruption of intact DMS when administered, at an in-tissue concentration of about 10^{-5} M or less, to an experimental animal or to a test preparation of human postmortem brain that has received an intracerebral injection of DMS. In one preferred embodiment, the compound thus administered impedes disruption of intact DMS by reducing the mean tissue volume of disrupted DMS material. The compound acts on DMS components in such a way that disrupted DMS *in situ* attain a significantly reduced diameter, volume of tissue occupied by the redistributed transformed DMS protein material, and associated injury foci, when compared to the diameter, volume of tissue occupied, and injury foci of disrupted DMS in an untreated subject or in a subject treated with an inactive agent.

In accordance with another aspect of the present invention, there is provided a composition for treating cerebral amyloidosis, comprising a pharmaceutically effective amount of a compound that impedes disruption of intact DMS by: (i) reducing the mean tissue volume of disrupted DMS; (ii) reducing the ratio of the number of inflammatory cells per DMS; or (iii) increasing the ratio of the number of macrophages containing disrupted DMS material per DMS, each when compared to controls. The compound impedes the disruption of intact DMS when administered, at an in-tissue concentration of about 10^{-5} M or less, to an experimental animal or to a test preparation of human postmortem brain that has

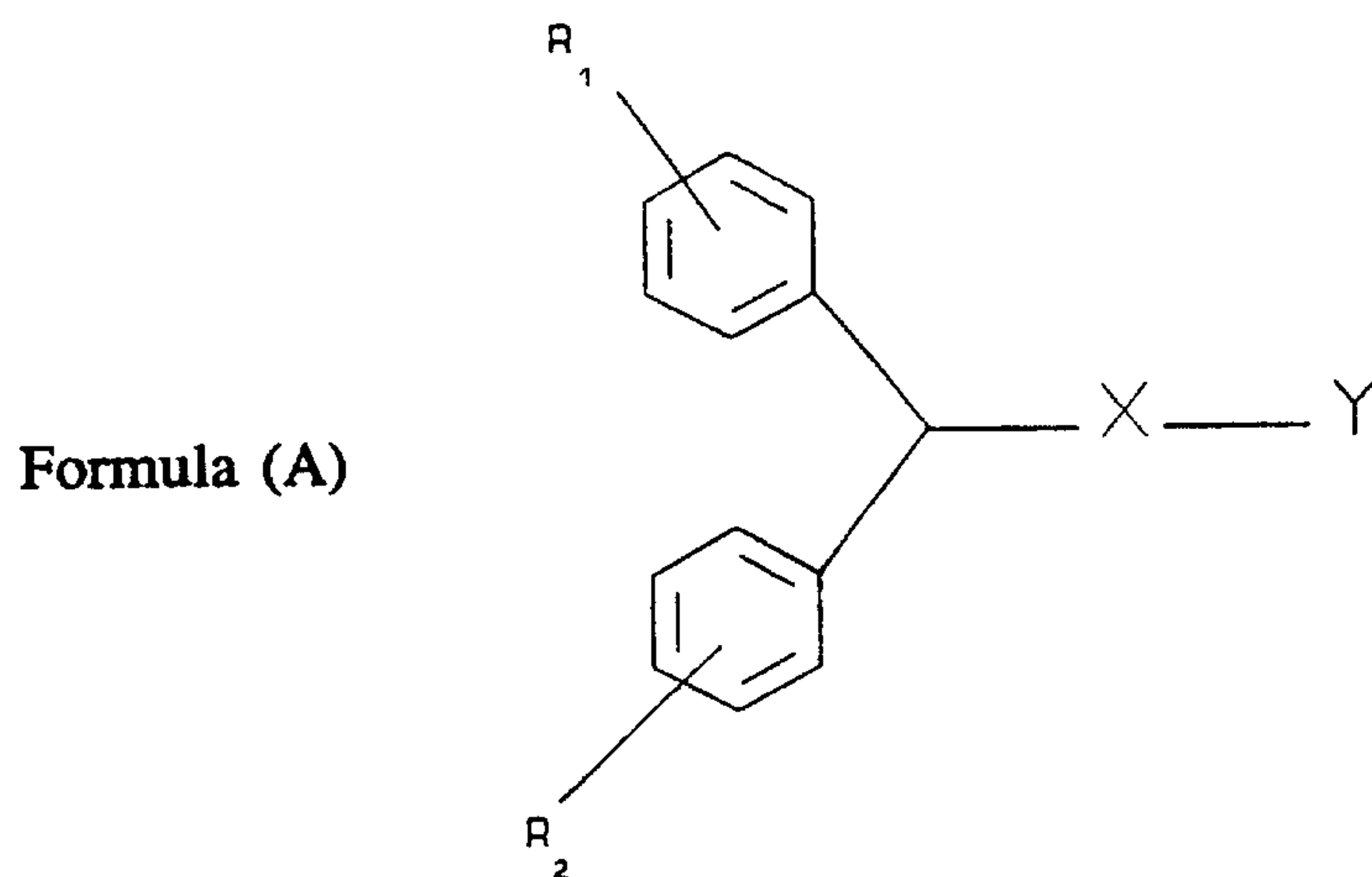
received an intracerebral injection of DMS. The compositions and methods of using the compositions typically include at least one of the aforementioned compounds in combination with a pharmaceutically acceptable sterile vehicle, as described in REMINGTON'S PHARMACEUTICAL SCIENCES: DRUG
5 RECEPTORS AND RECEPTOR THEORY, (18th ed.), Mack Publishing Co., Easton PA (1990).

Compounds also are provided that are effective in reducing the number of subsequent DMS disruptions brought about by an initial DMS disruption by reducing the ratio of the number of inflammatory cells per DMS, *i.e.*, reducing
10 the extent of the injury foci associated with the transformation and redistribution of protein material of an initial disrupted DMS. These compounds, when administered to a test animal that has received an injection of DMS at an in-tissue concentration of about 10^{-5} M or less, result in less evidence of inflammatory reaction in relation to persistent DMS materials at the DMS injection site
15 measured, for example, by the number of inflammatory cells such as polymorphonuclear leukocytes and mononuclear leukocytes per DMS in each injury focus, or the tissue volume taken up by non-macrophage inflammatory cells per DMS in each injury focus, when compared to controls at the same time intervals, for instance, at 24 hours and 48 hours, respectively.

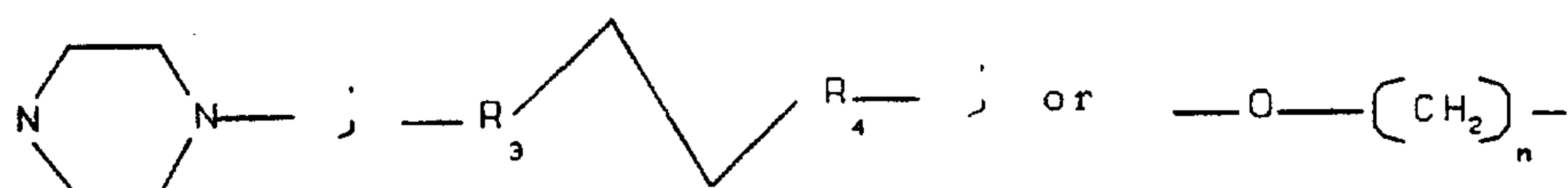
20 In accordance with yet another aspect of the present invention, compounds are provided that are effective in reducing the number of subsequent DMS disruptions brought about by an initial DMS disruption by increasing the ratio of the number of macrophages containing disrupted DMS material per DMS, *i.e.*, promoting the digestion and removal of the DMS material through the
25 reticuloendothelial system. By promoting the digestion and removal of the DMS material, the compounds reduce the extent and duration of the effect of the disrupted DMS and therefore impede subsequent DMS disruptions. These compounds, when administered to a test animal that has received an injection of DMS at an in-tissue concentration of about 10^{-5} M or less, result in more evidence
30 of digestion and removal by the reticuloendothelial system, when compared to controls. Digestion and removal can be measured, for example, by the presence of intracellular intact DMS protein material within the cytoplasm of macrophages and mononuclear phagocytes and the numbers thereof at or near the site of DMS

disruption, or the presence of intracellular digested, altered, proteolyzed or otherwise transformed DMS protein material within the cytoplasm of macrophages and mononuclear phagocytes and the numbers thereof at or near the site of DMS disruption.

- 5 Illustrative of the compounds within the present invention include those of general formula (A):



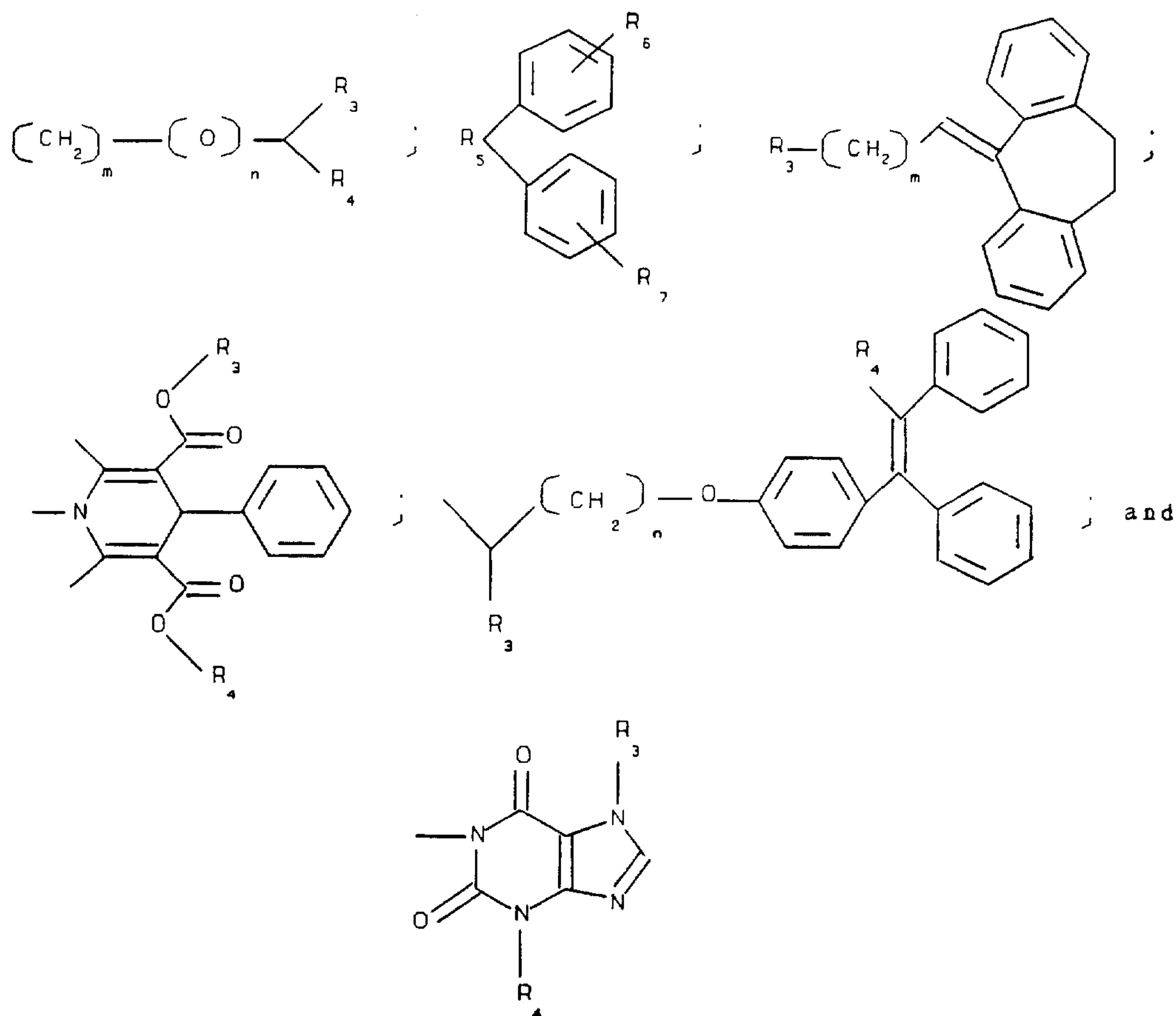
where X is selected from



and Y is selected from

- 10 In the general formula (A) above:

R_1 and R_2 are each one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 alkoxy, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein if R_1 or R_2 are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety
 15 selected from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or unsubstituted C1-C5 alkylamino; wherein if R_1 or R_2 are alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted



or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl, and if R_1 or R_2 is a fused ring system, each individual R_1 or R_2 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; wherein each ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_3 is hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, amino, C1-C5 alkyl-substituted amino, sulfur, oxygen, phenyl, benzyl, naphthyl and anthracenyl; wherein each aromatic ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_4 is hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, amino, C1-C5 alkyl-substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, oxygen, phenyl, benzyl, naphthyl and anthracenyl;
5 wherein each aromatic ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_5 is nitrogen or carbon;

R_6 and R_7 are each one or more independent substitutions selected from
10 hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein if R_6 or R_7 is piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or
15 unsubstituted C1-C5 alkylamino; wherein if R_6 or R_7 is alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl, and if R_6 or R_7 is a fused ring system, each individual R_6 and R_7 involved in the fused ring system, together with the phenyl to which it is attached,
20 forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a
25 nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; wherein each ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

n is an integer of from 0 to 5;

m is an integer of from 0 to 5;

30 or a pharmaceutically acceptable salt of such compound.

Other objects, features and advantages of the present invention will become apparent from the following detailed description. It should be understood, however, that the detailed description and the specific examples,

while indicating preferred embodiments of the invention, are given by way of illustration only, since various changes and modifications within the spirit and scope of the invention will become apparent to those skilled in the art from this detailed description. Unless otherwise specified, the respective contents of documents cited in the following description are hereby incorporated by reference.

BRIEF DESCRIPTION OF THE DRAWINGS

Figures 1-12 illustrate schematically DMS disruption and stimulation of subsequent DMS disruption of other DMS on nerve fibers which are associated with the region of the disrupted DMS. Figures 13-26 illustrate how disruption of one DMS can stimulate and bring about the disruption of numerous DMS.

FIGURE 1 illustrates five nerve fibers (lines and triangles) with five individual DMS (black circles), one on each fiber.

FIGURE 2 illustrates the initiation of a DMS disruption on one of the five nerve fibers.

FIGURE 3 illustrates continued disruption of the disrupted DMS of **FIGURE 2**, and the inducement of disruption of a DMS on an adjacent fiber.

FIGURE 4 illustrates a fully disrupted DMS from **FIGURE 2**, and the initiation of disruption of the DMS on the adjacent fiber of **FIGURE 3**.

FIGURE 5 illustrates continued disruption of the disrupted DMS of **FIGURE 4**, and the inducement of disruption of a DMS on an adjacent fiber.

FIGURE 6 illustrates a fully disrupted DMS from **FIGURE 4**, and the initiation of disruption of the DMS on the adjacent fiber of **FIGURE 5**.

FIGURE 7 illustrates continued disruption of the disrupted DMS of **FIGURE 6**, and the inducement of disruption of a DMS on an adjacent fiber.

FIGURE 8 illustrates a fully disrupted DMS from **FIGURE 6**, and the initiation of disruption of the DMS on the adjacent fiber of **FIGURE 7**.

FIGURE 9 illustrates continued disruption of the disrupted DMS of **FIGURE 7**, and the inducement of disruption of a DMS on an adjacent fiber.

FIGURE 10 illustrates a fully disrupted DMS from **FIGURE 8**, and the initiation of disruption of the DMS on the adjacent fiber of **FIGURE 9**.

FIGURE 11 illustrates continued disruption of the disrupted DMS of **FIGURE 10**.

FIGURE 12 illustrates fully disrupted DMS on all 5 fibers.

FIGURE 13 illustrates nerve fibers with one undisrupted DMS in the center, wherein some of the nerve fibers contain undisrupted DMS.

FIGURE 14 illustrates the same nerve fibers with the DMS in the center beginning to disrupt.

FIGURE 15 illustrates the same nerve fibers with the DMS in the center continuing to disrupt and attaining a disrupted volume where it is now in the region of an adjacent nerve fiber which contains one undisrupted DMS.

FIGURE 16 illustrates the same nerve fibers whereby the undisrupted DMS on the adjacent nerve fiber has begun to disrupt.

FIGURE 17 illustrates a continuation of the disruption of the DMS on the adjacent nerve fiber of **FIGURE 16**.

FIGURE 18 illustrates a continuation of the disruption of the DMS on the adjacent nerve fiber of **FIGURE 17**.

FIGURE 19 illustrates the same initial nerve fibers whereby the DMS in the center has disrupted to a greater extent than in **FIGURES 14** and **15**, and has now impinged on two additional adjacent nerve fibers, each containing one undisrupted DMS.

FIGURE 20 illustrates the undisrupted DMS on the adjacent nerve fibers of **FIGURE 19** beginning to disrupt.

FIGURE 21 illustrates a continuation of the disruption of the DMS of **FIGURE 20**.

FIGURE 22 illustrates a continuation of the disruption of the DMS of **FIGURE 21**.

FIGURE 23 illustrates the center DMS disrupting to an even greater extent than in **FIGURES 14, 15** and **19**, and impinging on 8 adjacent DMS-containing nerve fibers.

FIGURE 24 illustrates many undisrupted DMS on the adjacent nerve fibers of **FIGURE 23** beginning to disrupt.

FIGURE 25 illustrates a continuation of the disruption of the many DMS of **FIGURE 24**.

FIGURE 26 illustrates a continuation of the disruption of the many DMS of FIGURE 25.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

5 It has been discovered that the development of amyloid fibrils associated, for example, with the evolution of cerebral amyloidosis is tied to the unchecked disruption of DMS *in vivo*. The connection between DMS disruption and amyloid formation is evidenced by the fact that disrupted DMS treated with Congo Red stain display a red-green congophilic birefringence identical to that found in senile-plaque amyloid. Thus, the most significant aspect of brain damage that
10 characterizes cerebral amyloidosis can be reproduced using material derived, pursuant to the present invention, from normal mammalian brain samples.

A hitherto unrecognized aspect of cerebral amyloid plaque formation by DMS transformation and/or disruption concerns the initiation of the underlying DMS disruption. An important mechanism of initiation and promotion of DMS
15 disruption has been discovered that involves a distinctive autocatalytic phenomenon, whereby the disruption, degeneration, and evolution of an individual DMS into an individual cerebral amyloid plaque provides the stimulus for a group or field of other DMS to disrupt, degenerate and evolve in a recurring set of waves (see Figures 1-12). This unchecked, autocatalytic phenomenon causes an
20 exponential growth pattern: small, perhaps statistically insignificant differences (between individual brains) in starting numbers of disrupted DMS *in situ* evolve into statistically significant differences after generations of the cycle. For example, if all other factors were equal, a subject having an initial group of 100 DMS would not be statistically or symptomatically different from a second subject
25 having an initial group of 98 DMS. However, if over time each of the initial DMS initiated 10 subsequent DMS to disrupt, each of which in turn initiated 10 subsequent DMS disruptions, then group 1 after 20 generations would have 2×10^{20} more disrupted DMS than group 2, which is significant.

The compounds of the present invention impede DMS disruption by
30 previously unknown mechanisms that reduce the number of subsequent DMS

disruptions brought about by an initial DMS disruption and hence, reduce the cerebral amyloid burden and formation of amyloid plaques. The compounds of the present invention therefore impede disruption of DMS by: (i) reducing the mean volume of tissue occupied by disrupted DMS (*i.e.*, redistributed transformed
5 DMS protein material and the associated injury foci); (ii) reducing the ratio of the number of inflammatory cells per DMS (*i.e.*, persistence of injury and inflammation associated with the disrupted DMS); or (iii) increasing the ratio of the number of macrophages containing disrupted DMS material per DMS (*i.e.*, digestion and removal of DMS material through the reticuloendothelial system).
10 Additionally, a compound of the present invention may prevent the initial DMS disruption and, hence, prevent any subsequent DMS disruptions.

The usefulness of precluding (inhibiting) cerebral amyloid formation is recognized, and the usefulness of blocking the transformation of cerebral amyloid precursors into amyloid is evident (known as "blocker therapy"). In accordance
15 with blocker therapy, DMS transformation into amyloid plaques is interrupted or inhibited after DMS disruption has occurred. By contrast, the present invention comprehends a therapy, leading to less DMS-derived cerebral amyloid by impeding DMS disruption and/or altering the disruption process by acting on the DMS prior to disruption in such a manner that either prevents disruption, or,
20 when disrupted, reduces subsequent DMS disruptions. Compounds that are effective in blocking DMS transformation in accordance with blocker therapy do not necessarily impede DMS disruption or alter the disruption process by acting on the DMS prior to disruption in the manners described herein, and vice versa.

In accordance with the present invention, if the mean volume of tissue
25 occupied by the disrupted DMS is reduced when compared to the volume of tissue occupied by disrupted DMS in a control subject, the volume of the disrupted DMS preferably is reduced by greater than 10%, more preferably greater than 20%, and most preferably, greater than 30%. (The expression "control subject" in this context denotes either an untreated subject or a subject that has been
30 treated with an inactive placebo.) If the ratio of number of acute inflammatory cells per DMS is reduced when compared to the ratio in a control subject, the ratio is reduced by greater than 10%, preferably 50% and more preferably 100%. In addition, if the ratio of the number of macrophage containing disrupted DMS

material per DMS is increased when compared to the ratio in a control subject, the ratio is increased by greater than 10%, preferably greater than 50% and more preferably greater than 100%.

5 Figures 1-12 illustrate how the disruption of one DMS can stimulate subsequent disruption of other DMS on nerve fibers which contact fully disrupted DMS. This phenomenon is addressed by the expressions "subsequent DMS disruptions brought about by an initial DMS disruption," "DMS disruptions brought about by an initial DMS disruption," and "the autocatalytic phenomenon," respectively. See also Figures 13-26, which are discussed in greater detail below.

10 Figure 1 illustrates five nerve fibers (lines and triangles) with five individual DMS (black circles), one on each fiber, whereby nerve fiber 1 contains undisrupted and intact DMS 11, fiber 2 contains DMS 12, fiber 3 contains DMS 13, fiber 4 contains DMS 14 and fiber 5 contains DMS15. Figure 2 shows the initiation of disruption of DMS 11 on fiber 1.

15 As shown in Figure 3, DMS 11 of Figure 2 which had begun to disrupt, continues disrupting and reaches a size which induces disruption of DMS 12 on adjacent fiber 2. If the DMS could be prevented from ever disrupting, or the disruption mechanism altered to an extent which would: (i) prevent the disrupted DMS from attaining a size sufficient to induce subsequent DMS disruptions; (ii)
20 reduce the persistence of injury and inflammation associated with the disrupted DMS which would prevent subsequent DMS disruptions; or (iii) increase the digestion and removal of DMS material through the reticuloendothelial system which also prevents subsequent DMS disruptions, then the autocatalytic phenomenon that is illustrated in Figures 1-12 could be prevented.

25 Figure 4 illustrates the DMS from FIG. 2, now fully disrupted to form disrupted DMS 21, and the initiation of disruption of DMS 12 on adjacent fiber 2 of FIG. 3. As shown in Figure 5, DMS 12 of FIG. 4, which began disruption, continues to disrupt, and begins to induce disruption of DMS 13 on adjacent fiber 3. This process is repeated for the remaining fibers as shown in Figures 6-12.
30 Figure 6 shows disrupted DMS 22, Figure 8 shows disrupted DMS 23 on fiber 3, Figure 10 shows disrupted DMS 24 on fiber 4, and Figure 12 shows disrupted DMS 25 on fiber 5. As mentioned above, disrupted DMS 21, 22, 23, 24 and 25 result in the formation of cerebral amyloid plaque. Based on the teachings herein,

those skilled in the art will appreciate that significant reductions in the size and amount of cerebral amyloid plaque formation can be achieved if the initial DMS disruption were prevented (*i.e.*, prevent the disruption of DMS 11 to disrupted DMS 21), or any subsequent DMS disruption were prevented (DMS 12 into disrupted DMS 22, DMS 13 into disrupted DMS 23, etc.), or if the mechanism of DMS disruption altered so that the resulting disrupted DMS initiate less subsequent DMS disruptions when compared to disrupted DMS in control subjects. Hence, the method of the present invention is effective in treating cerebral amyloidosis, including Alzheimer's disease, because the compounds of the invention, when administered, either (a) inhibit DMS disruption, or (b) alter the DMS disruption process in such a manner that subsequent DMS disruptions are prevented or reduced.

Figures 13-26 illustrate how one single DMS 50 can stimulate the growth and disruption of 8 more DMS (51, 52, 53, 54, 55, 56, 57, 58) and, therefore, illustrate the autocatalytic phenomenon brought about by the growth and disruption of DMS in brain. Figure 13 shows one intact and undisrupted DMS 50 in the center with about 12 nerve fibers in the near vicinity, which contain 8 other DMS (51, 52, 53, 54, 55, 56, 57, 58). The small round objects are intact and undisrupted DMS, the lines are nerve fibers and the empty triangles signify nerve cell bodies. In Figure 13, DMS 50, and the remaining DMS (51, 52, 53, 54, 55, 56, 57, 58) are stable.

As shown in Figure 14, DMS 50 is beginning to disrupt and in Figure 15, it has disrupted to the extent that the disrupted region is impinging on a nerve fiber that contains another DMS 51. In Figures 16, 17 and 18, this second DMS 51 begins to disrupt and the transformed redistributed disrupted DMS region enlarges to form fully disrupted DMS 61. In Figure 19, DMS 50 has disrupted to a greater extent than in FIG. 15 and thus impinges on two (2) additional DMS containing nerve fibers containing DMS 52, 53, such that these DMS, in Figures 20, 21 and 22, subsequently disrupt to form disrupted DMS 62 and 63, respectively.

As shown in Figure 23, the central DMS (originally DMS 50) now has disrupted to an even greater extent to form disrupted DMS 60, and impinges on all DMS containing nerve fibers in the vicinity such that the DMS 54, 55, 56, 57

and 58 in these fibers, in Figures 24, 25 and 26 subsequently disrupt to form disrupted DMS 64, 65, 66, 67 and 68, respectively. A comparison of the DMS in Figures 15, 19 and 23 shows that a small disruption (Figure 15) of DMS 50 results in a total of only one secondary disruption (DMS 51 disrupts to form disrupted DMS 61); a larger disruption (Figure 19) of DMS 50 results in a total of three secondary disruptions (DMS 51 to disrupted DMS 61, DMS 52 into disrupted DMS 62 and DMS 53 into disrupted DMS 63); and the largest disruption (Figure 13) to form disrupted DMS 60 resulted in a total of eight secondary disruptions. The geometric progression of subsequent DMS disruption is readily apparent when the eight secondary disruptions each individually cause eight more disruptions for a total of 64, and then those 64 each cause eight more for a total of 512 disruptions, and so on.

The newly discovered mechanism, whereby an initial DMS disruption facilitates subsequent DMS disruptions elsewhere (on other nerve fibers, etc.) through an autocatalytic phenomenon, also implicates a phenomenon of senile amyloid-plaque formation leading to the formation of other senile amyloid plaques. The connection between *de novo* DMS disruption and DMS disruption elsewhere in the brain is evidenced, for example, by observations that:

A. Intact DMS are found within tiny nerve fibers and endings, and the latter are injured by the thousands when a DMS is disrupted and the ensuing injury reaction proceeds to form its associated injury foci of cerebral senile plaque lesions. An initial disrupted DMS injury to a fiber that contains a subsequent DMS at a different location from the initial disrupted DMS explains the initiation of the subsequent DMS disruption. This is illustrated in Figures 1 to 12.

B. Initial DMS disruption leading to subsequent DMS disruption through the autocatalytic phenomenon resulting in exponentially greater numbers of disrupted DMS implies an accelerated course of senile plaque progression in comparatively younger subjects where nerve fiber density per unit volume is greater than in comparatively more elderly subjects. In other words, if there are more fibers per unit volume, then more fibers will pass through an area of disruption, or injury foci. Thus, these fibers that pass through the injury foci, and contain intact DMS, will be injured and as a consequence, more DMS disruptions will result. It has been found in some studies that younger cases of

cerebral amyloidosis or Alzheimer's disease run a more aggressive, faster course. Kono *et al.*, BASIC, CLINICAL AND THERAPEUTIC ASPECTS OF ALZHEIMER'S AND PARKINSON'S DISEASES, Vol. 2, Plenum Press, N.Y., 143-146; Brandt, *et al.*, *Neuropsychiatr. Neuropsychol. Behav. Neurol.*, 2(2): 93-101 (1989); Knesivich, *et al.*, *Psych. Res.*, 14: 255-263 (1984).

5 C. The total number of disrupted plus intact DMS in Alzheimer's disease brain has not been found to be significantly higher than in normal controls, but there obviously are greater numbers of disrupted DMS in Alzheimer's disease brain. Averbach, *Neurology*, 32(2): A227, (1982). The autocatalytic
10 phenomenon accounts simultaneously for: a) more numbers of disrupted DMS in disease compared to controls; b) the absence of a statistically significant difference in starting material quantity (numbers of intact DMS); and c) the sum of intact plus disrupted DMS being equal in Alzheimer's disease and normal individuals. In other words, Alzheimer's disease individuals and normal
15 individuals start with roughly equal numbers of DMS, but the former group have a higher (faster) rate of transformation to disrupted DMS due to the autocatalytic phenomenon whereby an initial DMS disruption causes subsequent DMS disruptions, and so on.

Treatments that will reduce the number of starting DMS, inhibit the
20 growth of DMS, or alternatively that will delay the time of initiation of the process of DMS disruption, will therefore impede the kinetics of the autocatalytic phenomenon. Delay of the initiation can be achieved by (1) delaying the start of the whole process, for example, by inhibiting the growth of DMS, or (2) by delaying an individual DMS disruption. Retardation of the autocatalytic
25 phenomenon also can be effected by reducing the number of subsequent DMS disruptions brought about by an initial DMS disruption. Throughout this description, the expression "reduce the number and/or rate of subsequent DMS disruptions" denotes a process whereby a given DMS (i) does not disrupt, (ii) disrupts to a lesser extent (*i.e.*, smaller injury foci), (iii) disrupts but reduces the
30 inflammatory reaction in relation to persistent DMS materials, or (iv) disrupts but increases the digestion and removal of the disrupted DMS material. Thus, the overall number of subsequent DMS disruptions will be reduced in accordance with the present invention. A treatment that produces a small, perhaps insignificant,

reduction in subsequent DMS disruptions per cycle will, as described above, produce in this exponential process a huge and important reduction in quantities of DMS disruption. This result is that the individual so treated can shift from a high quantity group to a low quantity group and thereby remain asymptomatic, or have fewer symptoms, or have slower progressing symptoms. Quantitative reduction of subsequent DMS disruptions implies that certain individual DMS will have delay of onset of disruption, will not disrupt at all or that disruption will be altered to produce disrupted DMS that do not initiate, or initiate to a lesser extent, further disruptions.

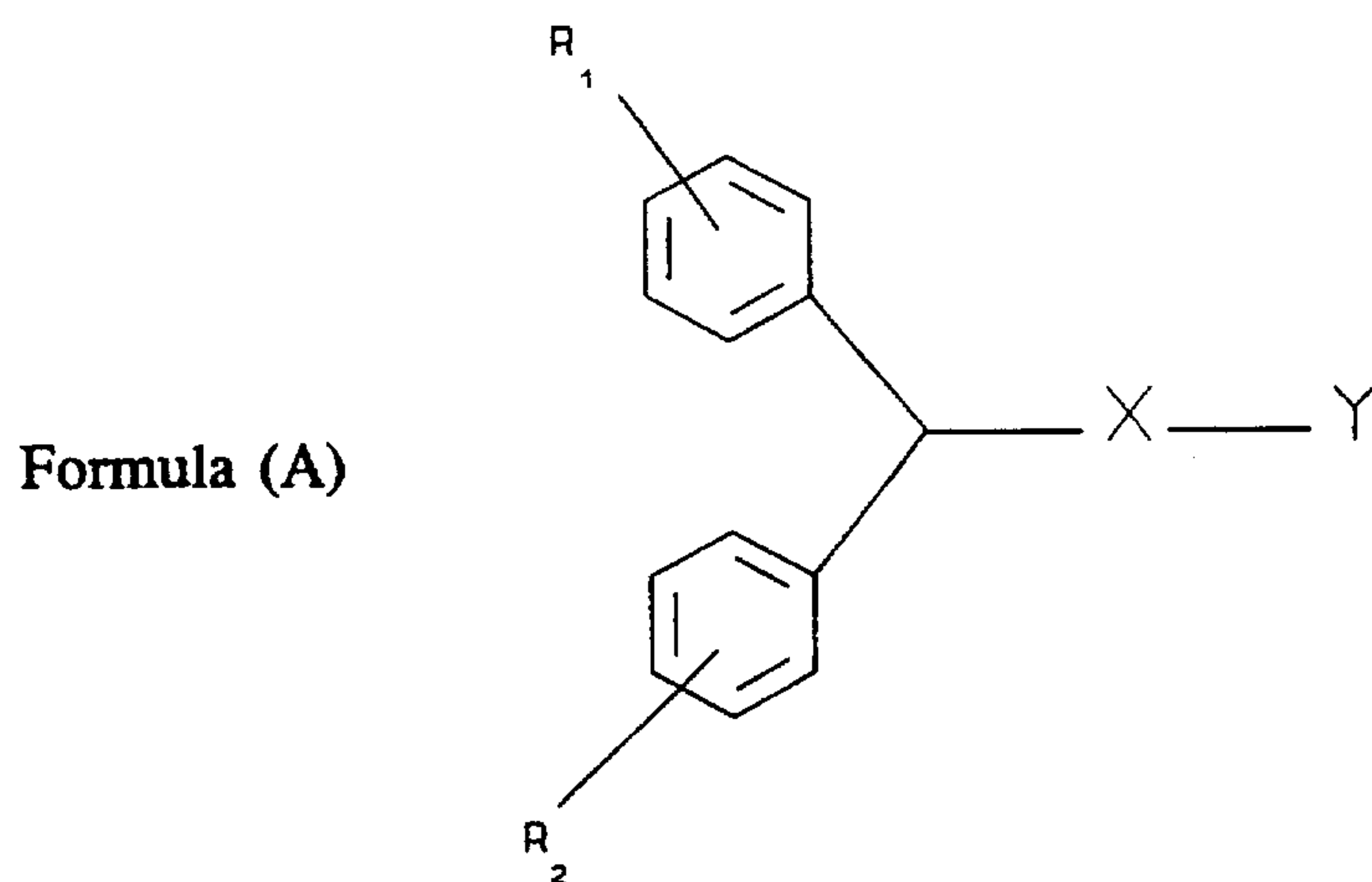
10 The extent of subsequent DMS disruptions brought about by an initial DMS disruption and brain injury process is proportional to the number of DMS containing fibers that are injured by the initial DMS disruption. For example, in an individual with Alzheimer's disease, a large hippocampal cortical senile plaque can have an injury focus with a diameter of about 100 micrometers and a volume
15 of about 525,000 cubic microns. The plaque effects the injury of thousands of fibers which pass through this volume. If the diameter of the injury focus was reduced to about 80 micrometers, with a corresponding volume of about 268,200 cubic microns and many fewer injured fibers, then the number of subsequent DMS disruptions would be reduced by approximately one-half, because the
20 volume has been halved.

 Thus, reducing the diameter of the disrupted DMS by only 20% results in the first instance in a 50% reduction in subsequent DMS disruption. By virtue of the newly discovered autocatalytic phenomenon, this reduction ultimately results in significantly less DMS disruptions overall. A modest diameter
25 reduction in disrupted DMS thereby occasions, at a given point in time, a shift from a high to a low quantity of cerebral amyloid plaques, preventing the subject from becoming symptomatic at that time (see Figures 13-26).

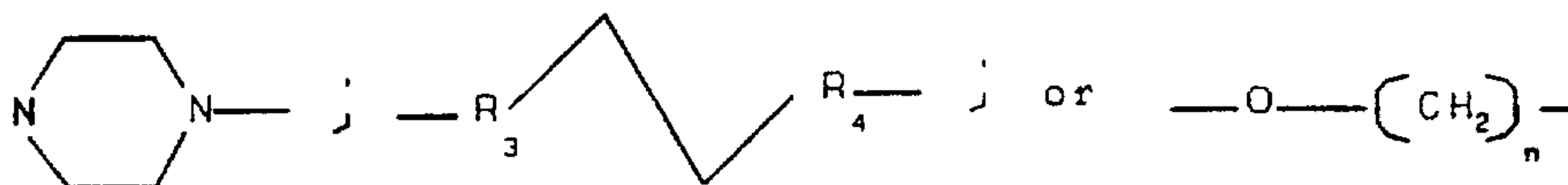
 Compounds that are effective in reducing the quantity or volume of initial disrupted DMS and, hence, the quantity and volume of disrupted DMS injury
30 foci, can be used to treat cerebral amyloidosis, including Alzheimer's disease. Particularly effective in this regard are compounds that act on DMS protein components or DMS membrane, for example, via intracellular or extracellular binding, so as to limit initial DMS disruption by either preventing DMS disruption

altogether or by altering the initial DMS prior to disruption in such a manner that, when disrupted, subsequent DMS disruption is curtailed. Compounds of the present invention are capable of reducing subsequent DMS disruptions brought about by an initial DMS disruption by (i) decreasing the mean tissue volume of
 5 disrupted DMS, (ii) reducing the ratio of number of inflammatory cells per DMS, or (iii) increasing the ratio of the number of macrophages containing disrupted DMS material per DMS, *i.e.*, increasing the digestion and removal of the disrupted DMS material through the reticuloendothelial system.

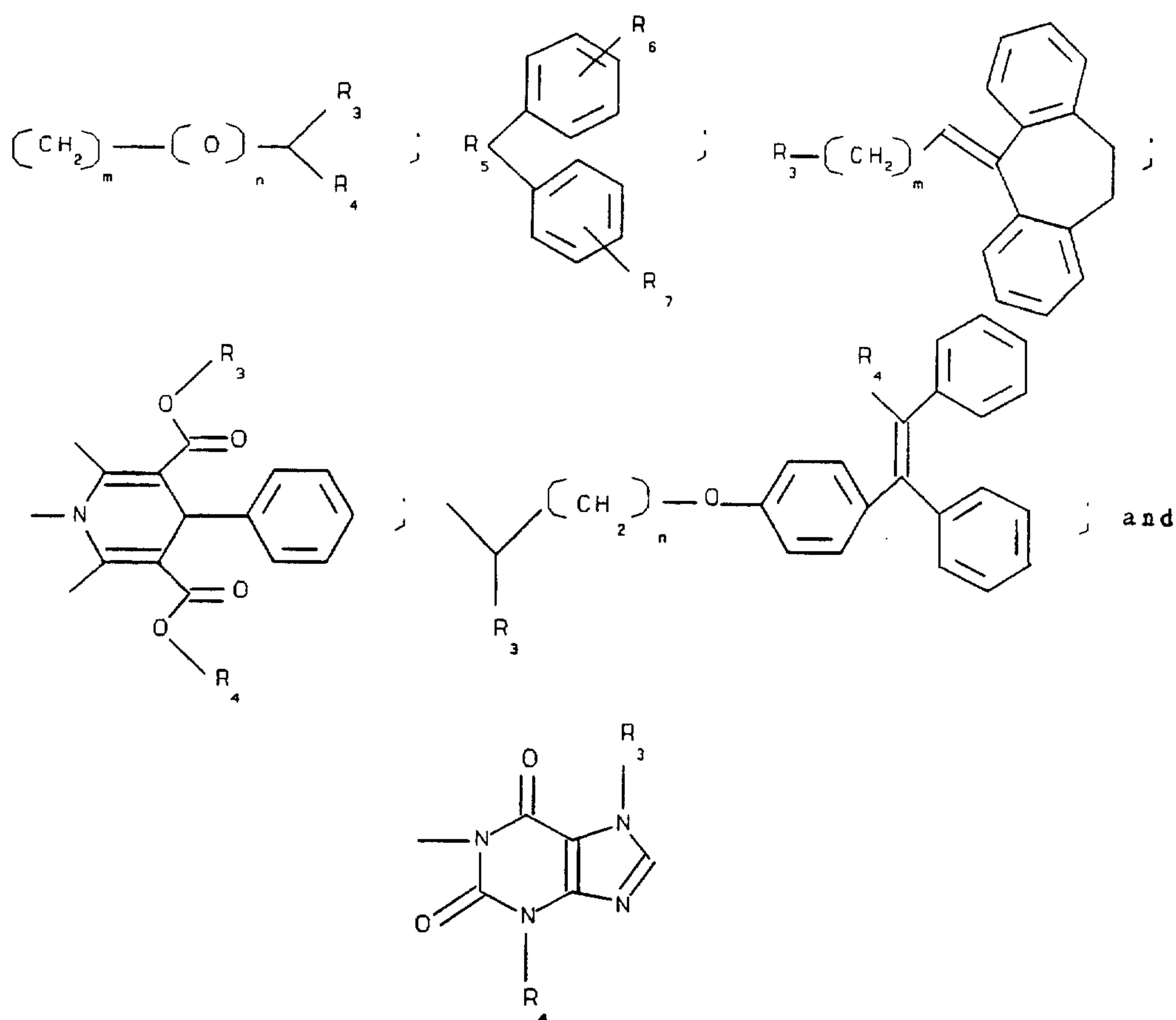
Compounds found to be effective in either preventing DMS disruption or
 10 altering pre-disrupted DMS in any of the manners described above can be represented by the general formula (A):



where X is selected from



and Y is selected from



In the general formula (A) above:

R_1 and R_2 are each one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 alkoxy, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein if R_1 or R_2 are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or unsubstituted C1-C5 alkylamino; wherein if R_1 or R_2 are alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl, and if R_1 or R_2 is a fused ring system, each individual R_1 or R_2 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene,

aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; 5 wherein each ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_3 is hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, amino, C1-C5 alkyl-substituted amino, sulfur, oxygen, phenyl, benzyl, naphthyl and anthracenyl; wherein each aromatic ring may be substituted by one 10 or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_4 is hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, amino, C1-C5 alkyl-substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted 15 C2-C5 alkenylamino, sulfur, oxygen, phenyl, benzyl, naphthyl and anthracenyl; wherein each aromatic ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_5 is nitrogen or carbon;

20 R_6 and R_7 are each one or more independent substitutions selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein if R_6 or R_7 is piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected 25 from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or unsubstituted C1-C5 alkylamino; wherein if R_6 or R_7 is alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl, and if R_6 or R_7 is a fused ring system, each individual R_6 and R_7 30 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline,

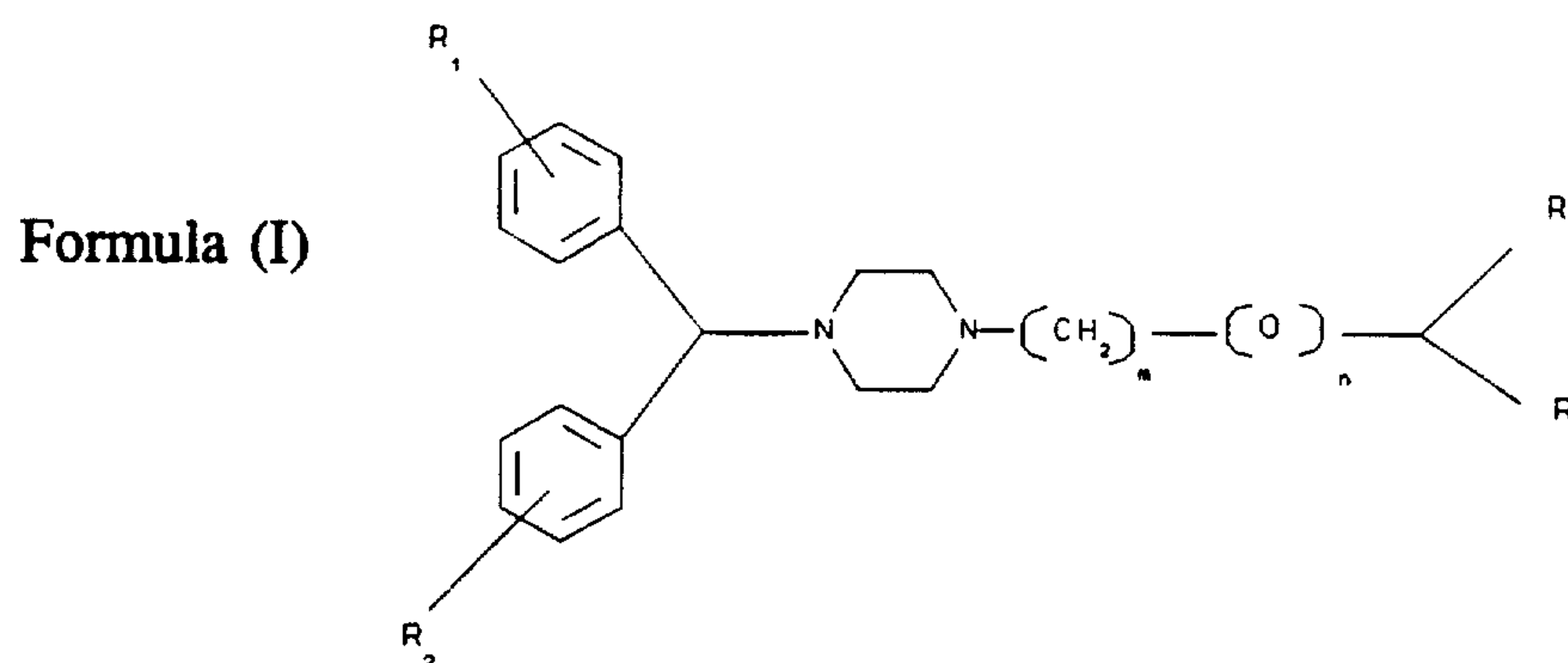
phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; wherein each ring may be substituted by one or more substituents selected from
 5 halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

n is an integer of from 0 to 5;

m is an integer of from 0 to 5;

or a pharmaceutically acceptable salt of such compound.

Particularly preferred in this regard are compounds of any of formula (I)-
 10 (VI) below.

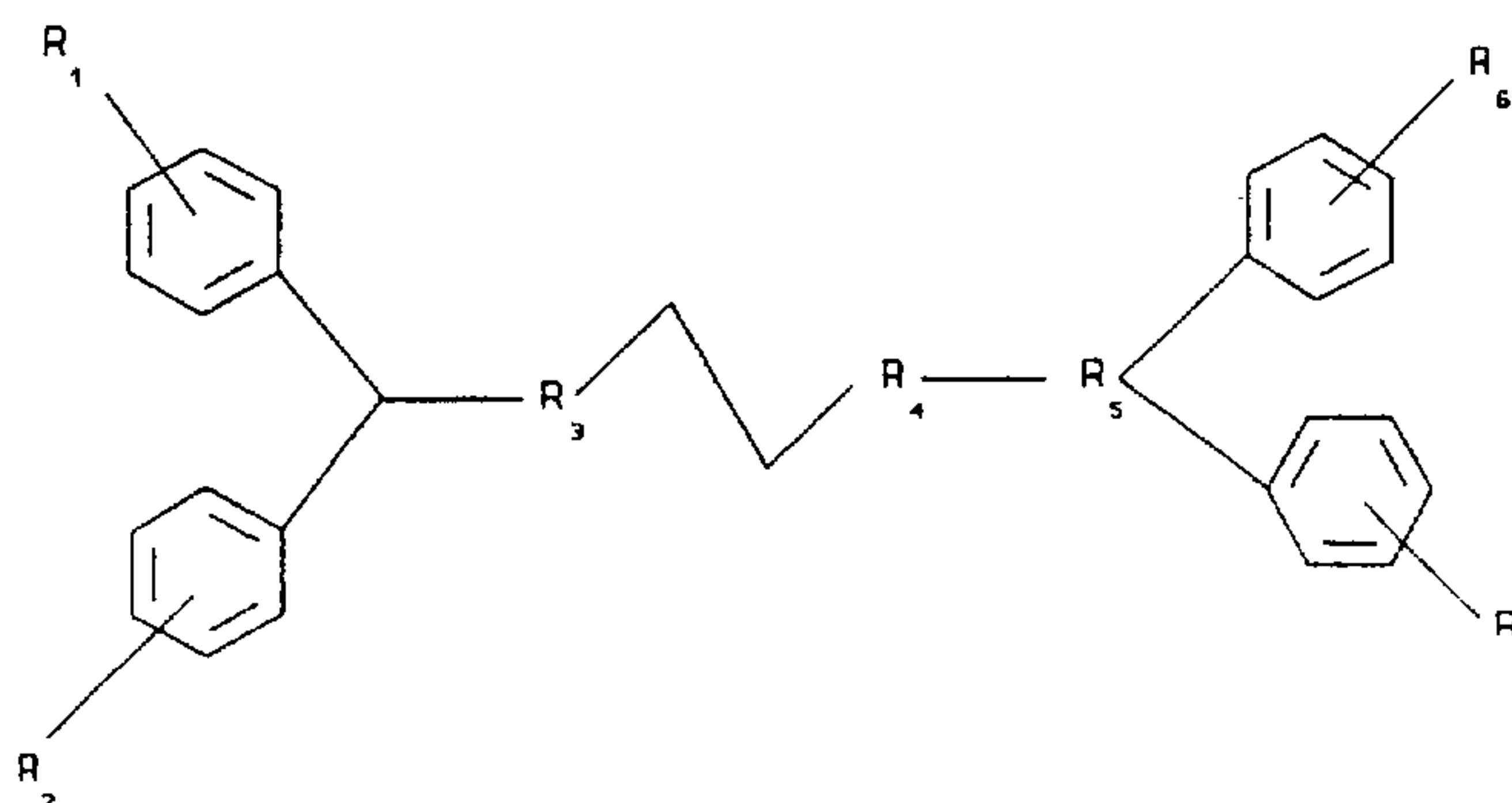


wherein R_1 and R_2 can each be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, nitro and halogen; R_3 and R_4 can independently be selected from C1-C5 alkyl,
 15 C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl and anthracenyl. Each aromatic ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl. In Formula (I) m is 0 to 5 inclusive, and n is 0 or 1. Pharmaceutically acceptable salts of these compounds also are encompassed by
 20 the present invention.

Particularly preferred compounds in accordance with Formula (I) are those where R_1 and R_2 are each hydrogen, R_3 and R_4 are selected from C1-C5 alkyl and benzyl, with each phenyl moiety being unsubstituted or substituted with one or more substituents selected from halogen and haloalkyl, and m and n are each 0.
 25 Other compounds of Formula (I) include those where R_1 and R_2 are each

hydrogen, R_3 is phenyl, R_4 is selected from C1-C5 alkyl and C1-C5 haloalkyl, m is 1 to 5 inclusive and n is 0. Additional compounds include those of Formula (I) where R_1 and R_2 are each hydrogen, R_3 is methyl and R_4 is phenyl, wherein R_4 is the only substituted phenyl moiety, the substitution selected from halogen and trifluoromethyl. Other preferred compounds include those of Formula (I) where n is 1 or where R_1 and R_2 are each hydrogen and R_3 and R_4 are each phenyl.

Formula (II)



wherein R_1 , R_2 , R_6 and R_7 can each be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems. If any of R_1 , R_2 , R_6 and R_7 are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or unsubstituted C1-C5 alkylamino. Moreover, if any of R_1 , R_2 , R_6 and R_7 are alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl, and, if any of R_1 , R_2 , R_6 and R_7 is a fused ring system, then each individual R moiety involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part. In this case, each

ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl. In Formula (II), R₃ can be selected from C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur and oxygen; R₄ can be selected from C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur and oxygen; and R₅ can be nitrogen or carbon. Pharmaceutically acceptable salts of such compounds also are encompassed by the present invention.

10 Illustrative preferred compounds within Formula (II) are (A) those in which each of R₁, R₂, R₆ and R₇ is an independent substituent and also (B) those where, if any of R₁, R₂, R₆ and R₇ is a fused ring system, each R₁, R₂, R₆ and R₇ moiety involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline. With respect to category (A), R₁, R₂, R₆ and R₇ preferably are selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, 15 halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, piperazinyl, and pyridylmi; if any of R₁, R₂, R₆ and R₇ are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, substituted or unsubstituted C1-C5 alkylamino, and if any of R₁, R₂, R₆ and R₇ are alkylamino, each alkylamino can consist of 1 to 5 carbon atoms and the amino group can be unsubstituted or mono-substituted or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl. Even more preferred in this regard are compounds of Formula (II) where R₁, R₂, R₆ and R₇ are each selected from hydrogen and C1-C5 alkyl, and R₅ is carbon. A more preferred compound is where R₁, R₂, R₆ and R₇ are each hydrogen, R₃ is C1-C5 alkyl and R₄ is selected from C1-C5 alkyl and oxygen.

Other preferred compounds useful in the present invention include those where R₁, R₂, R₆ and R₇ are each hydrogen, R₃ is selected from amino and C1-C5 alkyl substituted amino and R₄ is selected from oxygen, sulfur, or C1-C5 alkyl,

or where R_1 , R_2 , R_6 and R_7 are each hydrogen, R_3 is sulfur and R_4 is selected from oxygen, sulfur or C1-C5 alkyl, or where R_1 , R_2 , R_6 and R_7 are each hydrogen, R_3 is oxygen and R_4 is C1-C5 alkenyl. In addition to those compounds mentioned above, other preferred compounds useful in this invention include those of Formula (II) where R_1 and R_6 are each hydrogen, R_2 and R_7 are each methyl, R_3 is selected from amino and C1-C5 alkyl substituted amino, and R_4 is C1-C5 alkenyl, or where R_1 and R_6 are each methyl, R_2 and R_7 are each hydrogen, R_3 is oxygen and R_4 is C2-C5 alkenylamino. Other preferred compounds are those where R_5 is nitrogen, and where R_1 , R_2 , R_6 and R_7 are each hydrogen, R_3 is selected from oxygen, nitrogen and C1-C5 alkyl and R_4 is C1-C5 alkyl.

Other preferred compounds useful in the present invention include those where R_3 and R_4 are each oxygen and R_5 is carbon, and R_1 , R_2 , R_6 and R_7 are each a single substitution selected from hydrogen, halogen, C1-C5 haloalkyl, C1-C5 alkyl, C1-C5 alkoxy, substituted or unsubstituted piperazinyl, alkylamino, phenyl, and fused ring systems, and preferably R_1 , R_2 , R_6 and R_7 are selected from hydrogen, chlorine, methoxy, trifluoromethyl, methylamino, piperazinyl, phenyl and fused ring systems, and wherein if any of R_1 , R_2 , R_6 and R_7 is a fused ring system, each individual R_1 , R_2 , R_6 and R_7 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein each ring may be substituted by one or more substituents selected from halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl.

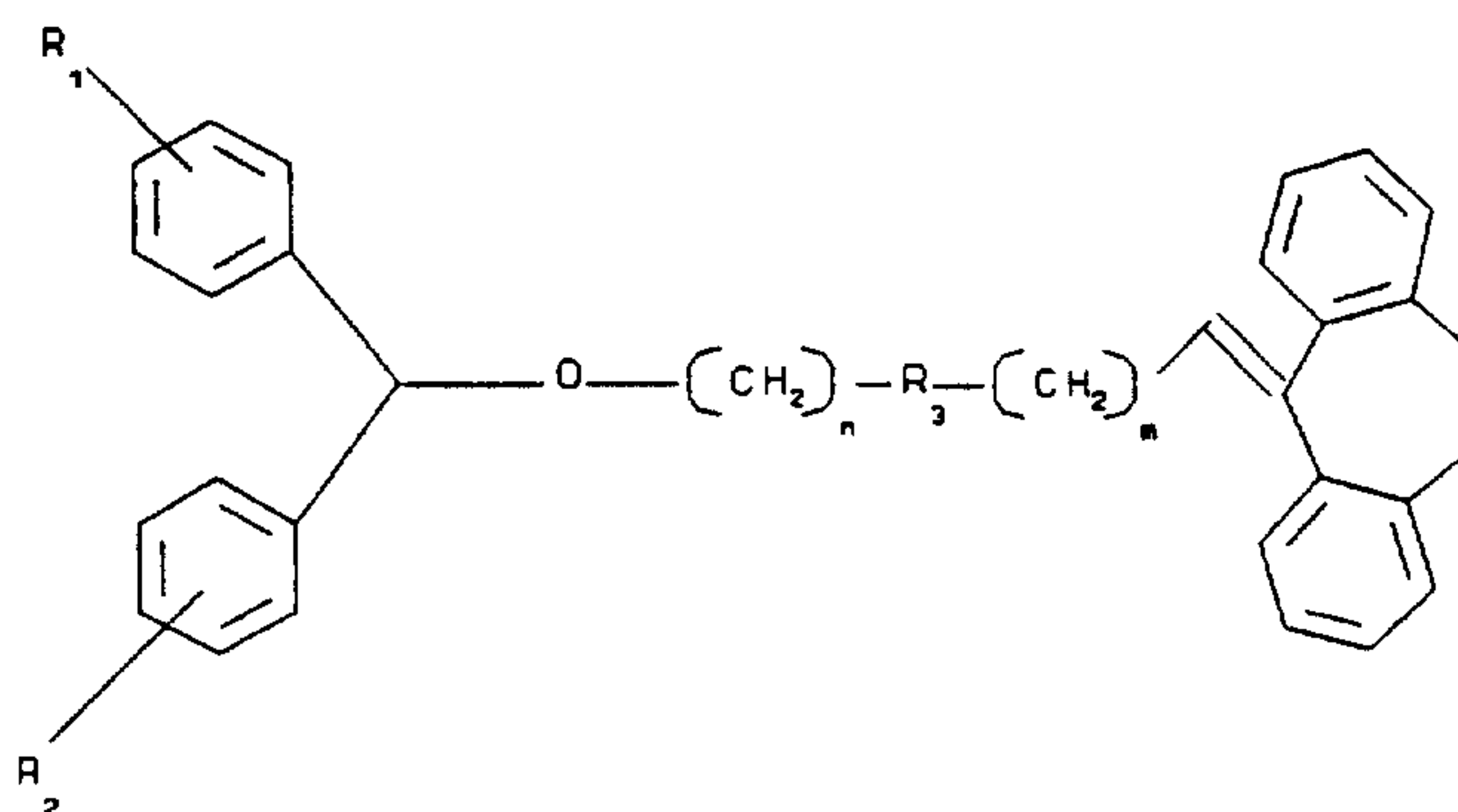
Even more preferred in this regard are compounds of Formula (II) where R_1 , R_2 , R_6 and R_7 are selected from hydrogen, chlorine, C1-C5 alkoxy, trifluoromethyl, substituted or unsubstituted methylamino, piperazinyl, and phenyl, preferably selected from hydrogen, chlorine, and trifluoromethyl and more preferably are hydrogen. Moreover, R_1 and R_6 may be hydrogen and R_2 and R_7 may be chlorine, or vice versa, or where R_1 and R_6 can be any one of trifluoromethyl, C1-C5 alkyl, methyl or a part of a fused ring, and R_2 and R_7 are hydrogen, or vice versa. Other compounds useful in the present invention are

those where R_1 , R_2 , R_6 and R_7 are each alkylamino, wherein each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted C1-C5 alkyl, C2-C5 alkenyl, or C2-C5 alkynyl.

Additional useful compounds of the present invention are those of formula (II) where R_1 and R_6 are each alkylamino, wherein each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted C1-C5 alkyl, C2-C5 alkenyl, or C2-C5 alkynyl, and R_2 and R_7 are hydrogen, or where each of R_1 , R_2 , R_6 and R_7 is selected from hydrogen, phenyl and fused ring systems, wherein if any of R_1 , R_2 , R_6 and R_7 is a fused ring system, each individual R group involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part, or more preferably, if any of R_1 , R_2 , R_6 and R_7 is a fused ring system, each individual R group involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, or even more preferably R_1 and R_6 are piperazinyl or alkoxy and R_2 and R_7 are hydrogen.

25

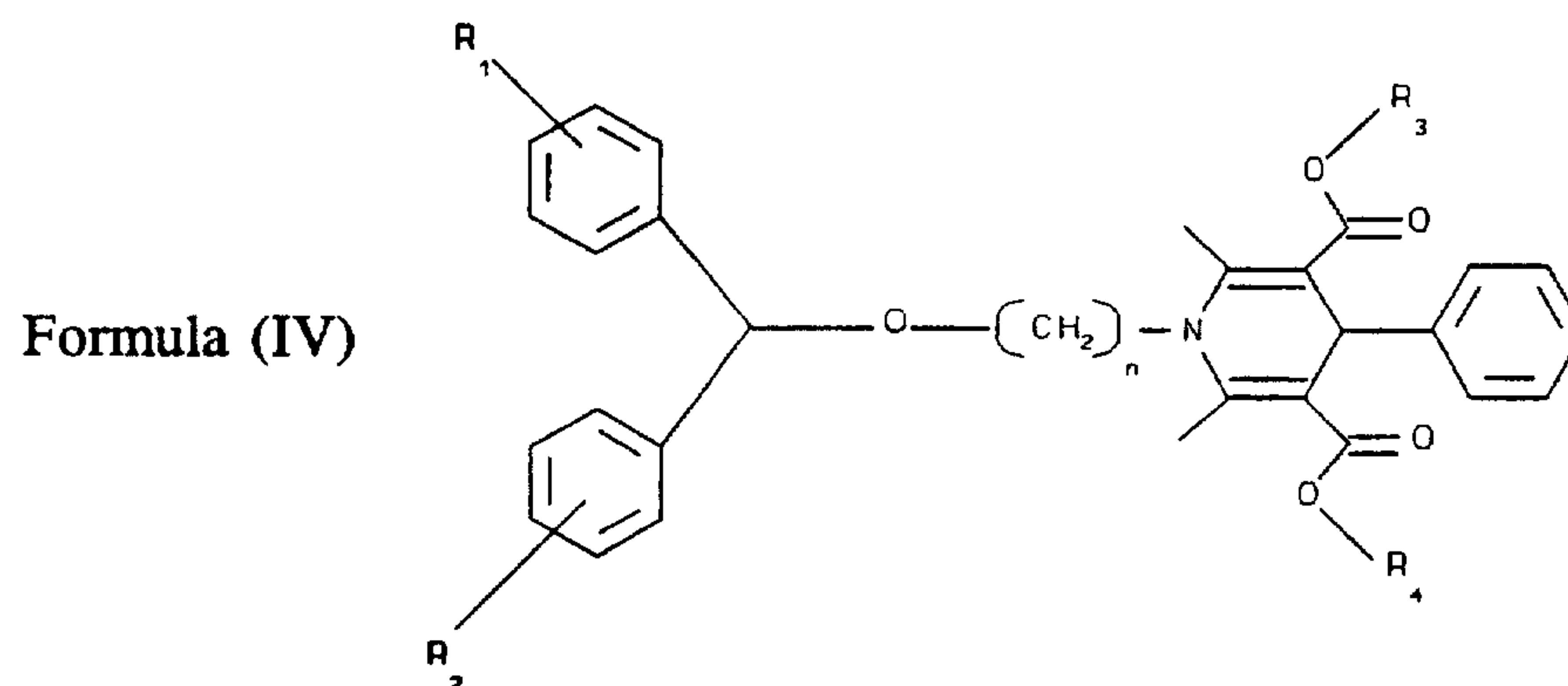
Formula (III)



wherein R_1 and R_3 can each be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy,

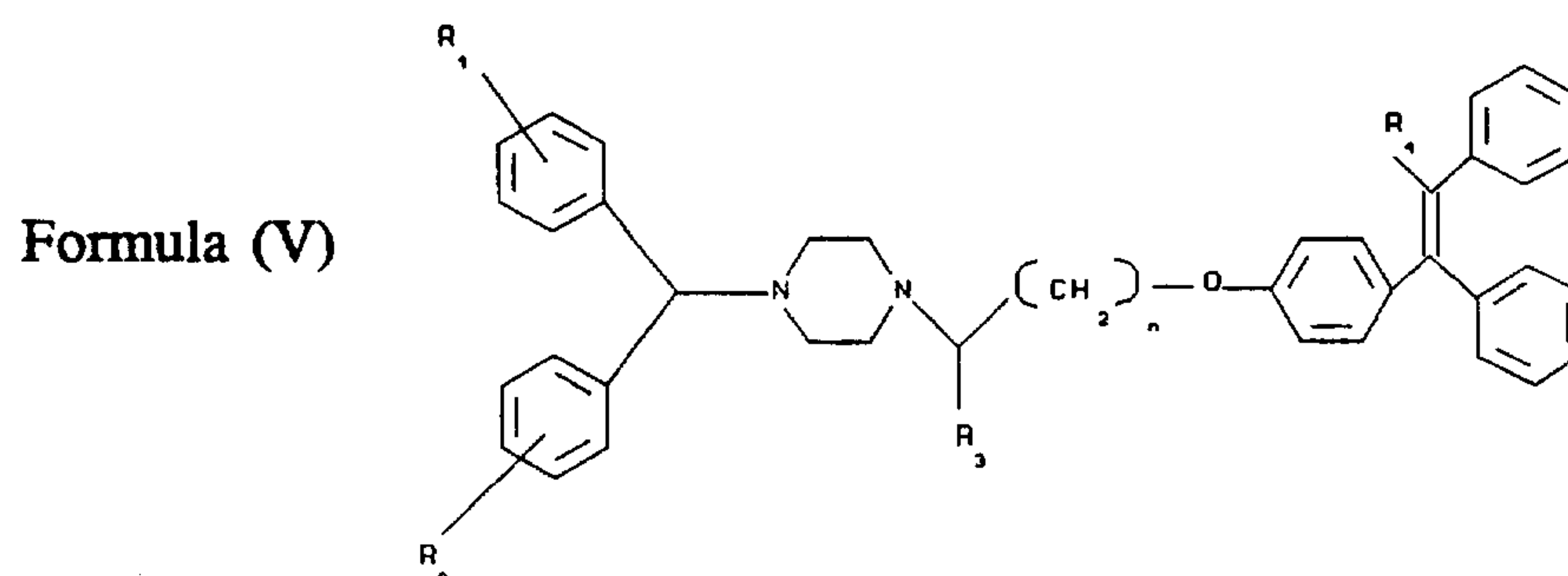
C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro and carboxyl. In Formula (III), R_3 can be selected from amino, C1-C5 substituted amino and CH_2 ; and n and m can independently be an integer of from 0-5. Pharmaceutically acceptable salts of these compounds also are encompassed by the present invention.

Particularly preferred compounds of Formula (III) include those where R_1 and R_2 are each hydrogen, R_3 is methylamino, and n and m are each 2, and more preferably where R_1 and R_2 are each hydrogen and R_3 is CH_2 .



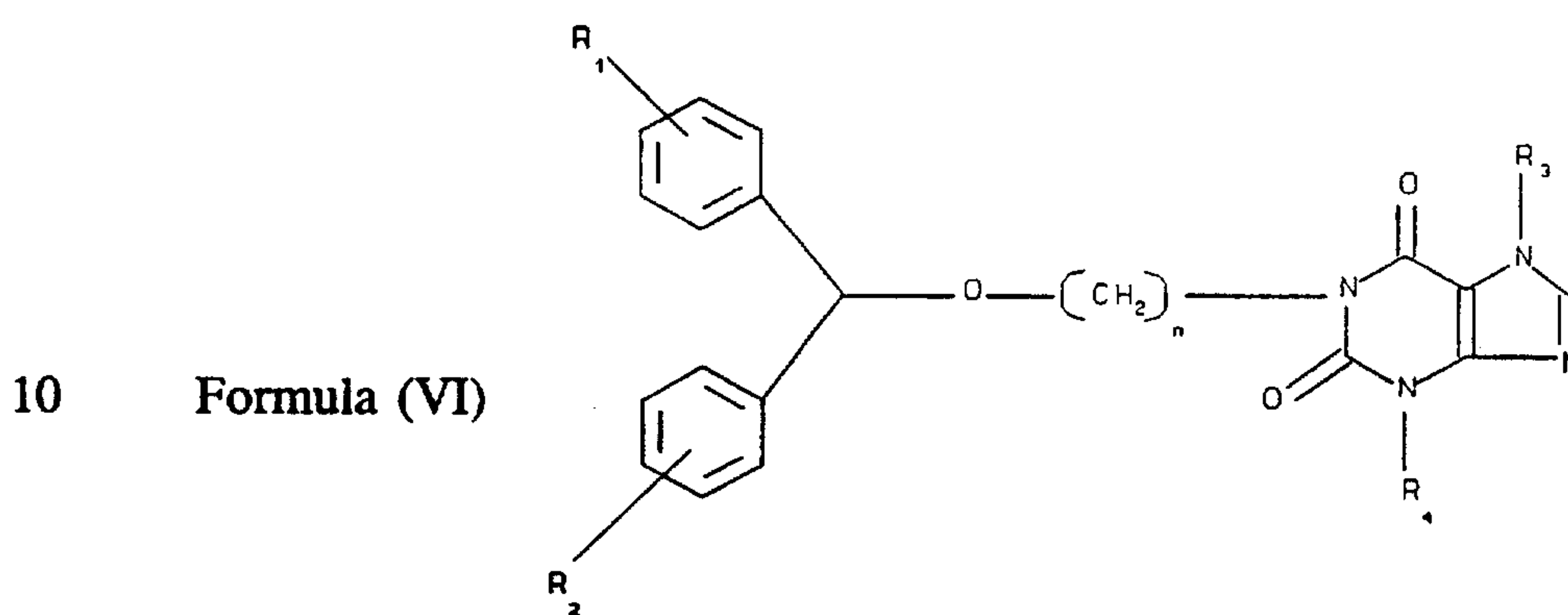
wherein R_1 and R_2 each can be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro and carboxyl. R_3 and R_4 each can be independent and selected from hydrogen and C1-C5 alkyl, and n can be 1 to 5 inclusive. Pharmaceutically acceptable salts of these compounds also are encompassed by the present invention.

Particularly preferred compounds of Formula (IV) include those where R_1 and R_2 are each hydrogen, R_3 and R_4 are each methyl, and n is 2.



wherein R_1 and R_2 each can be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro and carboxyl. R_3 can be selected from hydrogen and C1-C5 alkyl, R_4 can be selected from hydrogen and C1-C5 alkyl, and n can be 1 to 5 inclusive. Pharmaceutically acceptable salts of these compounds also are encompassed by the present invention.

Particularly preferred compounds of formula (V) are those where R_1 and R_2 are each hydrogen, or where R_3 is methyl, and n is 1.



wherein R_1 and R_2 each can be one or more independent substituents selected from hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro and carboxyl. R_3 and R_4 each can be independent and selected from hydrogen and C1-C5 alkyl, and n is 1 to 5 inclusive. Pharmaceutically acceptable salts of these compounds also are encompassed by the present invention.

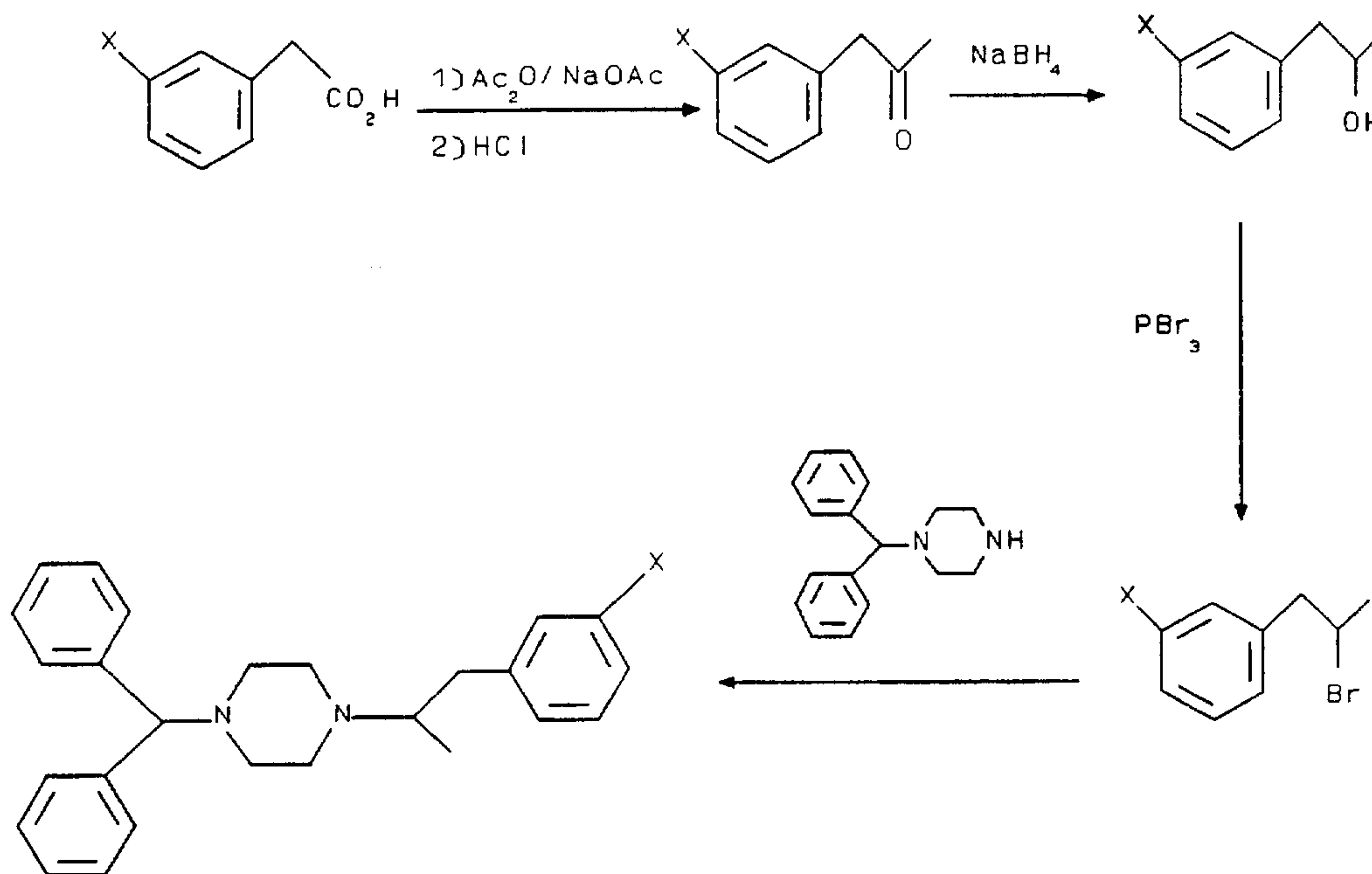
Particularly preferred compounds of Formula (VI) include those where R_1 and R_2 are hydrogen, where R_3 is hydrogen, R_4 is methyl and n is 2, or where R_3 is methyl, R_4 is methyl and n is 2.

Any of these compounds can be formulated into a pharmaceutically acceptable composition for administration to a mammal or test animal whereby the composition would include a pharmaceutically acceptable carrier and a therapeutically effective amount of the compound.

Skilled artisans are capable of synthesizing any of the aforementioned compounds using methods known to those skilled in the art. The following

synthesis pathways illustrate how various compounds can be made in accordance with the present invention.

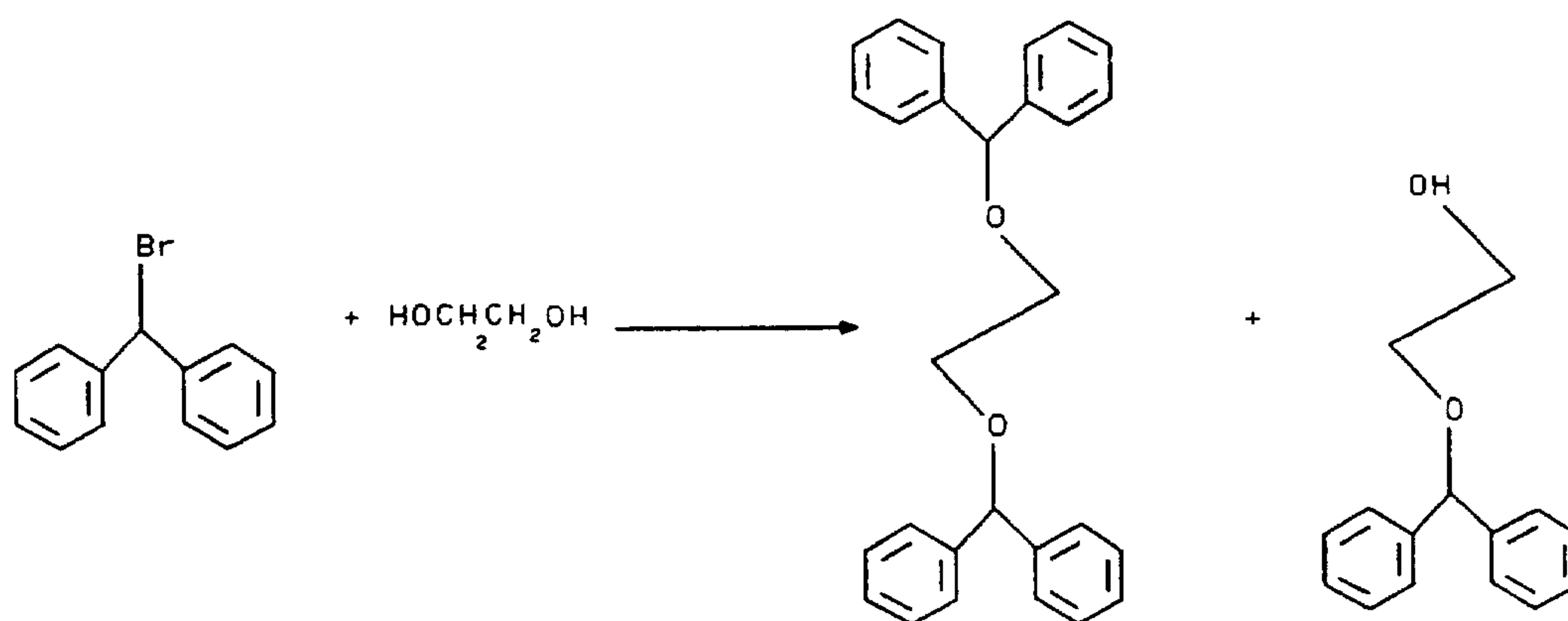
Synthesis Pathway for Compounds of Formula (I):



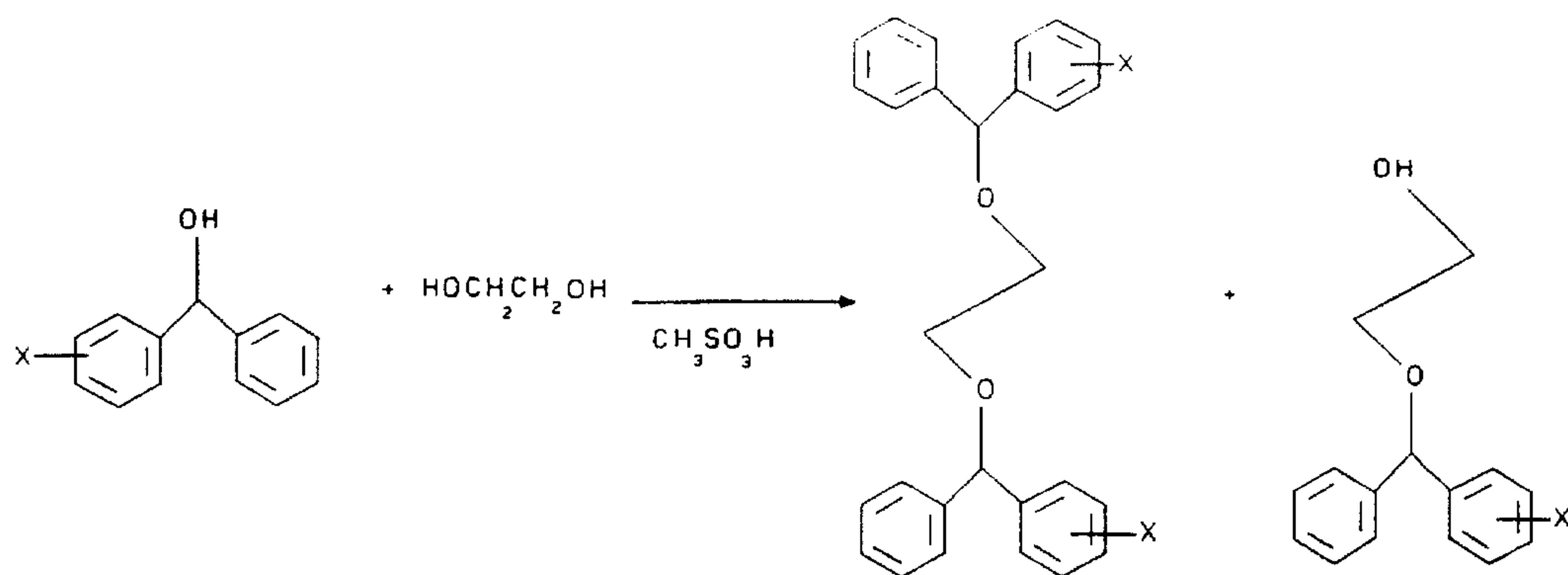
where x can be selected from H, Cl, CF₃ or C1-C5 alkyl.

5

Synthesis Pathway for Compounds of Formula (II):

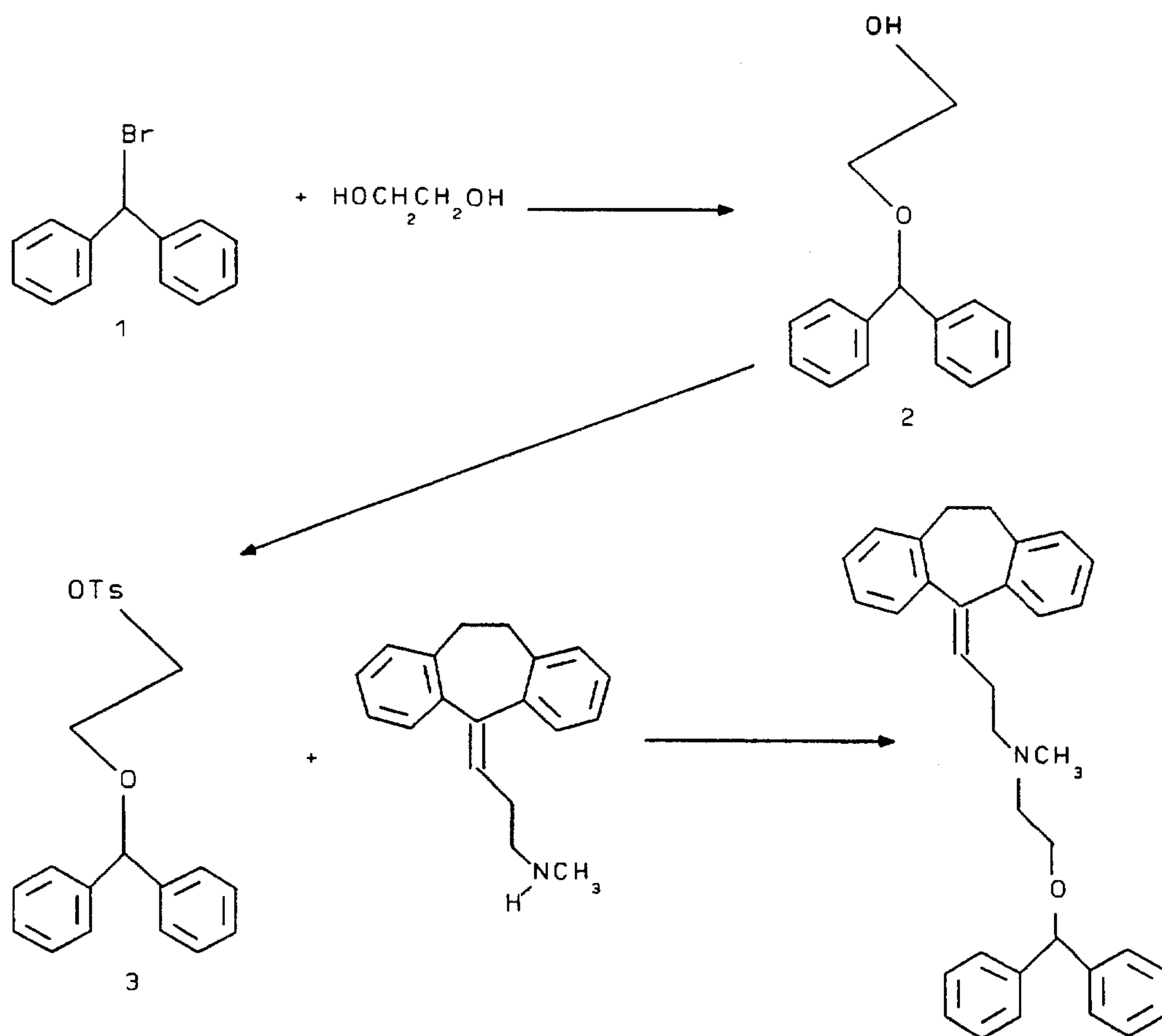


OR



where x can be CH_3 , OCH_3 , CF_3 , Cl, Br, F or other halogen, CO_2R , NR_3 , cyclohexane, naphthyl, SO_3 or CONH_2 , where R can be hydrogen or C1-C5 alkyl.

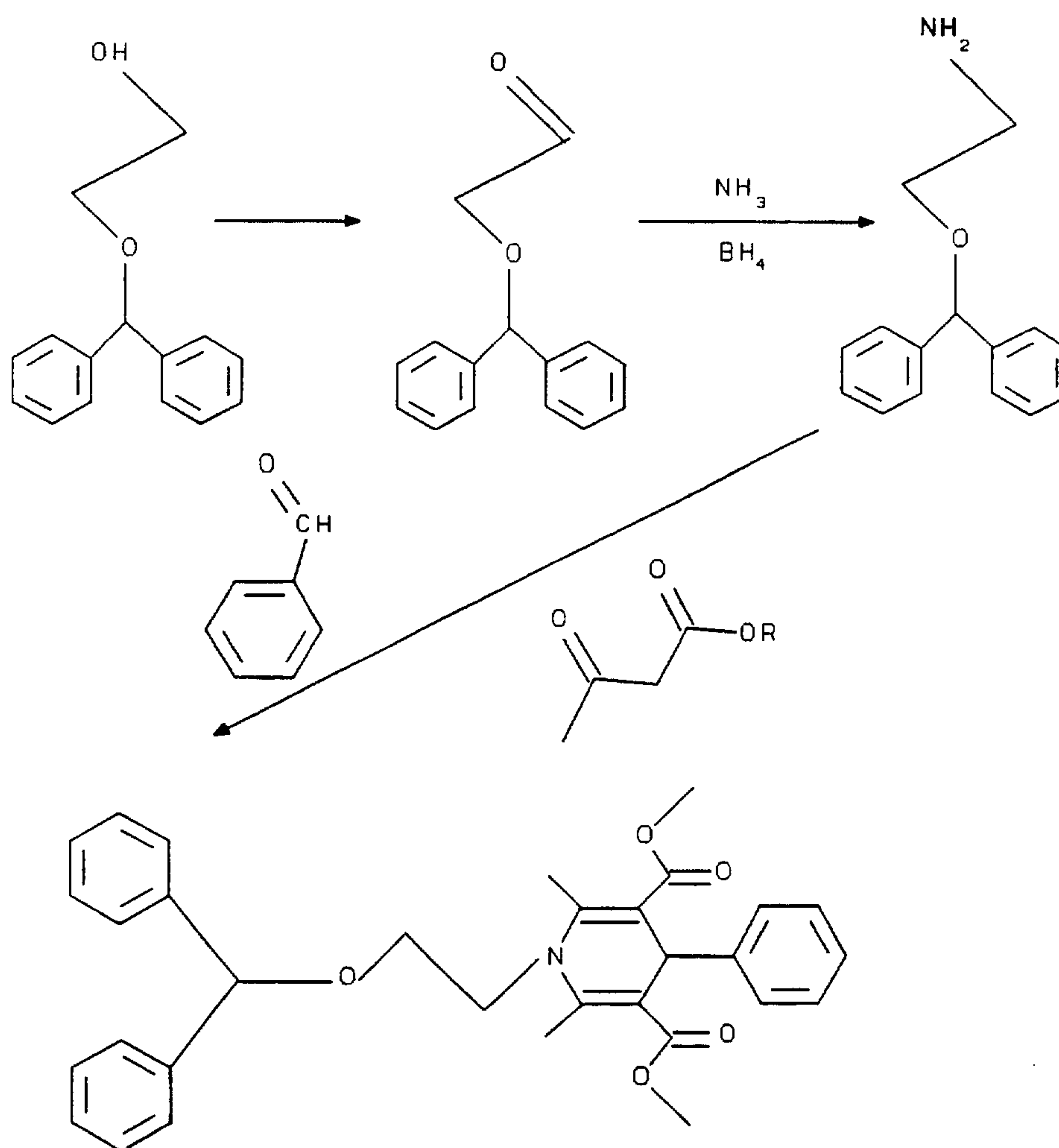
Synthesis Pathway for Compounds of Formula (III):



where Ts denotes a tosylate derivative.

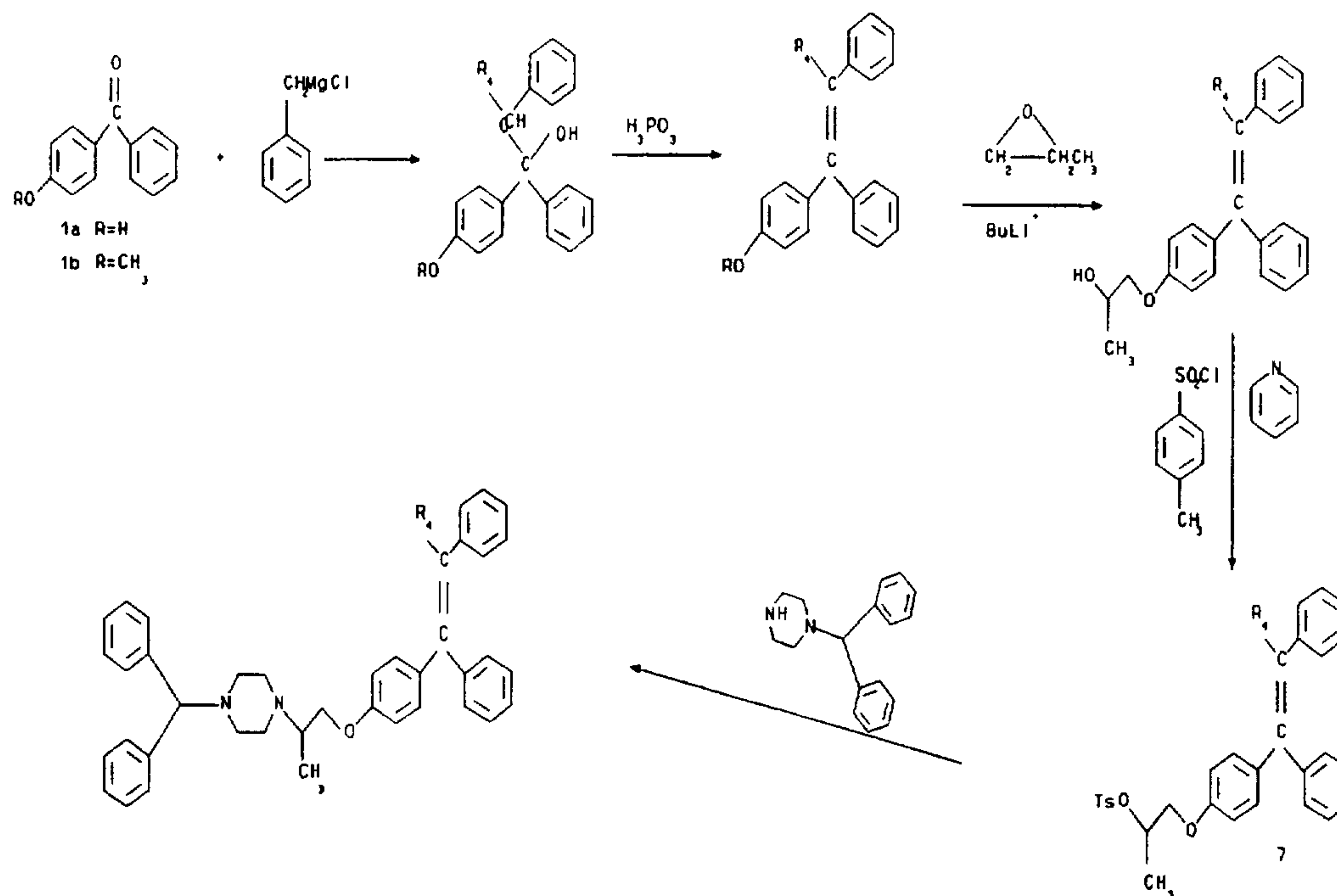
Synthesis Pathway for Compounds of Formula (IV):

Compounds of Formula (IV) can be prepared using techniques known in the art. For example, compound 3 from the Formula (II) compound synthesis can be modified by replacing the OH with NH₂, and then this compound can be used in the Hantzsch pyridine synthesis, together with benzylaldehyde and a β-keto ester. the Hantzsch pyridine synthesis is described, for example, in Streitwieser, INTRODUCTION TO ORGANIC CHEMISTRY, Macmillan Publishing Co., Inc., N.Y. 1097 (1976). An exemplary synthesis pathway is illustrated below.



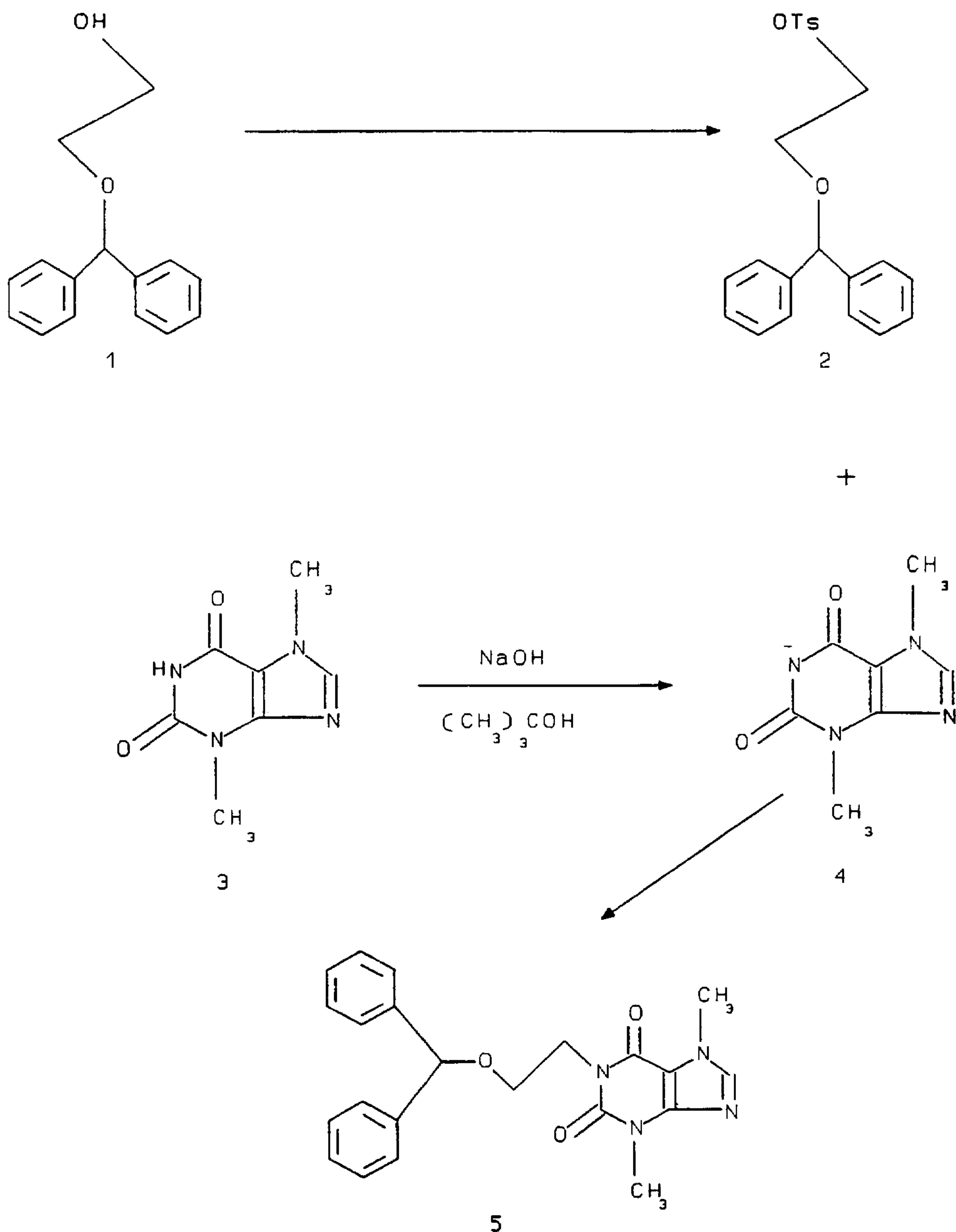
where R can be any unsubstituted or substituted alkyl radical having from 1-5 carbon atoms.

Synthesis Pathway for Compounds of Formula (V):



where R₄ can be H or C1-C5 alkyl, and where Ts denotes tosylate. If R is CH₃, the methyl group can be removed by conventional means, including, for example, reaction with NaCN.

Synthesis Pathway for Compounds of Formula (VI):



where Ts represents a tosylate derivative, and compound number 2 is the same as compound number 3 in the synthesis of Formula (III) compounds. Those skilled in the art will appreciate that other routes can be used to synthesize any of the compounds of Formula (I)-(VI), using the guidelines provided herein.

The microspheric bodies (DMS) employed according to the present invention are derived from mammalian brain tissue and are characterized, in essentially homogeneous form, by a range of diameters from about 0.1 μm to

about 15 μm , by the proteinaceous core structure of DMS, and by certain stainability properties. (In this regard, "homogeneous" means that the DMS represent the only protein structure discernible in the subject composition at the light-microscopic level.) For example, the microscopic bodies of the present invention are homogeneously electron-dense when stained with osmium and lead, and can be visualized by thin-section electron microscopy; under optical microscopic examination, they appear eosinophilic and phloxinophilic, and are nonbirefringent when stained with Congo Red. When the microspheric bodies of the present invention are disrupted, a material is produced that displays congophilic birefringence; that is, when stained with Congo Red the material becomes optically anisotropic to the extent of splitting an incident light wave into two waves with mutually perpendicular vibrational planes and produces a red-green color change when viewed with cross polarizers.

DMS are spherical, membrane-bounded, intracellular structures, about 0.1 and 15 μm in diameter, that are found in human and other mammalian brains. More specifically, the normal location for DMS is in gray-matter neuropil, where the spherical structures are enclosed in tiny, neuronal cellular processes. DMS are solitary, non-perikaryal and non-confluent, and are not found in cerebellum or in white matter. With regard to inter-DMS distances, the spatial distribution of DMS in gray matter regions is random. Compositions of DMS in homogeneous form can be produced by extraction, according to the present invention, to give homogeneous samples of DMS.

The following procedure can be followed to extract DMS from brain tissue:

(1) Whole brain is removed from the skull postmortem, by use of standard postmortem techniques for humans or animals. The best results are obtained if the organism has been in circulatory arrest for less than six hours at the time of brain removal and if the body has been refrigerated as early as possible postmortem. DMS are still extractable at postmortem intervals greater than six hours and are still extractable if body cooling has been delayed or absent, but these two factors will usually greatly decrease the overall average yield of DMS per individual brain. In addition to the effect of postcirculatory arrest interval and temperature on DMS yield, there is considerable individual variation

in DMS content per brain, and also individual variation in DMS extractability, which may be related to agonal metabolic state, overall disease status or other factors. All of the factors which determine total DMS yield per brain can have an impact on DMS extraction, since the volume of homogeneous DMS will decrease proportionally to any reduction in percentage extractability; such a decrease may be sufficient to hinder accurate recognition during the extraction procedure. Furthermore, the screening of putative anti-amyloidosis therapies and the characterization of isolated samples of DMS, in accordance with the present invention, are rendered correspondingly more difficult and costly, and ultimately may be impossible at critically small volumes of DMS.

(2) By means of clean instruments, the freshly removed brain is immediately dissected. Dissection is optimally performed in a cold room at 10°C. By means of careful, but rapid, sharp and blunt dissection, the internal capsules, corona radiata, centra semi-ovale, brain stem, cerebellum, lepto and pachymeninges, arachnoid granulations, coroid plexi, and blood vessels are separated and discarded, and the remaining mass of brain is rapidly used for the subsequent steps. (Standard blocks for microscopic study can be removed at this stage and stored separately in histological fixative.) The dissection brain mass ("DBM") is optimally utilized immediately after dissection. It may also be stored frozen at temperatures of -10°C to -70°C, but this decreases the overall average yield of DMS per individual brain.

(3) The extraction of DMS material from DBM can be carried out by a combination of centrifugation steps. In an exemplary extraction, DBM mechanically homogenized in a 2:1 volume of 0.5 M TRIS-HCL buffer (pH 7.5) is subjected to centrifugation at about 200 rpm for some 10 minutes. (All manipulations are carried out at around 4°C.) The sediment thus obtained ("Sediment I") is separated across a sucrose gradient (1.589 M, or 45%; 1.895 M, or 52%; 2.3895 M, or 62.5%) via centrifugation at 26,000 rpm for 30 minutes. It has been found that the material that settles at the interface between 1.895 M and 2.1015 M (56.7% sucrose) is the DMS containing fraction, as may be confirmed by microscopic examination, with eosin staining, of the fraction.

The DMS-containing fraction obtained from Sediment I consists essentially of the dense microspheres described above, and can be used in a screening

method according to the present invention. It is preferable, however, for the fraction to be subjected to additional manipulations in order to enrich the DMS concentration. To this end, it has proved useful, for example, to wash the DMS-containing fraction in buffer -- the above-mentioned homogenization buffer is
5 suitable for this purpose -- and to spin the resulting mixture again (10,000 rpm for 7 minutes) to obtain DMS-enriched sediment ("Sediment II").

As with Sediment I, Sediment II can be run through a density gradient to enrich further the yield of DMS. It has been discovered that the carbohydrate Percoll® (Pharmacia) is particularly useful in this context. A commercially
10 available formulation of 80% Percoll® (1.13 g/ml) in 0.15 M NaCl provides a iso-osmolar gradient to which Sediment II can be subjected (30,000 rpm for 15 minutes); successive samples, on the order of 0.25 to 1 cc each, then can be taken along the length of the gradient and the DMS-enriched fractions isolated. If these fractions are washed again in buffer, they can be spun down once again (15,000
15 rpm for 10 minutes) to obtain a sediment ("Sediment III") that is substantially pure DMS.

The DMS materials obtained as described above can be used, pursuant to the present invention, in screening anti-amyloidosis therapies. In particular, homogeneous DMS material within the present invention can be employed to
20 ascertain effectiveness *in vitro*, on the part of an active agent or a treatment comprising a putative therapy for cerebral amyloidosis, in reducing the extent of DMS disruption induced by autocatalytic DMS disruption.

By whatever means DMS are disrupted in control samples, a putative anti-amyloidosis agent or therapy can be screened by virtue of its ability to reduce the
25 volume of tissue occupied by the redistributed transformed DMS protein material and associated injury foci under test conditions. For example, *in vitro* DMS disruption on an appropriate viewing surface, such as a glass or plastic slide (see Test 1 below), can be accomplished by mechanical means; by the action of an enzyme treatment, or other chemical exposure. Disruption of DMS can also be
30 effected by injecting DMS material of the present invention into an isolated tissue sample (see Test 2). Brain slices are preferred for this purpose, but liver, pancreas and other organs are also acceptable sources for tissue samples.

An active agent or a treatment that proves effective *in vitro* is then tested, pursuant to the present invention, for *in vivo* efficacy in preventing amyloid-fibril formation by reducing the number, quantity and volume of DMS disruption (to thereby reduce and/or prevent future DMS disruption) in an animal model for cerebral amyloidosis comprised, for example, of a rat, dog, cat or other suitable laboratory animal that has been injected with homogeneous DMS material, pursuant to the present invention. Although intracerebral injection is preferred, injection sites in a test animal's body other than in the brain, such as in skin and in muscle, are suitable for determining the ability of a proposed active agent or treatment step to hinder the resulting formation of amyloid. Because simple injection of DMS onto a glass slide does not result in amyloid formation, it is understood that *in vivo* production of amyloid fibrils upon injection of DMS occurs in the extracellular spaces of the injected tissue.

For the present invention, an active agent is useful, when present at in-tissue concentrations of 10^{-5} M or less (e.g., between 10^{-5} M and 10^{-6} M) in DMS-injected test animals, if it prevents DMS disruption or, in relation to an untreated or placebo control, if it: (i) reduces the mean volume of tissue occupied by the redistributed, transformed DMS protein material; (ii) reduces the ratio of the number of inflammatory cells per DMS; or (iii) increases the ratio of the number of macrophages containing disrupted DMS material per DMS.

As a further refinement, substances falling within this newly-defined category of pharmaceutically active agents - that is, the class of compounds that, at $\leq 10^{-5}$ M concentration levels in tissues, inhibit DMS disruption and/or transformation and hence, inhibit induced amyloid formation - can be tested, pursuant to the present invention, in a second *in vivo* assay. Particularly preferred for this purpose is the "senile animal" model for cerebral amyloidosis, where animals such as aged dogs or monkeys, which are known to develop variable numbers of Alzheimer-type cerebral senile plaques, see Wisniewski, *et al.*, *J. Neuropathol. & Exp. Neurol.* 32: 566 (1973); Selkoe, *et al.*, *Science* 235:873 (1987), are tested for amyloid inhibition. This *in vivo* assay involves initial pretreatment- and control-biopsy monitoring, to confirm and quantify the presence of senile plaque, and serial cerebral biopsy to monitor quantitatively the

evolution of DMS and senile plaque *in situ* and the presence (or absence) of amyloid-formation inhibition.

The method of the present invention for treating cerebral amyloidosis is used with subjects in whom amyloid formation is anticipated. The treatment can be applied, for example, to those who are at risk of developing cerebral amyloid, as in senile plaques, including the elderly, nondemented population and patients with the diagnoses listed above under the cerebral-amyloidosis rubric. In addition, such prophylactic therapy can be effected, pursuant to the present invention, to inhibit or prevent less severe forms of brain-function decline correlated with the formation of smaller amounts of disrupted DMS in elderly, nondemented subjects for whom there is not expectation of dementia associated with a diseases under the cerebral-amyloidosis rubric.

Compounds within the present invention can be administered to such subjects orally, rectally, via a nasal route, parenterally (including by skin or other routes), in spray or aerosol form, or via inhalation. A compound within the present invention can thus be administered in a pharmaceutically-acceptable carrier therefor, such as physiological saline solution.

Compounds of the present invention are particularly preferred that, in addition to possessing DMS disruption inhibiting activity in the aforementioned concentration range, also are nontoxic at an appropriate dosage level, have a satisfactory duration of effect, and display an adequate ability to cross the blood-brain barrier. In this regard, U.S. Pat. No. 4,540,564 discloses an approach for enhancing blood-brain barrier-penetrating ability by attaching a centrally acting drug species to a reduced, biooxidizable, lipoidal form of a dihydropyridine $\rightarrow$ pyridinium salt redox carrier. Also particularly preferred are compounds that have specific, selective binding affinity for DMS components. Skilled artisans recognize that any of the known methods of enhancing blood-brain barrier-penetrating ability can be used in the present invention, using the guidelines provided herein.

Determining a pharmaceutically-effective amount of a compound administered in accordance with the present invention entails standard evaluations of pharmacokinetic data and clinical efficacy. For instance, see GOODMAN AND GILMAN'S THE PHARMACOLOGICAL BASIS FOR THERAPEUTICS

(7th ed.). Thus, the above-described *in vivo* animal testing will provide the basis for a range of dosages and dosage schedules to be assessed clinically in humans. An assessment in this regard would generate pharmacokinetic data, for example, regarding bioavailability, absorption, metabolism, serum levels and excretion.

5 Such data would be evaluated against clinical data obtained concurrently from neurobehavioral testing, for example, memory testing and testing of cognitive function, and from clinical medical assessment. If a dosage halts progression of deterioration in clinical parameters for a symptomatic patient, *i.e.*, a subject diagnosed as suffering from cerebral amyloidosis, that dosage should
10 also have a prophylactic effect in the elderly, nondemented population. In addition, a pharmaceutical composition within the present invention could be employed to ameliorate or prevent a decline in brain function, associated with amyloid formation, that is less severe than dementia, for example, where the subject does not require supervision or nursing care.

15 Prophylactic therapy in the aforementioned population could be effected, pursuant to the present invention, for all persons of normal brain function who fall within a prescribed age group, for example, 65- or 70-to-75-years old. Alternatively, prophylactic therapy could be applied to nondemented persons of any age who, while displaying normal brain function, are identified via diagnostic
20 testing that reveals evidence of DMS disruption in the brain.

 Diagnostic testing of this sort can be conducted by assaying, immunologically or otherwise, for the presence of DMS components such as DMS membrane, DMS protein or fragments thereof in biological samples other than brain tissue, such as samples of serum, spinal fluid and other bodily fluids.
25 Testing can also be directed to detection in a subject of antibodies against one or more DMS components. In addition, prophylactic therapy according to the present invention can be administered to the nondemented population on the basis of other factors, suggesting a risk for dementia, which are revealed by radiological or diagnostic imaging, genetic testing, electroencephalography or
30 other means.

 The following test paradigms illustrate ways in which DMS material, as described above, can be employed routinely, according to the present invention,

in identifying anti-amyloidosis agents within the aforementioned class of compounds.

TEST 1: *In vitro* disruption of DMS in a human brain slice

Human brain postmortem samples of histological block size (usually 1-5cm x 1-5cm x 1-3mm) are removed, by sterile techniques, with the aid of sterile gloves, scalpel and forceps, and then are placed in sterile empty plastic containers, such as a Petri dish before extracted DMS are injected into each brain sample at room temperature. After a one-hour incubation at room temperature, the brain samples are immersed in histological fixative and processed from histology by techniques that are standard for optical microscopy. Controls, size of inoculum, preparations of slides and interpretation of results are described below.

TEST 2: Formation of disrupted DMS induced *in vivo* by injection of DMS into live tissue

Laboratory rodents are anesthetized and their brains immobilized by routine methods, and injections of homogeneous DMS are made into superficial cerebral cortex regions through sterile needles inserted through the skull and meninges. (Sham control injections of DMS negative material can be put into either the contralateral cortex or into separate animals.) The method of anesthesia, type of craniotomy, site of injections in the brain parenchyma, size of needle and syringe or other vehicle, wound closure technique, and numbers of animals used are not crucial to the test and will vary depending on the animal used. Thus, a small mouse may not need a skull flap whereas a larger mammal may need a burr hole; size of needles or vehicles may vary with animal brain size, etc. (see Example 1). The size of injection is elective; smaller injections are more difficult and costly to trace histologically (see below), but larger injections are more costly in terms of numbers of DMS used. An exemplary protocol is detailed in Example 1.

The animal is painlessly sacrificed about 30 minutes or more after injection. The exact time of sacrifice is elective; generally, a period of 1 to 24 hours is preferable, but the DMS transformation will persist and can be recognized at many time intervals. After sacrifice the brain is removed by

standard methods and immersion fixed in histological fixative. Perfusion fixation is not recommended because perfusion pressures will usually disrupt the injection cavity and render the results inaccessible.

5 According to standard methods, the brain is fixed in toto for several days (correspondingly longer for larger animal brains), sliced, embedded, cut, mounted and stained for histological study. A dissecting microscope is used to locate the injection site and accurately place it in the block, and sections are carefully inspected during microtomy to ensure that the injection site is in the section and not discarded during trimming. The mounted slides are stained with routine stains
10 such as hematoxylin-eosin, or Congo Red, according to standard methodology. The sections are examined with the optical microscope fitted with polarizing lenses.

Variations are possible by virtue of the fact that compounds can be administered *in vivo*, via injection, ingestion or other routes, before, after or
15 during the introduction of DMS, and concurrently with or separately from the DMS. In addition, therapeutic strategies other than those based on the action of a pharmacological agent can be studied in whole animals. By means of the foregoing tests, nontoxic compounds suitable for clinical testing in human beings can be identified, pursuant to the present invention, that reduce the number,
20 quantity and/or volume of disrupted DMS.

Other details of the present invention are further described by reference to the following illustrative examples.

EXAMPLE ONE

Two human brain postmortem samples of 1.5cm. x 1.5cm. x 2mm were
25 removed by sterile technique and placed in two sterile Petri dishes. Approximately 80,000 homogeneous extracted DMS were injected into each through a sterile 20 gauge stainless steel needle at 37°C. Acetylcholine 20 mm in physiological saline was added to the second injection as a possible test compound. After one hour's incubation both samples were immersed and
30 processed as in Test 1 above.

EXAMPLE TWO

This example illustrates the identification of compounds, via an *in vivo* assay, as effective in reducing the number, quantity and/or volume of disrupted DMS, and hence, as effective amyloid-formation inhibitors at physiologically-compatible concentrations.

IN VIVO ASSAY: Male Wistar rats of three-months age are anesthetized by ether inhalation. Their heads are immobilized by means of a stereo-tactic head brace. Bilateral parieto-occipital scalp incisions (1 cm) are made with a sterile scalpel blade. Bilateral parieto-occipital 0.5 mm burr holes are made with a 0.5 mm drill.

For each compound to be assayed (see below), six rats are each injected on one side, through the burr hole from a sterile 1 cc syringe fitted with a sterile 22 gauge needle, with sterile physiological saline containing about 400,000 human DMS and the compound (total volume: 100 μ l). The injection is made in to the cerebral cortex to a depth of a few millimeters, so that the injection is within the parenchyma, not on the surface or in the ventricles. Consequently, the in-tissue concentration of the compound at the site of injection corresponds to the concentration of the compound in the saline.

On the contralateral side, each of the six test rats also receive an injection of sterile physiological saline (100 μ l). In addition to this internal control, a control group of six rats that do not receive any injection is associated with each test group. As an additional control group, a group of six rats that receive an injection of DMS without the compound, or that receive an injection of DMS and a compound found inactive using the testing protocols described herein, is associated with each test group.

After injection of the test animals, a sterile suture is placed through the scalp incision to cover the wound, and the animals are observed. Injection can be intracerebral, or via in-tissue injection at other places on the test animal. At post-injection intervals of one hour, twelve hours and twenty-four hours, respectively, four animals (two from the test group and two controls) are painlessly sacrificed by ether inhalation and CO₂ insufflation. Their brains are removed and fixed via immersion for twenty-four hours in 10% formalin. The fixed tissue is then sliced coronally, in sections of between 0.5 mm and 1 mm in

thickness, and the areas of injection are dissected out and blocked as described above under "Test 2."

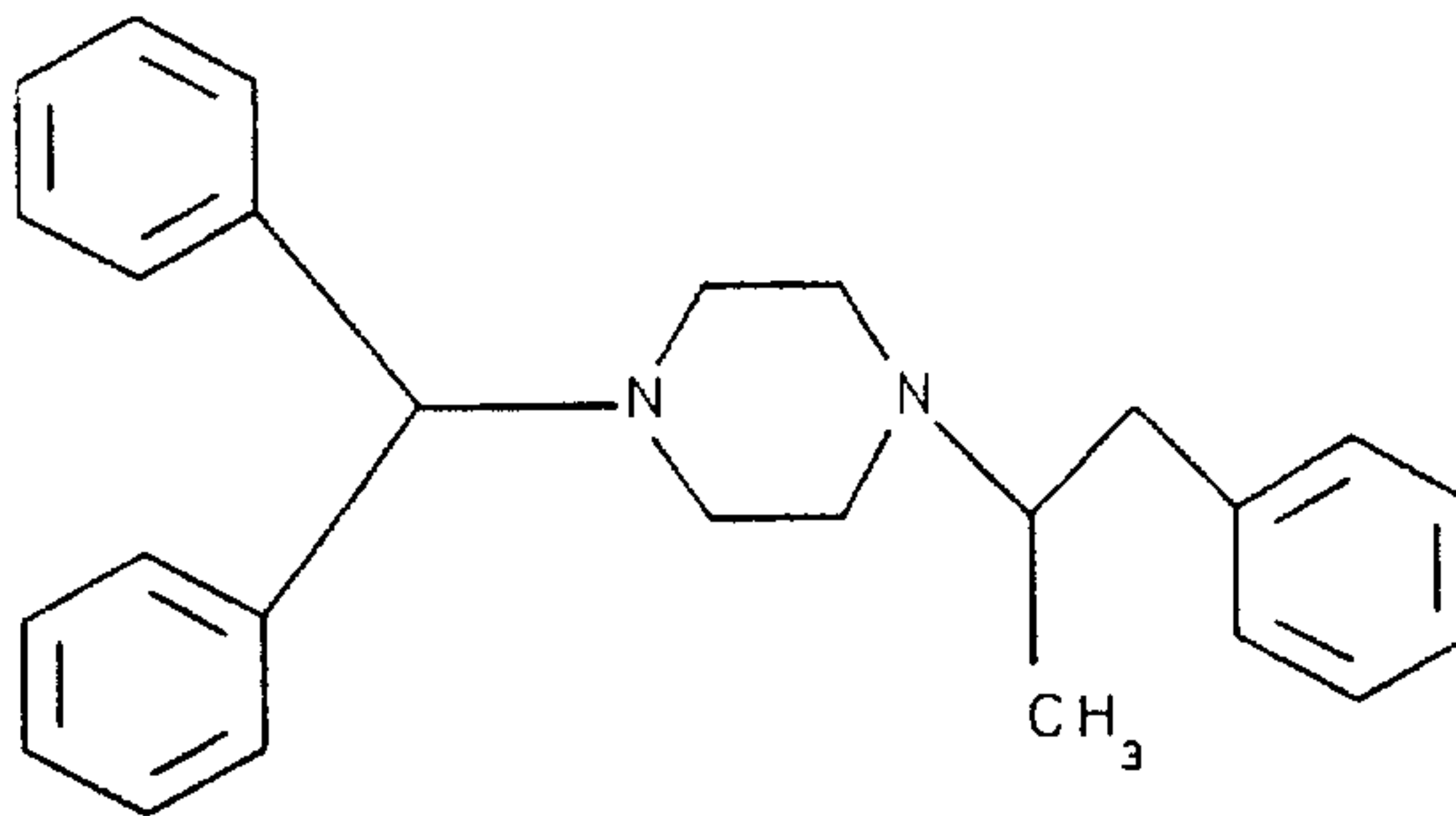
5 The blocks with the injection site are sectioned at a thickness of 6 μm , and every tenth section is mounted to provide a total of ten technically intact sections containing the injection site. The mounted sections are processed and stained with Congo Red, as previously described, and examined, via optical microscopy, with and without polarized illumination. By means of optical micrometer and standard graticles, mean volumes of DMS is determined and compared to the controls. Compounds that were found to reduce the number, quantity and/or volume of
10 disrupted DMS, when compared to the controls, were selected according to the present invention. Those compounds that did not reduce the number, quantity and/or volume of disrupted DMS, or that did not significantly reduce the number, quantity and/or volume of disrupted DMS, when compared to the controls, were considered inactive. Some of these inactive compounds were used in subsequent
15 experiments as controls.

EXAMPLE 3

Approximately 500 compounds were tested as described in example 2 above. Those compounds that were found to reduce the number, quantity and/or volume of disrupted DMS by at least 10%, when compared to the controls, were
20 considered useful in the present invention. The compounds found to be useful were found to fall within six (6) general formula, described below in Examples 4 and 5, whereas those compounds that did not reduce the number, quantity and/or volume of disrupted DMS by at least 10%, when compared to the controls, did not fall within the six (6) general formula.

EXAMPLE 4

25 This example illustrates the synthesis of various compounds used in Examples 1 and 2 above, which were found to be effective.

1. Synthesis of compounds of formula (I)*Preparation of ($\pm$)-1-(1-Phenylisopropyl)-4-(diphenylmethyl)piperazine*Compound 1

The general pathway for making compounds of Formula (I) is outlined above. In accordance with the general pathway, approximately 17 g of 0.1 mol 3-Chlorophenylacetic acid was refluxed in acetic anhydride (75 ml, 0.8 mol) and sodium acetate (8.2 g., 0.1 mol) for 2 days. The resulting mixture was allowed to cool to room temperature and diluted with water (150 ml). The organic layer was then refluxed in concentrated HCl (80 ml) for one hour, cooled to room temperature and then 200 ml of water was added to the mixture. The mixture was extracted with ethyl ether to produce 2-chlorophenyl acetone at about a 41% yield. The intermediate product had a boiling point of about 76°C.

To a solution of 3-chlorophenyl acetone (8.1 g, 48 mmol) in ethanol (50 ml) was added NaBH₄ (1.5 g, 40 mmoles). The mixture was stirred at room temperature overnight and quenched with water, extracted with ether to produce about 88% of the desired alcohol. 3-(Trifluoromethyl)phenyl-1-propanol was obtained in about 72% yield using this procedure. A solution of the alcohol (0.1 mol) and PBr₃ in 100 ml of chloroform then was stirred at room temperature overnight, and poured into water (100 ml). The organic phase was washed with water (2 × 10 ml), dried and evaporated to produce the corresponding bromide. In accordance with this method, 2-bromo-1-phenylpropanes were prepared; 2-Bromo-1-(3-chlorophenyl)propane was obtained as a colorless oil with a yield of about 51%, and 2-Bromo-1-(3-trifluoromethyl)phenylpropane was obtained as a colorless oil with a yield of about 42%.

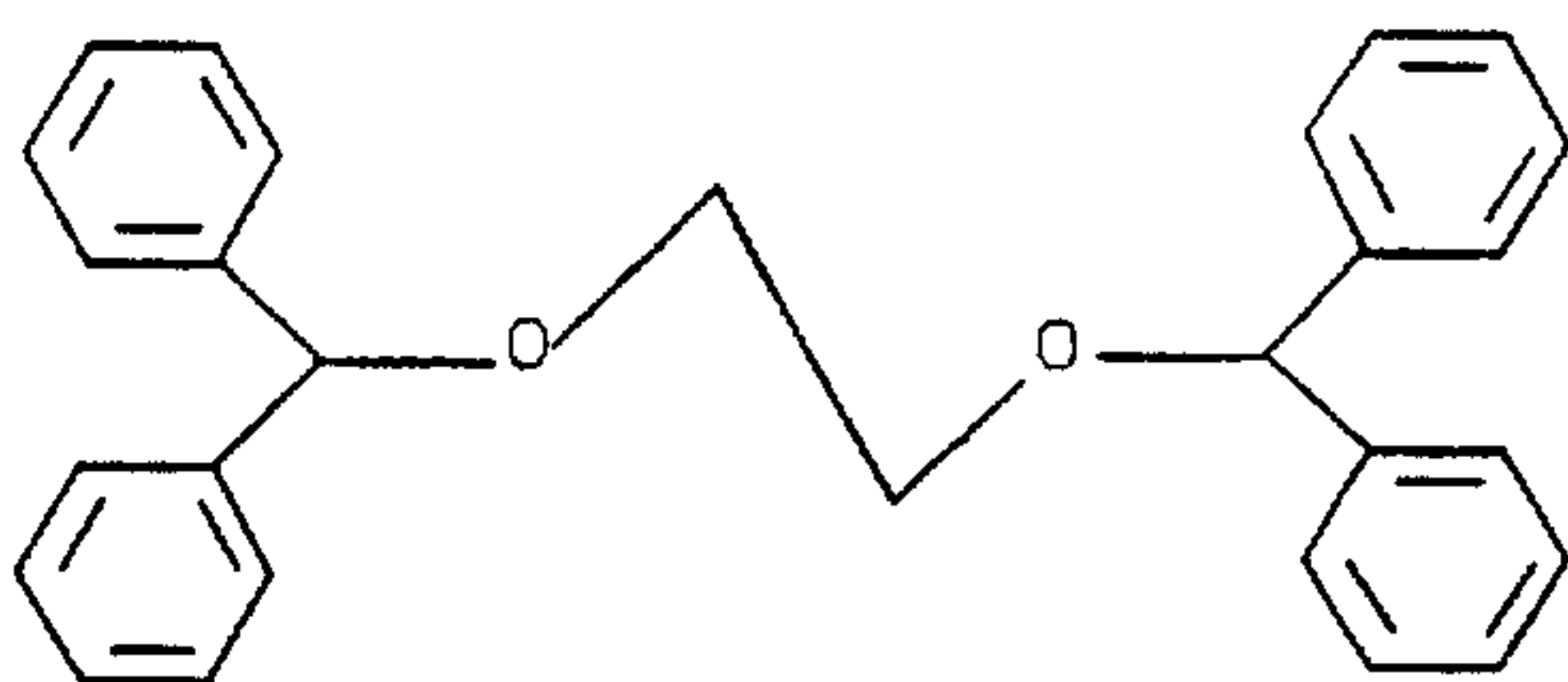
These products were alkylated to produce the compound 1 above. A solution of 1-(diphenyl)methylpiperazine (0.1 mol), 2-bromo-1-phenylpropane (commercially available, 0.11 mol) and triethylamine (0.2 ml) in 20 ml DMF was

refluxed for about 4 hours. The mixture was evaporated to dryness in vacuum. The residue was passed through silica gel column to produce compound 1 as a colorless liquid, which then was converted to its hydrochloride salt having a melting point within the range of 244-246°C. The final product (hydrochloride salt thereof) was a white solid having a molecular weight of about 435.9.

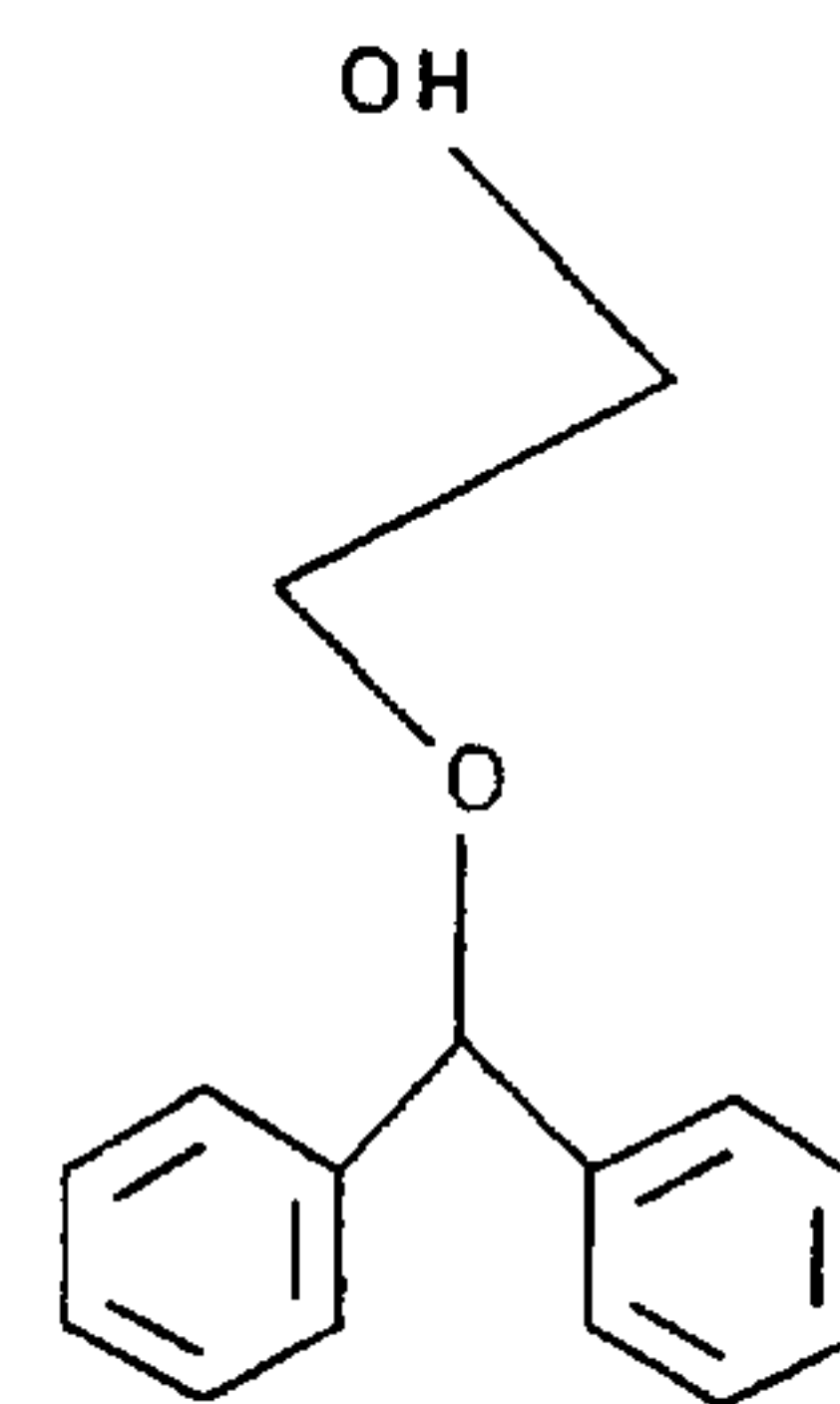
2. Synthesis of compounds of formula (II)

Preparation of 1,2-Bis(diphenylmethoxy)ethane and 2-Diphenylmethoxyethanol

Compound 2; 1,2-Bis(diphenylmethoxy)ethane was prepared in accordance with the general pathway outlined above for preparation of compounds of Formula (II). Compound 3; 2-Diphenylmethoxyethanol is a by-product of that pathway synthesis, and can be used in other pathways to produce other compounds useful in the present invention.



Compound 2



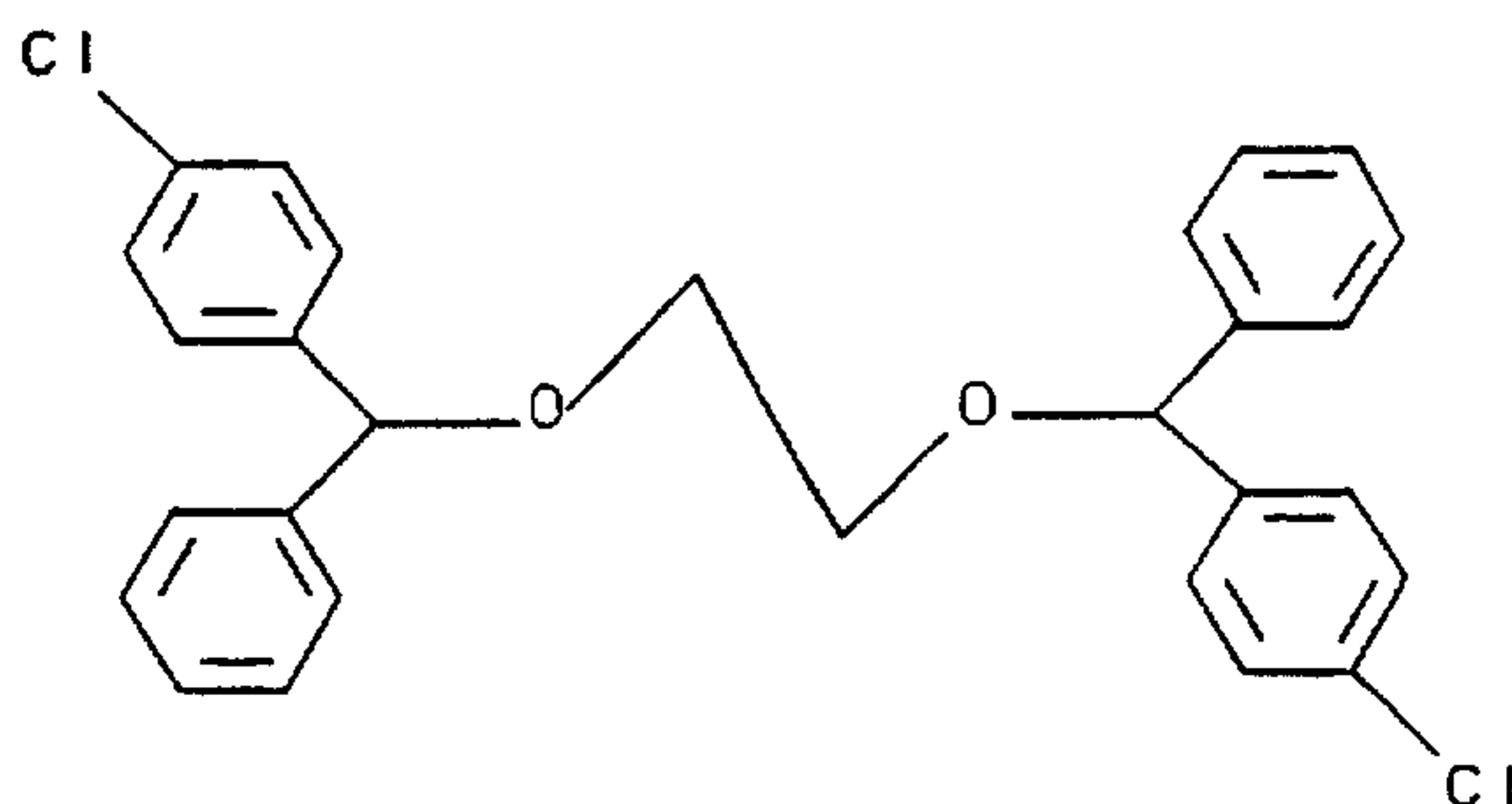
Compound 3

In accordance with the pathway synthesis for making compounds of Formula (II), benzhydryl bromide (19 g, 0.077 mol) and ethylene glycol (27.8 g in 25 ml; 0.45 mol) were combined and stirred. Nitrogen was bubbled through the reaction. The mixture was warmed on a steam bath. As the mixture warmed, the benzhydryl bromide melted and formed a layer denser than the ethylene glycol. As the reaction proceeded, the layer became less dense than ethylene glycol. The reaction appeared to be complete after 10 minutes and TLC (methylene chloride) confirmed that the reaction was complete. The diether, compound 2, had an R_f of 0.6. Benzhydryl bromide reacted almost instantly with alcohols.

The reaction then was cooled to room temperature and then was diluted with 100 ml of water and extracted with 2 X 50 ml of methylene chloride. The methylene chloride solution was filtered through IPS filter paper and stripped. The resulting material (14 g) was then flash column chromatographed on 150 g of flash chromatography silica gel. After crystallization from hexane, compound 2 formed a white crystalline solid having a melting point within the range of from about 99-103°C, and a molecular weight of 394.5, and compound 3 formed a white crystalline solid having a melting point within the range of from about 68-70°C and a molecular weight of about 228.3.

10 *Preparation of 1,2-Bis(4-chlorophenylphenylmethoxy)ethane*

The general pathway for producing compounds such as compound 2, with various substituents X (X can be CH₃, OCH₃, CF₃, Cl, Br and other halogens, C1-C5 alkyl, etc.) is outlined above. Compound 4 below, where X is Cl was prepared in accordance with the aforementioned general pathway.



15

Compound 4

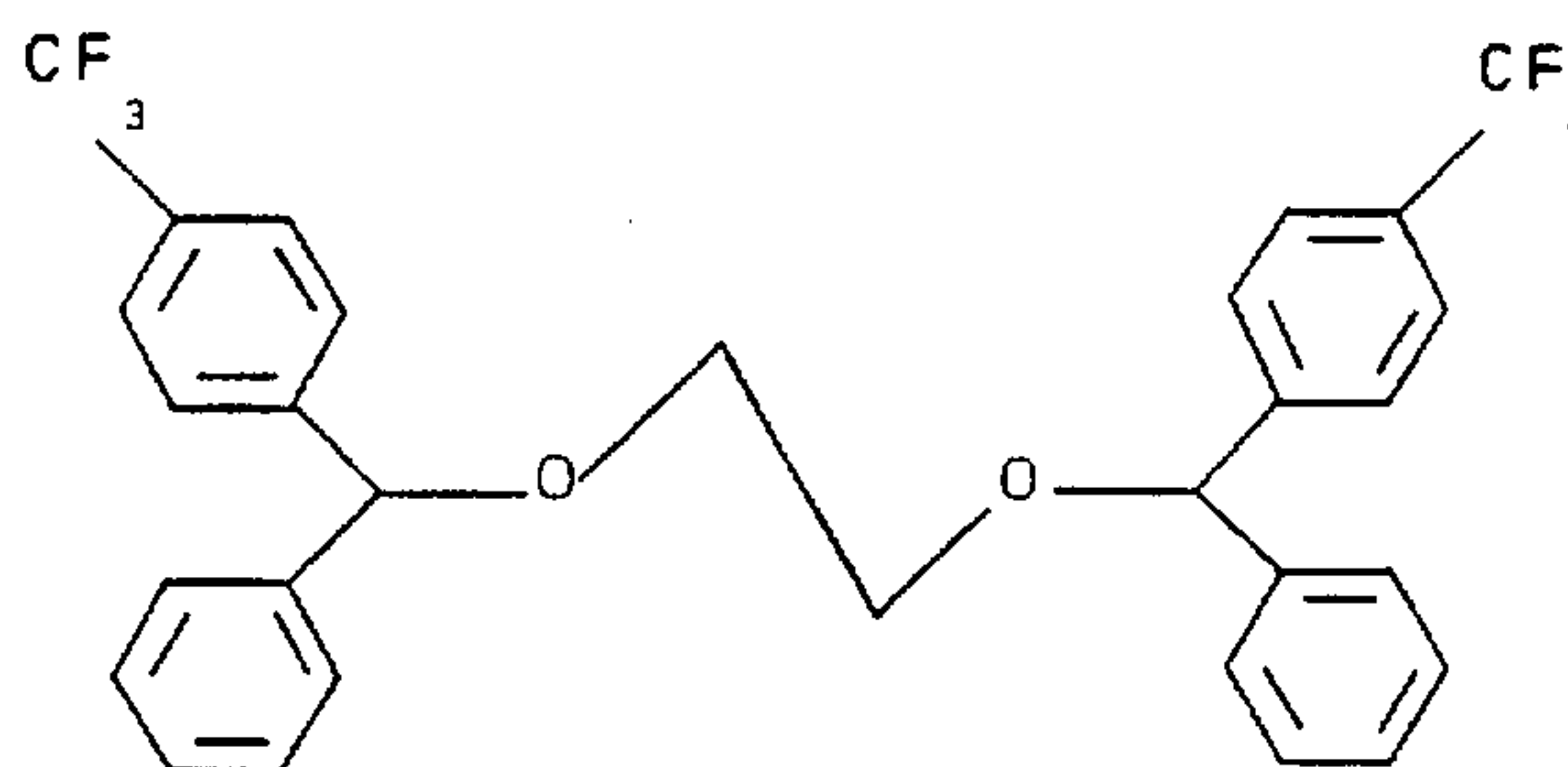
In accordance with the pathway synthesis for making compounds of Formula (II), benzhydrol (8.7 g, 0.044 mol) and ethylene glycol (33 g in 30 ml; 0.53 mol) were combined and warmed on a steam bath. Once the solution was complete, methane sulfonic acid (2 ml, 0.03 mol) was added and the reaction mixture immediately became cloudy. The TLC (methylene chloride) indicated that the reaction was complete on mixing. Compound 4 had an R_f of 0.7. The reaction then was cooled to room temperature and diluted with 100 ml of water and extracted with 2 X 50 ml of methylene chloride. The methylene chloride solution was filtered through IPS filter paper and stripped.

20

The resulting material then was flash chromatographed on 100 g of flash chromatography silica gel. Compound 4 was crystallized from hexane and formed a liquid at room temperature, and had a molecular weight of 463.4.

Preparation of 1,2-Bis(4-trifluoromethylphenylphenylmethoxy)ethane

- 5 A procedure identical to that above was used only X for compound 5 below was CF_3 instead of Cl.

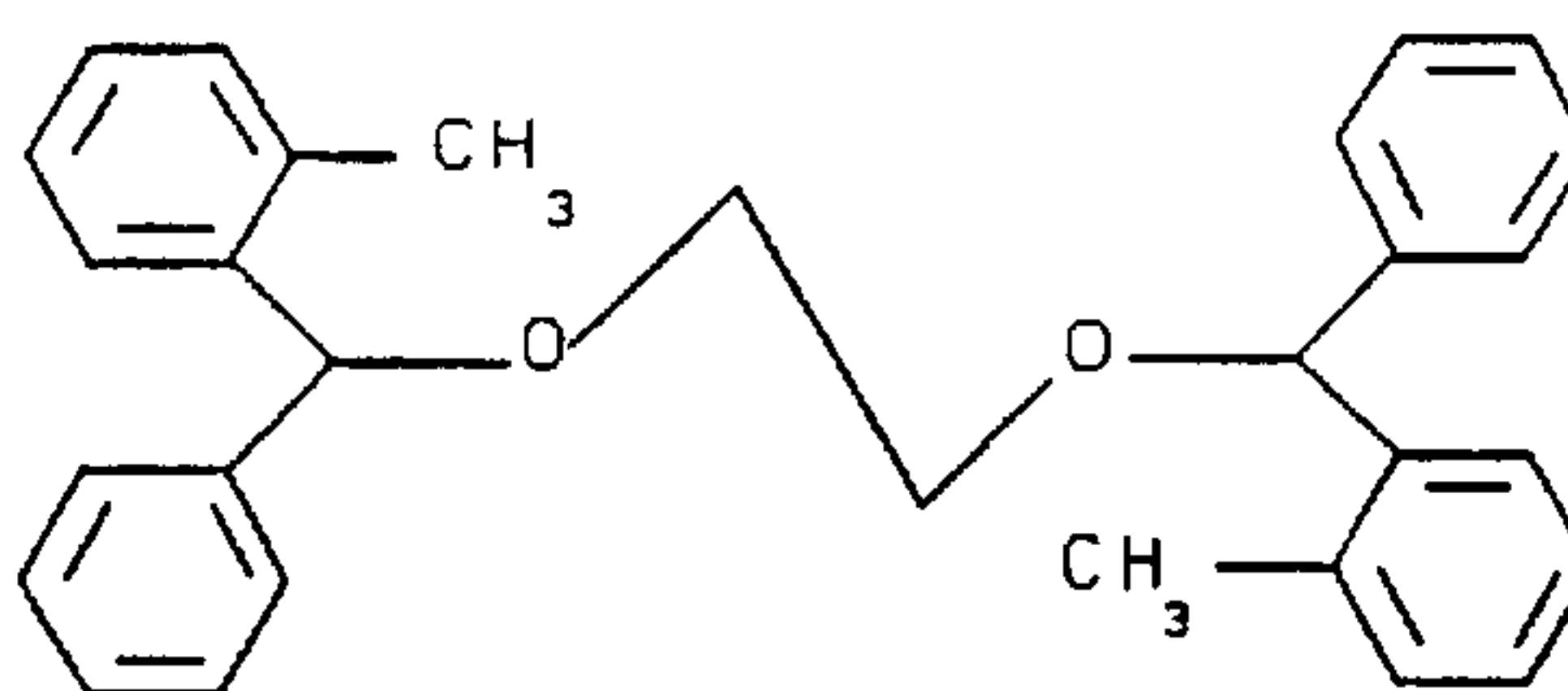


Compound 5

Compound 5, like compound 4 above, was crystallized from hexane and formed a liquid at room temperature and had a molecular weight of 530.5.

- 10 *Preparation of 1,2-Bis(2-methylphenyl phenylmethoxy)ethane*

A procedure identical to that above was used only X for compound 6 below was CH_3 instead of Cl or CF_3 .

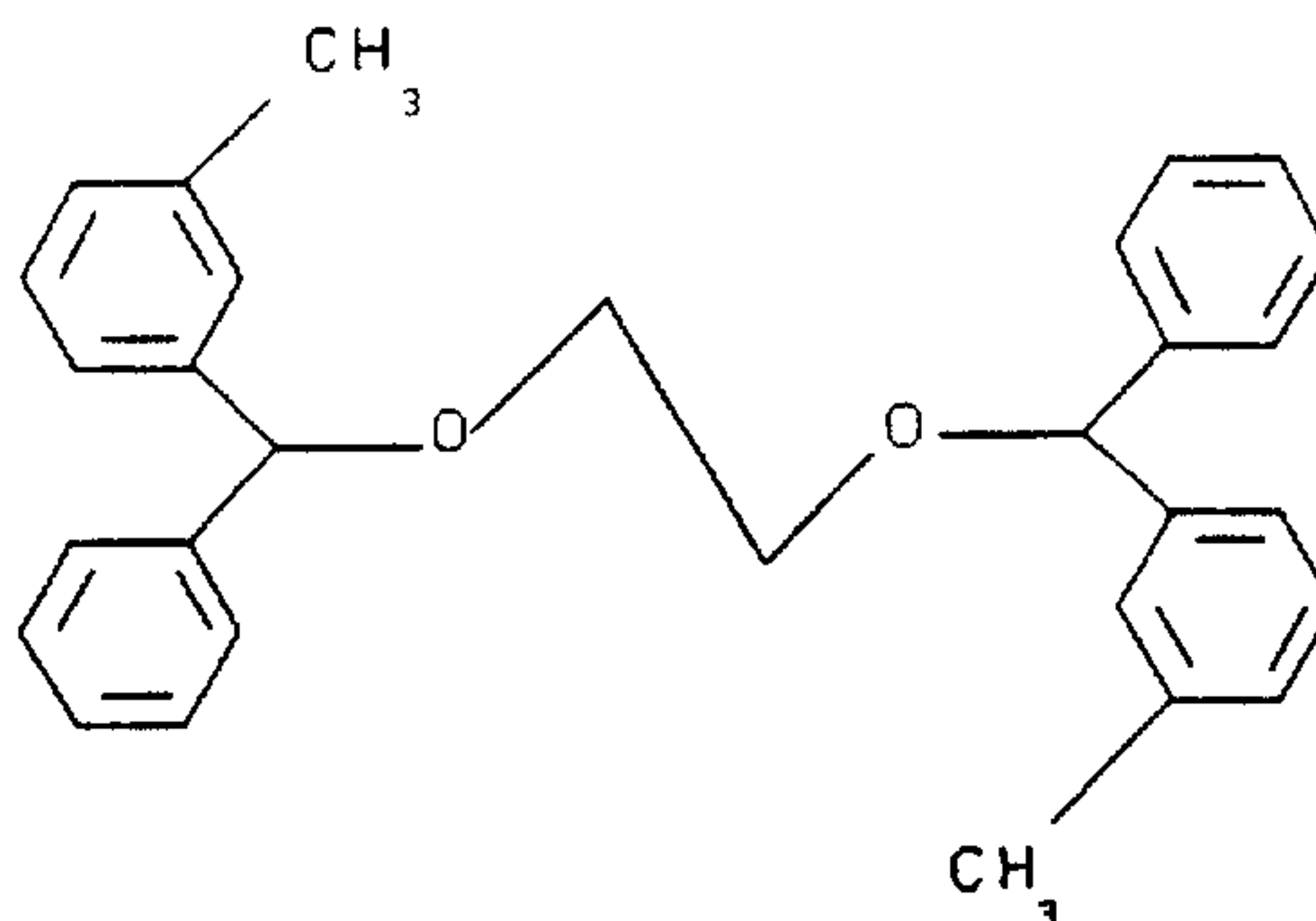


Compound 6

- 15 Compound 6, like compounds 4 and 5 above, was crystallized from hexane and formed a solid crystalline material having a melting point within the range of from about 105-109°C, and had a molecular weight of 422.6.

Preparation of 1,2-Bis(3-methylphenyl phenylmethoxy)ethane

A procedure substantially like that above was used; only X for compound 7 below, however, was CH_3 instead of Cl or CF_3 .

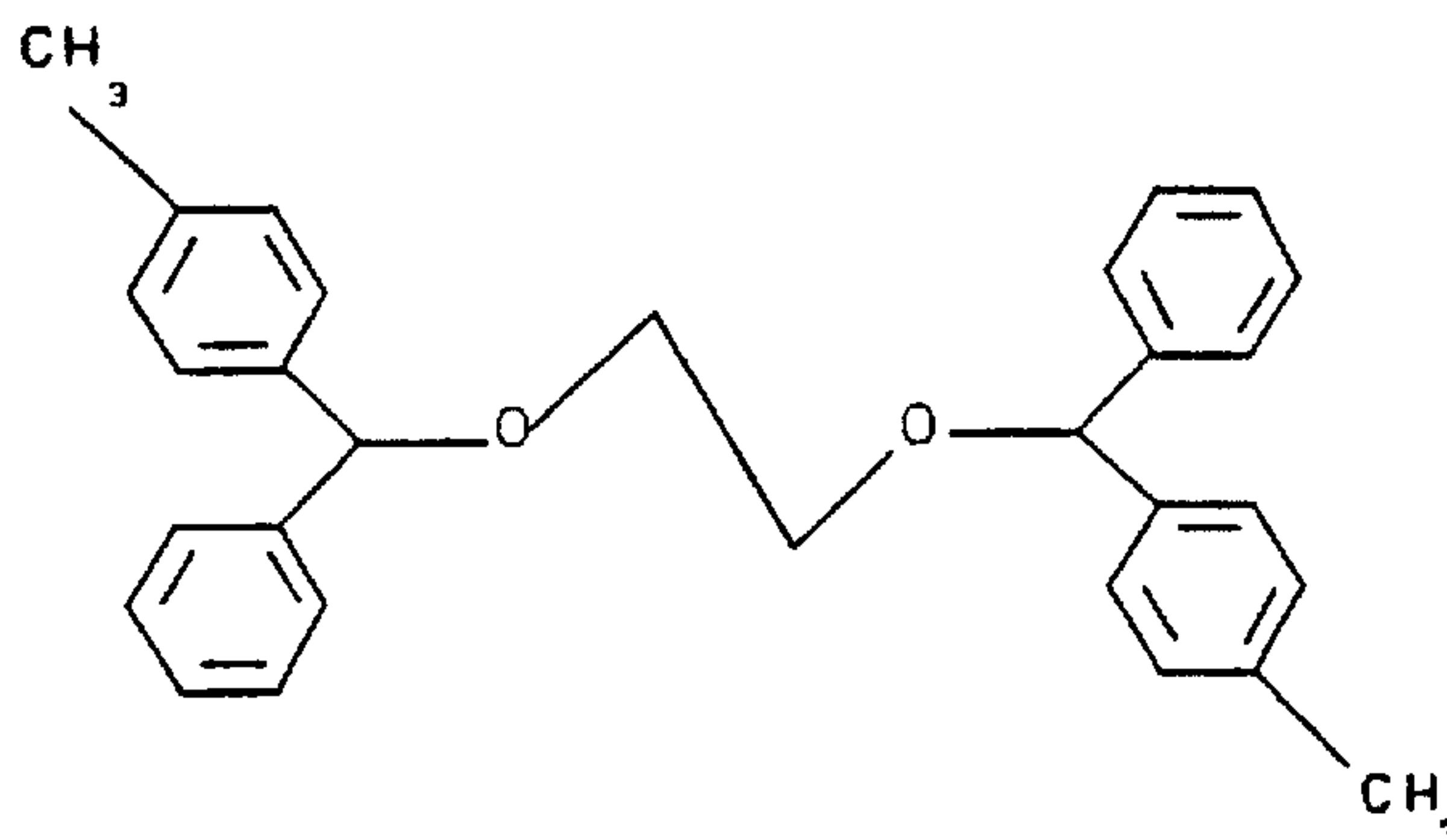


Compound 7

Compound 7, like compounds 4-6 above, was crystallized from hexane and formed a liquid oily substance having a molecular weight of 422.6.

Preparation of 1,2-Bis(4-methylphenyl phenylmethoxy)ethane

5 A procedure identical to that above was used only X for compound 8 below was CH₃ instead of Cl or CF₃.



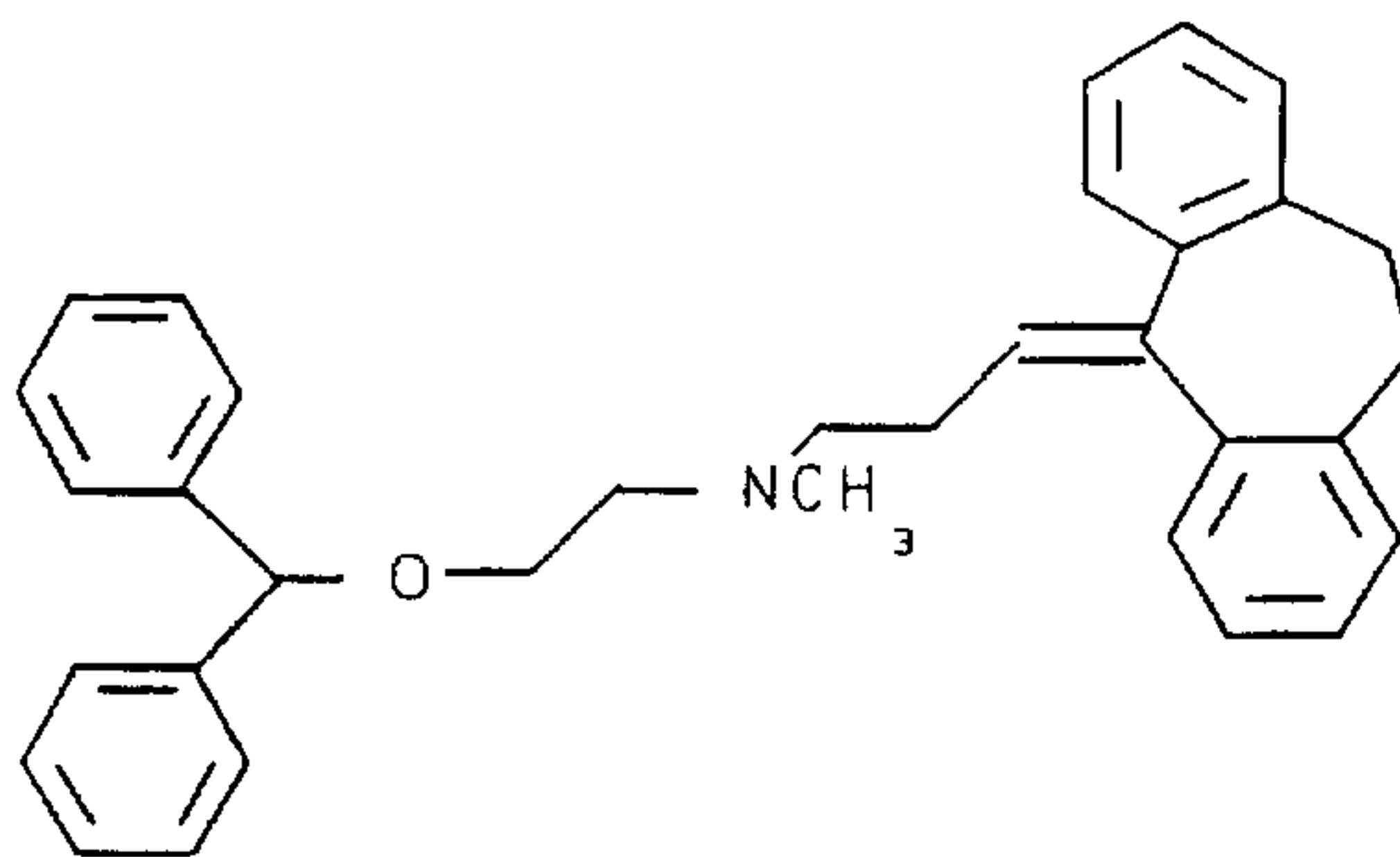
Compound 8

10 Compound 8, like compounds 4-7 above, was crystallized from hexane and formed a white crystalline solid material having a melting point in the range of from about 107-116°C, and had a molecular weight of 422.6.

3. Synthesis of compounds of formula (III)

Preparation of 3-(10,11-Dihydro-5H-dibenzo[a,d]cyclohepten-5-ylidene-N-methyl-N-[2-(diphenylmethoxy)ethyl]-1-propanamine

15 The general pathway for synthesizing compounds of formula (III) is outlined above. Compound 9 below was prepared in accordance with that general pathway.

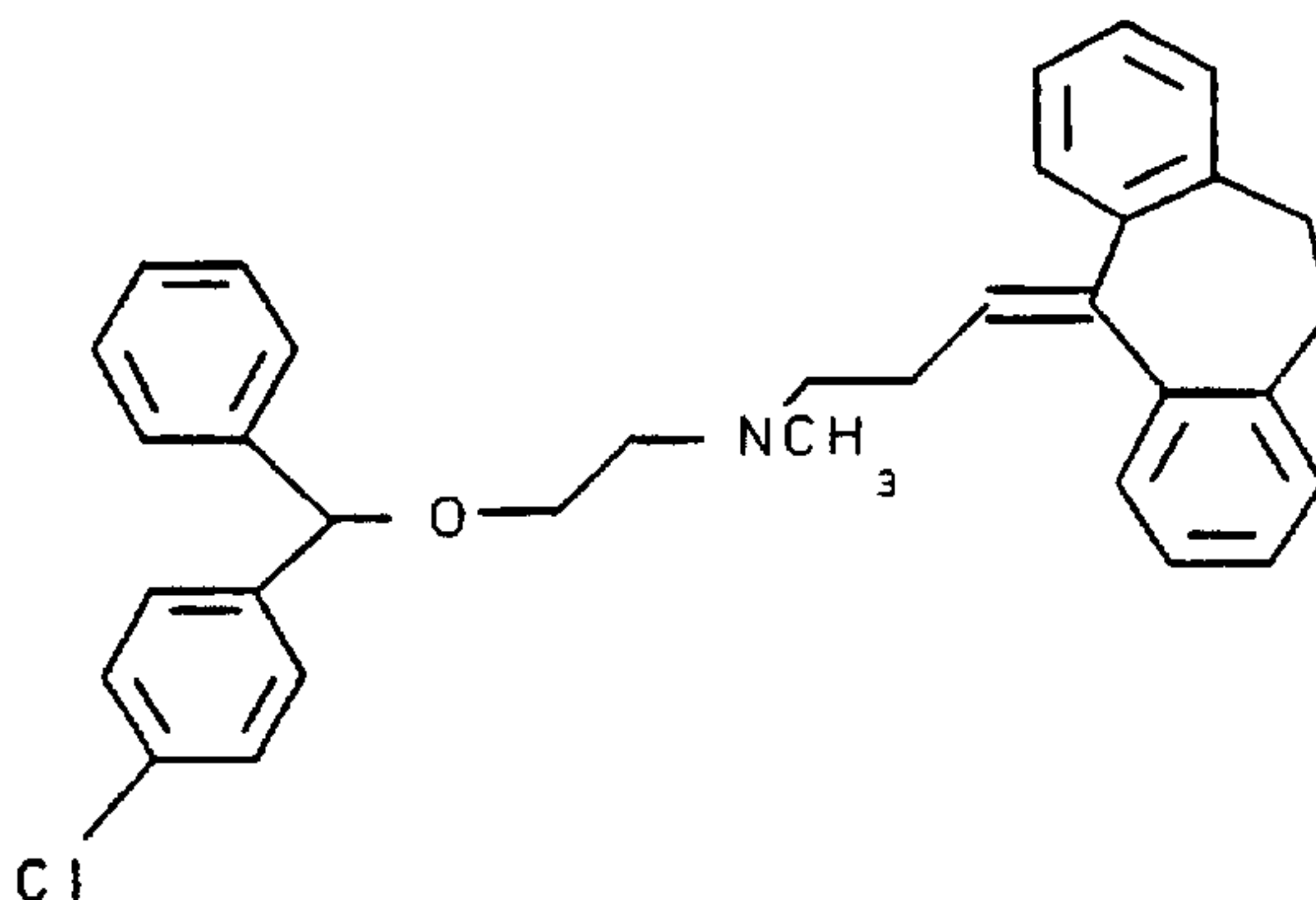


Compound 9

Compound 9 was prepared by first preparing compound 3 above, 2-diphenylmethoxyethanol, (0.54 g, 0.003 mol) and then treating equimolar amounts (0.045 mol) with tosyl chloride (0.6 g, 0.045 mol) in 30 ml dichloromethane and 15 ml pyridine, and stirred overnight at room temperature. The mixture was extracted with dichloromethane and was added to a methylene chloride solution of 3-(10,11-Dihydro-5H-dibenzo[a,d]cyclohepten-5-ylidene-N-methyl)-1-propanamine (nortryptylene; 0.79 g, 0.002 mol). This mixture then was stirred at room temperature for 24 hours and then neutralized and purified by chromatography to produce compound 9, which was subsequently formed into its fumaric acid salt. The salt of compound was a tan powder having a molecular weight of about 589.7, whereby the molecular weight of compound 9 was 473.7.

Preparation of 3-(10,11-Dihydro-5H-dibenzo[a,d]cyclohepten-5-ylidene-N-methyl-N-[2-(4-chlorophenyl phenylmethoxy)ethyl])-1-propaneamine

A procedure identical to that above was used, except that, for compound 10, one of the diphenyl moieties in compound 9 above was substituted with a chlorine.



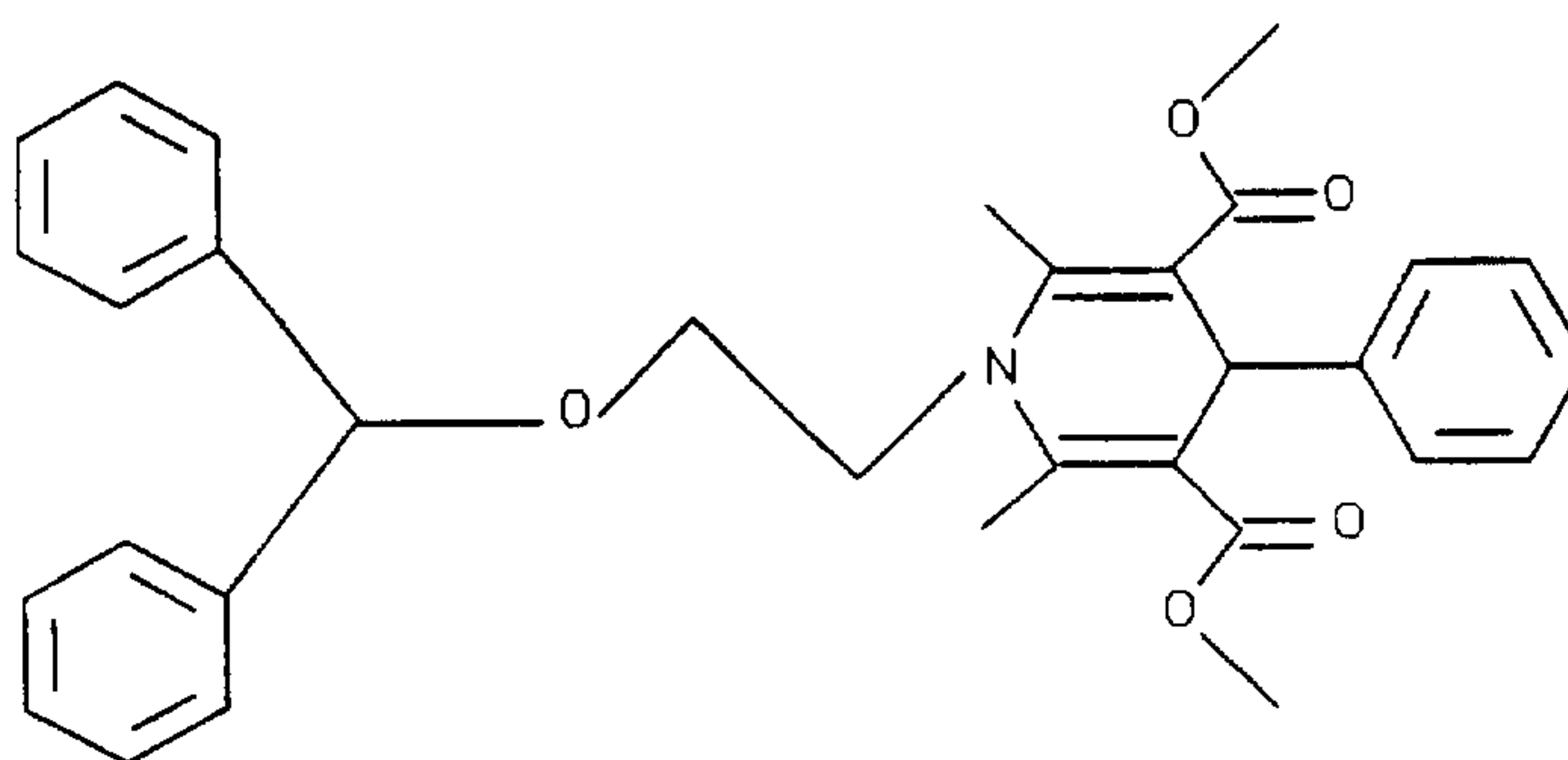
Compound 10

Compound 10, like compound 9 above, was formed into its fumaric acid salt which produced a hygroscopic, light-sensitive tan foam product having a molecular weight of 624.2, whereby compound 10 had a molecular weight of 508.1.

5

4. Synthesis of compounds of formula (IV)

Compound 11 below was prepared in accordance with the general pathway outlined above for preparing compounds of Formula (IV).



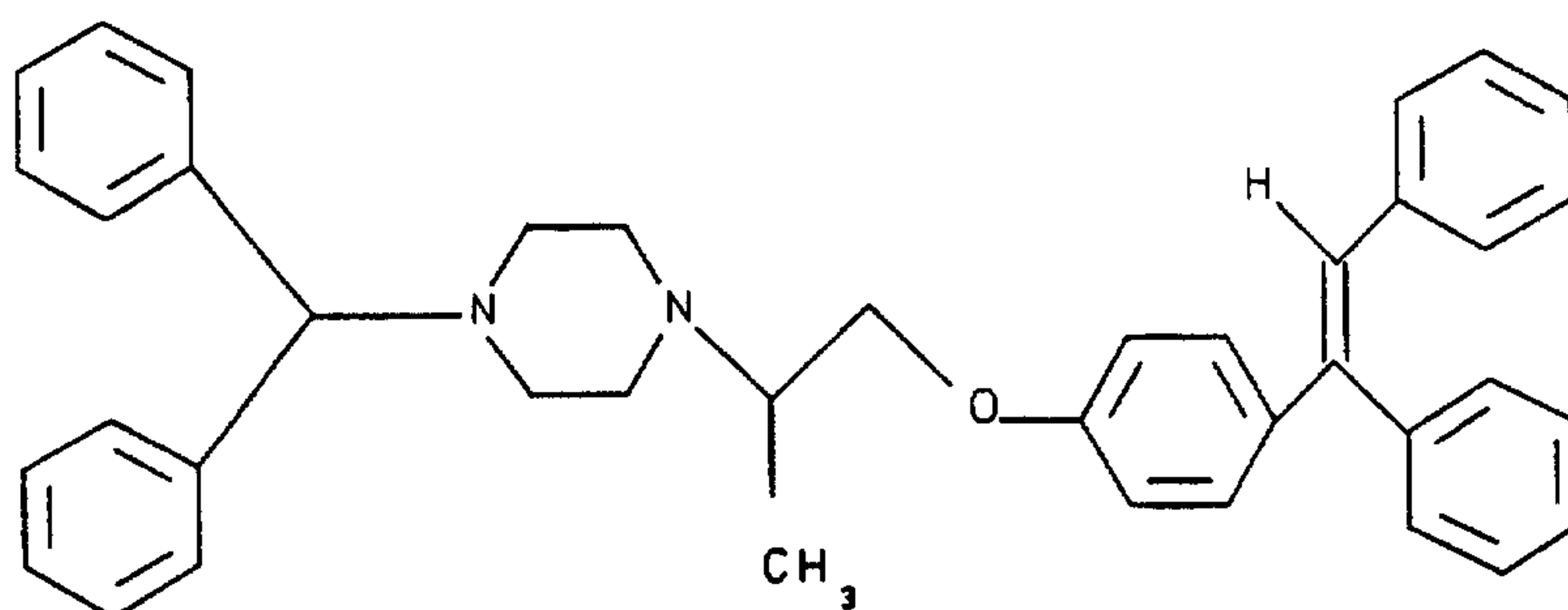
Compound 11

5. Synthesis of compounds of formula (V)

10

Preparation of 2-[4-(1,2-Diphenylethynyl)phenoxy]-diphenylmethylniprazine

Compound 12 below was prepared in accordance with the general pathway outlined above for preparing compounds of Formula (V):



Compound 12

15

Compound 12 was prepared in accordance with the pathway synthesis described above for preparing compounds of general formula (V). In accordance with this synthesis pathway. About 98 ml or a 0.2M solution of the grignard reagent, benzyl magnesium chloride in THF (0.196 mol), was added to a 1000

ml flask equipped with thermometer and dropping funnel, whereby the flask was charged with about 38 grams of 4-methoxybenzophenone (0.179 mole in 400 ml THF). The grignard reagent was rapidly added while maintaining the temperature below 20°C with an ice bath under a nitrogen atmosphere. The ice bath then was removed and the reactants heated to 40°C, was stirred for about 1 hour, poured into about 1L of cold 10% HCl and extracted with about 350 ml of ethyl acetate. This produced a yello-green oil, after concentration *in vacuo*, which subsequently was treated with 2L of boiling hexanes and the boiling solution was filtered through a plug of glass wool. The solution then was concentrated to a volume of about 700 ml and set aside to cool. After cooling, the solution was filtered, washed and air dried to yield a crystalline material.

Approximately 26 g of the crystalline material was mixed in a 500 ml flask with about 26 g of boric acid and slowly heated to 250°C under nitrogen. The temperature was maintained for 10 minutes, the reactants then were allowed to cool to room temperature and 350 ml of water was added. After boiling to dissolve any inorganics, the mixture was cooled to room temperature, extracted with toluene and washed twice with water. The intermediate product was filtered through a plug of silica to yield 18.5 g of pure material after combining fractions and concentrating.

About 12 grams of the intermediate product (0.042 mol) was mixed with about 51 g of NaCN and 75 ml of DMSO and magnetically stirred while heating at 160-170°C under nitrogen for about 6 hours. Heating was discontinued overnight, and resumed for 8 hours the remaining day. Again, heating was discontinued overnight, and resumed for 4 hours the remaining day. After heating, the reaction mixture was diluted with 1.5L of water and transferred to a 2L separation funnel. The mixture was acidified to pH 2 with concentrated HCl and extracted with 3 X 300 ml toluene. The combined toluene extract was back extracted twice with 300 ml of water, dried over sodium sulfate, nitrogen bubbled through the solution to displace HCN, filtered and concentrated *in vacuo* to yield about 12.06 g of a dark brown phenolic material.

About 2 grams of the dark brown phenolic material (7.34 mmol) was charged with 20 ml THF to a pressure vessel and about 4.0 ml of 2.5 mmolar n-BuLi (10 mmol) were added under a blanket of nitrogen. The vessel was sealed

and cooled to -78°C , the top removed and about 3 g of propylene oxide (51.65 mmol) was added. The vessel then was sealed with parafilm and allowed to warm to room temperature. Upon reaching room temperature, the vessel was sealed with a Teflon top and placed in a water bath at $55-60^{\circ}\text{C}$. After 5 days, the temperature was increased to 80°C and maintained there for 14 days.

After 14 days at 80°C , the reaction (now about 75% complete) was diluted with water, acidified with concentrated HCl and extracted 3 times with ethyl acetate. The combined extracts were back extracted twice with water, the organic layer dried, filtered and concentrated to yield 3.14 g of a crude reaction mixture. the mixture then was dissolved in dichloromethane and introduced onto a 5 X 12 cm column of silica and eluted first with EtAc/hexane 10/90 and then 15/85. Unreacted starting material eluted quickly and the products were eluted as an elongated spot by TLC. Multiple elutions with 7/93 EtAc/hexane resolved the mixture into two component mixtures, each containing E and Z isomers. After a series of acetylation and elutions, fractions containing about 1.17 g of a secondary alcohol product was obtained.

This secondary alcohol (910 mg, 2.75 mmol) was mixed together with 760 mg of Tosyl chloride (4.0 mmol) and 5 ml of pyridine, and magnetically stirred overnight in a 25 ml flask. An additional 320 mg of tosyl chloride then was added after 25 hours and stirring was continued for an additional 24 hours. Upon completion of the reaction, the solution was diluted to 500 ml with water, extracted with 200 ml of ethyl acetate and the ethyl acetate washed twice with water, once with 10% HCl and twice with water. The ethyl acetate was concentrated *in vacuo* to yield about 1.35 g of a syrup, which subsequently was introduced onto a 25 X 500 mm MPLC column dissolved in dichloromethane and the column eluted with EtAc/hexanes 10/90. About 1.21 grams of a tosylated product were obtained.

About 905 mg of the tosylated product in 20 ml of o-xylene was added to a 100 ml 3 necked flask. Meanwhile, a 50 ml flask as charged with about 1420 mg of diphenylmethyldipiperazine (5.6 mmol) and 2.24 ml of 2.5 mmolar n-BuLi (5.6 mmol), both in 20 ml of o-xylene, was added under nitrogen at room temperature. The solution was maintained for 30 minutes at room temperature and then added to the 3 necked flask containing the tosylated product. The

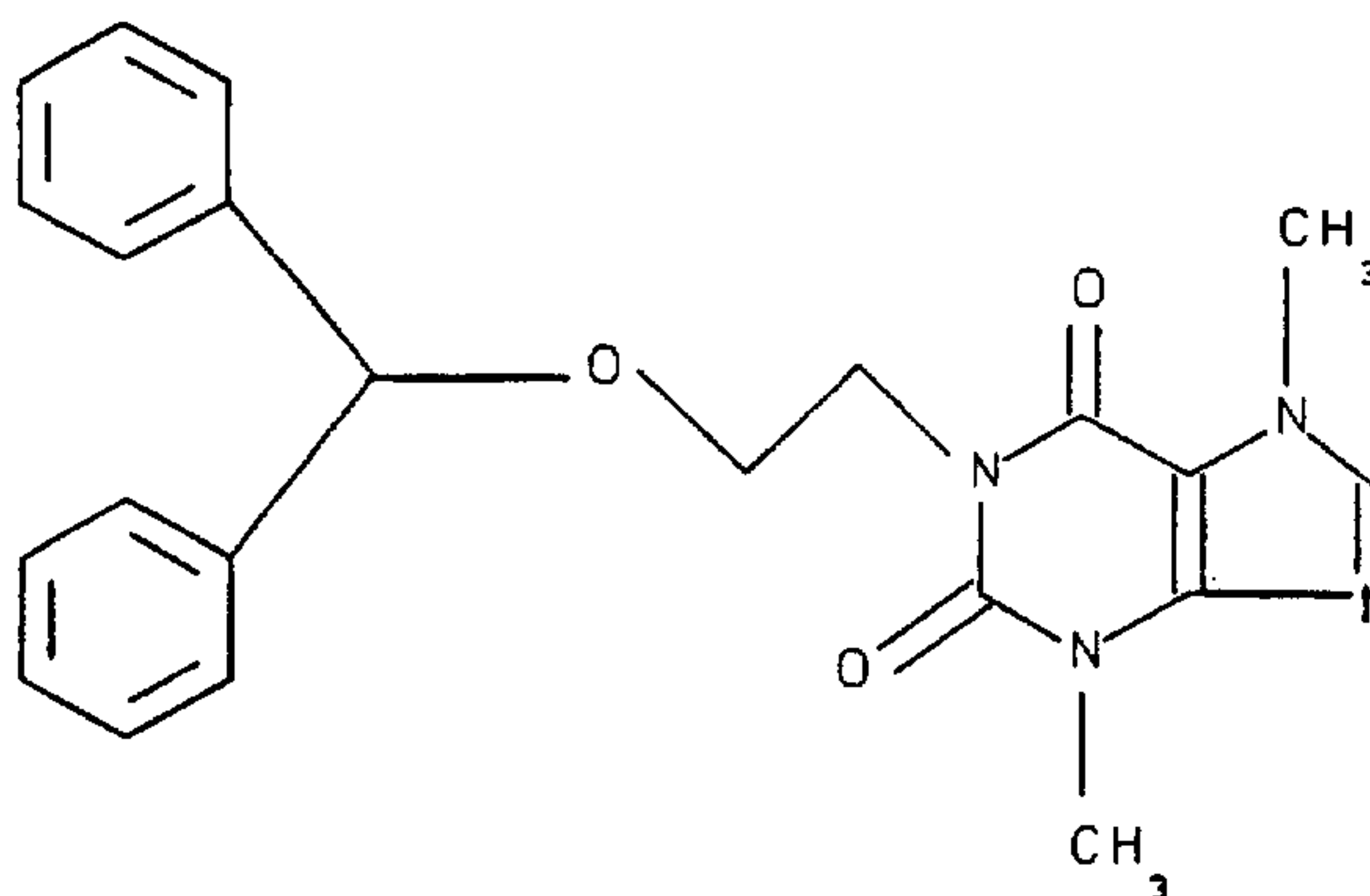
reaction was refluxed for 4 hours, the heat was removed and stirred overnight at room temperature. Refluxing was continued an additional hour.

The reaction was cooled to room temperature, transferred to a 125 ml separation funnel, the flask rinsed with toluene and combined with the original solution and extracted three times with 2ml portions of water. The o-xylene/toluene solution was dried over sodium sulfate, the solution filtered and concentrated *in vacuo* to yield 2 grams of a thick red syrup. The syrup then was dissolved in dichloromethane and filtered through a 5 X 10 cm plug of silica gel eluting with EtAc/hexanes 30/70. Fractions were combined and concentrated to yield about 326 mg of a red syrup. This syrup then was introduced onto a 15 X 500 mm MPLC column in dichloromethane and the column was eluted with EtAc/hexanes 20/80. Fractions again were combined and concentrated to yield about 200 mg of compound 12 which was present as a tan to brown syrup.

6. Synthesis of compounds of formula (VI)

Preparation of 3,7-Dimethyl-5-[2-(diphenylmethoxy)-ethyl]xanthine

Compound 13 below was prepared using the general pathway synthesis above for preparing compounds of Formula (VI).



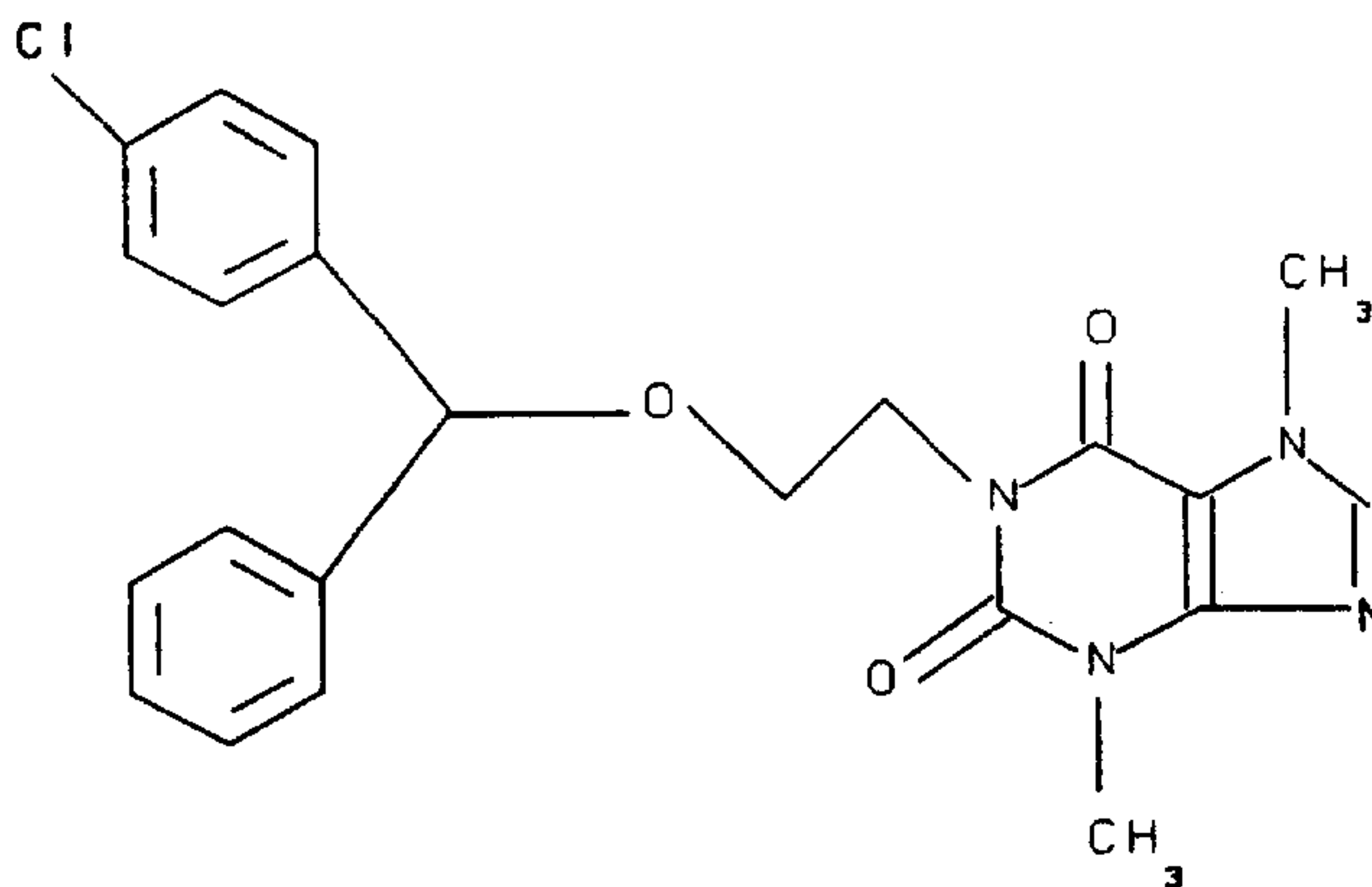
Compound 13

Compound 13 was prepared by preparing a solution of theobromine, (0.54 g, 0.003 mol), potassium t-butoxide (0.833 g, 0.002 mol) and sodium hydroxide (0.24 g, 0.006 mol) in 100 ml methanol, and stirring under nitrogen for about two hours. The tosylate of compound number 3 (2-diphenylmethoxyethanol) was formed in accordance with the general synthesis of formula (III) compounds and added to the mixture. The reaction mixture was stirred at room temperature for

about 24 hours, and the mixture was neutralized with hydrochloric acid (about 20 ml, 1N solution). The neutralized mixture then was purified by flash chromatography to produce about 25% of compound 13, which was a white crystalline material having a melting point within the range of from about 114-116°C, and a molecular weight of about 390.1.

Preparation of 3,7-Dimethyl-5-[2-(4-chlorophenyl phenylmethoxy)-ethyl]xanthine

Compound 14 below was prepared using the same procedure as described above for compound 13, except the chlorinated derivative of compound 3 was used instead of compound 3.



Compound 14

The procedure for preparing compound 13 was repeated, except that the theobromine solution was mixed with a chlorinated tosylate derivative of compound 3. The reaction mixture was stirred at room temperature for about 24 hours, and the mixture was neutralized with HCl (about 20 ml, 1N solution). The neutralized mixture then was purified by flash chromatography to produce compound 14, which was a hygroscopic white foam having a molecular weight of about 424.9.

EXAMPLE 5

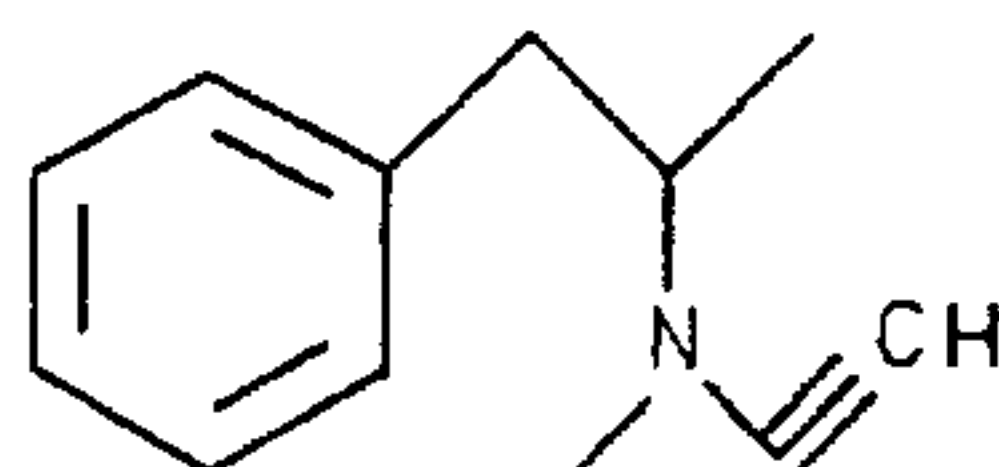
This example illustrates the effectiveness of a number of compounds of each of formula (I)-(VI) in reducing the number, quantity, and/or volume of disrupted DMS. This example illustrates the effectiveness of a number of compounds in: (i) reducing mean volume of tissue occupied by the disrupted DMS; (ii) reducing the ratio of the number of inflammatory cells per DMS; or

(iii) increasing the ratio of the number of macrophages containing disrupted DMS material per DMS. This example therefore illustrates the effectiveness of the inventive compounds in reducing the exponential DMS disruption autocatalytic phenomenon and hence, in reducing the cerebral amyloid burden and formation of amyloid plaque.

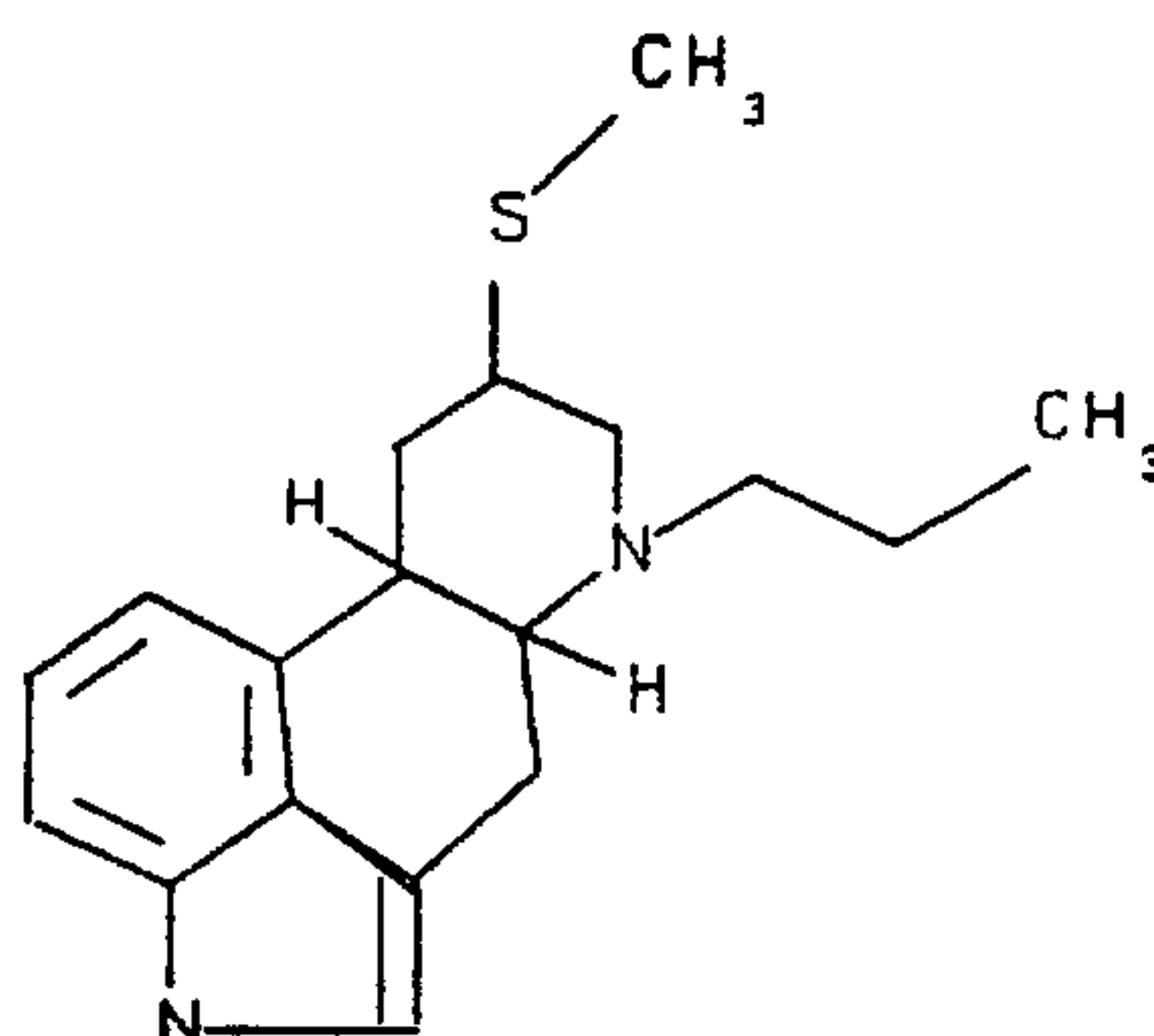
1. *In vivo* testing of compounds of formula (I)-(VI)

The compounds described above in Example 4 were tested in accordance with Example 2. In the following examples, the data of which is reported separately below in Tables I-VII, each of the compounds was administered to four (4) rats in sterile physiological saline containing about 100,000 human DMS and about 50 μ l of the compound at 10^{-5} M concentration, for a total volume of about 100 μ l. Controls were used whereby the control groups were comprised of: (i) four (4) rats that did not receive any injection of DMS or the compound; (ii) four (4) rats that received an injection of DMS and no compound; and (iii) two (2) rats that received an injection of DMS and a compound found to be inactive in Example 2 above. The inactive compounds are listed below as control compounds 1-26, and the results are formulated in Tables I-VII below.

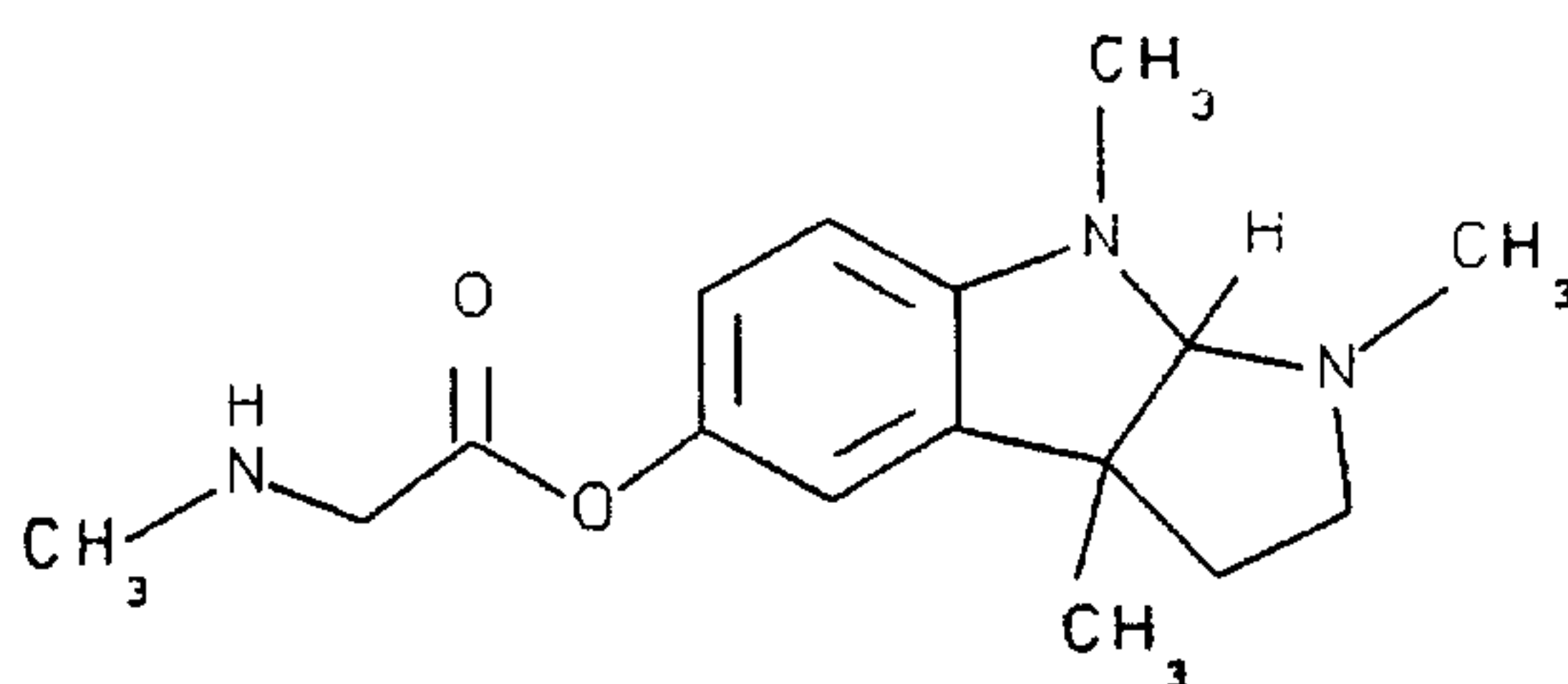
Inactive Control Compounds



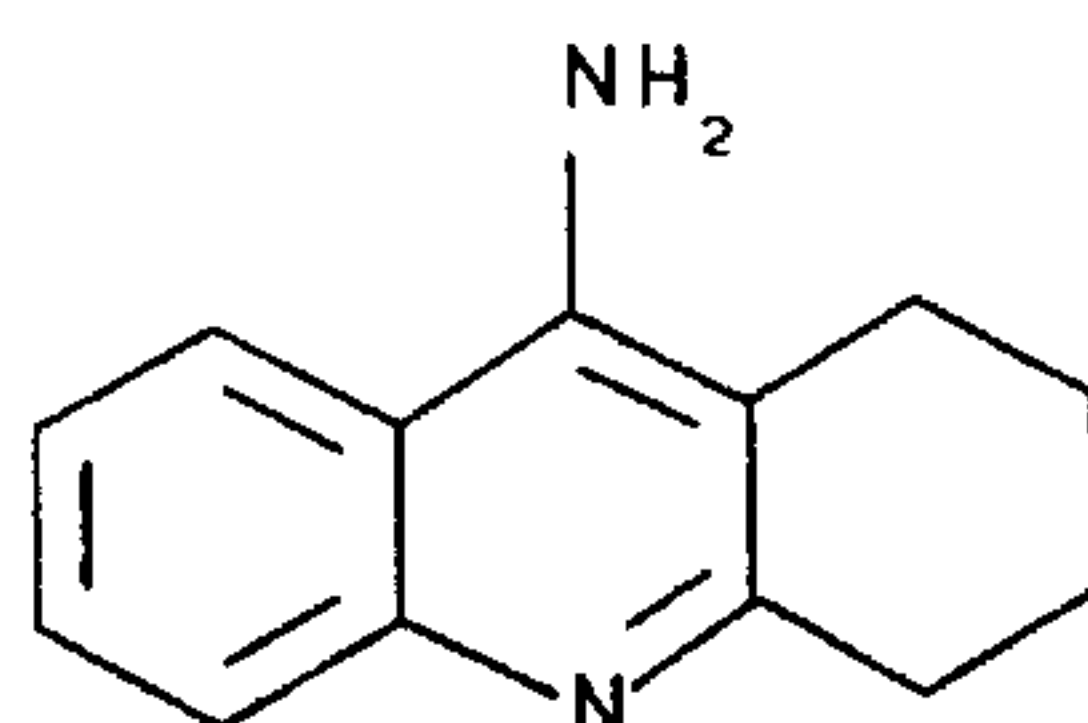
Control 1 - Selegiline (Deprenil)



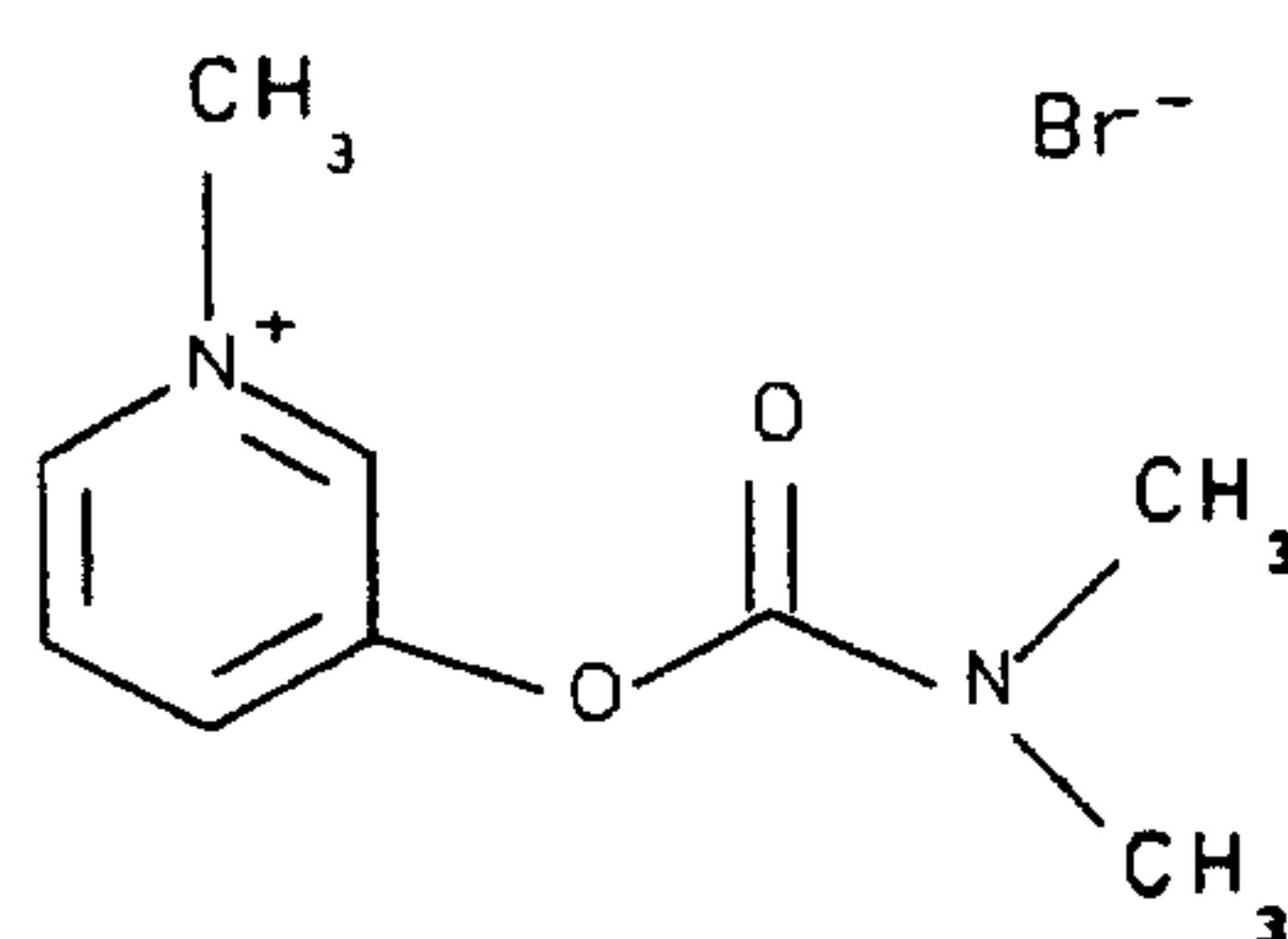
Control 2 - Pergolide



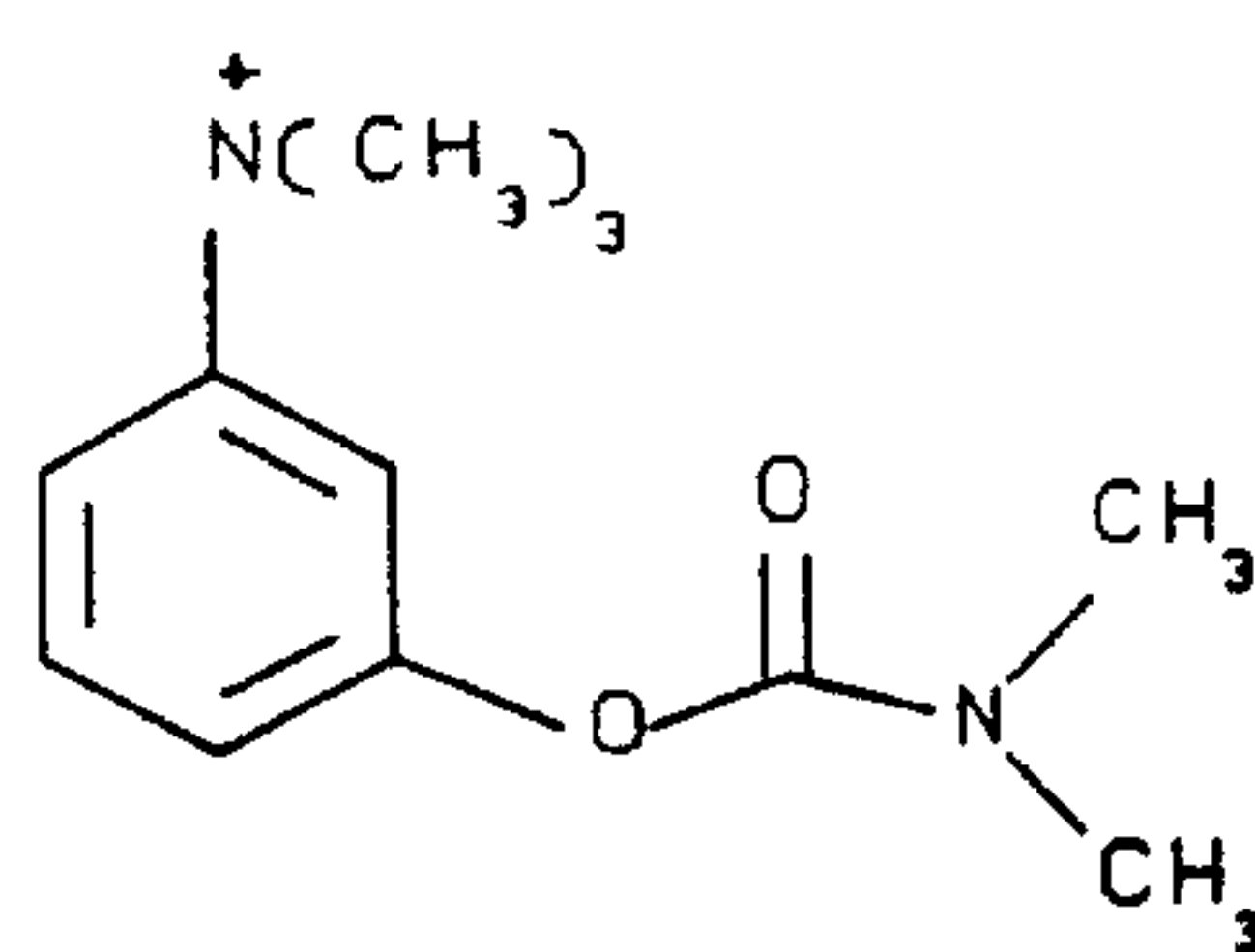
Control 3 - Physostigmine



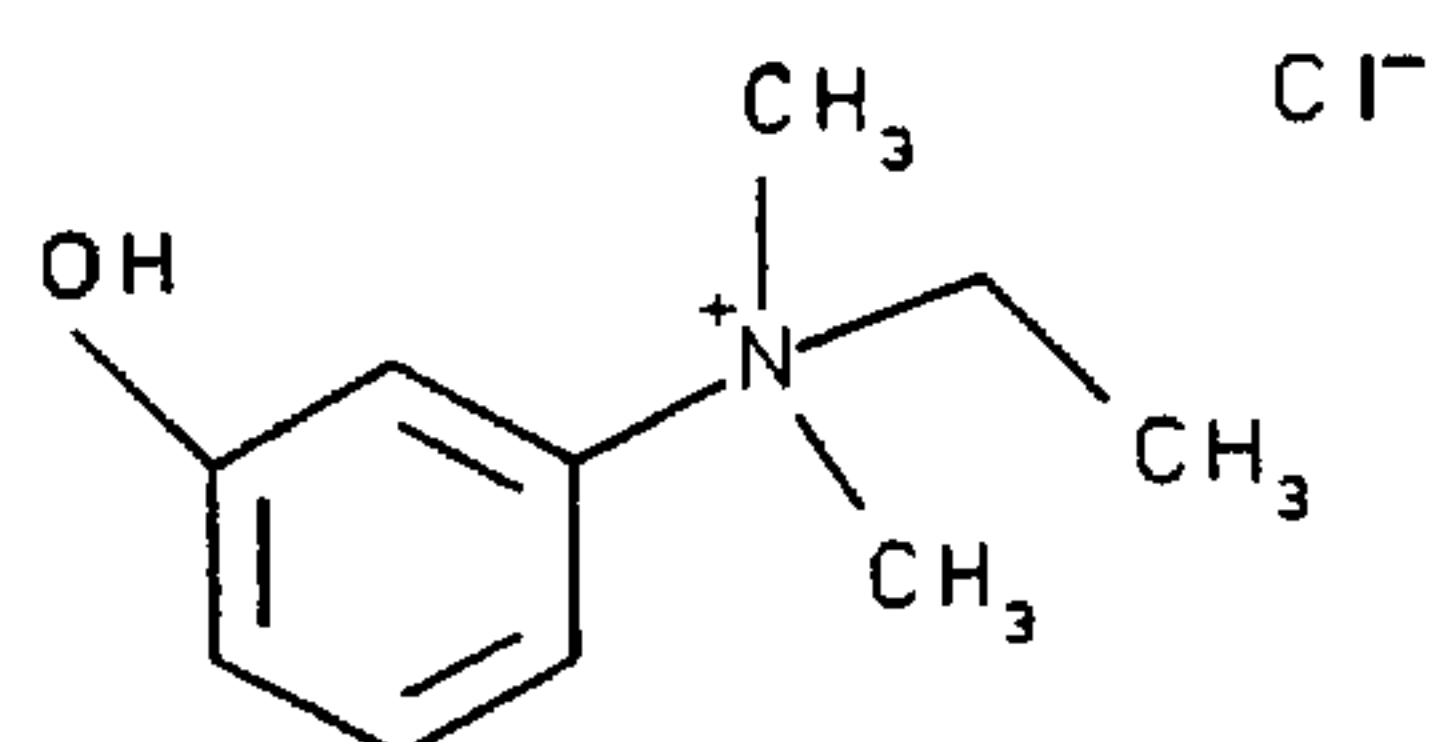
Control 4 - Tacrine



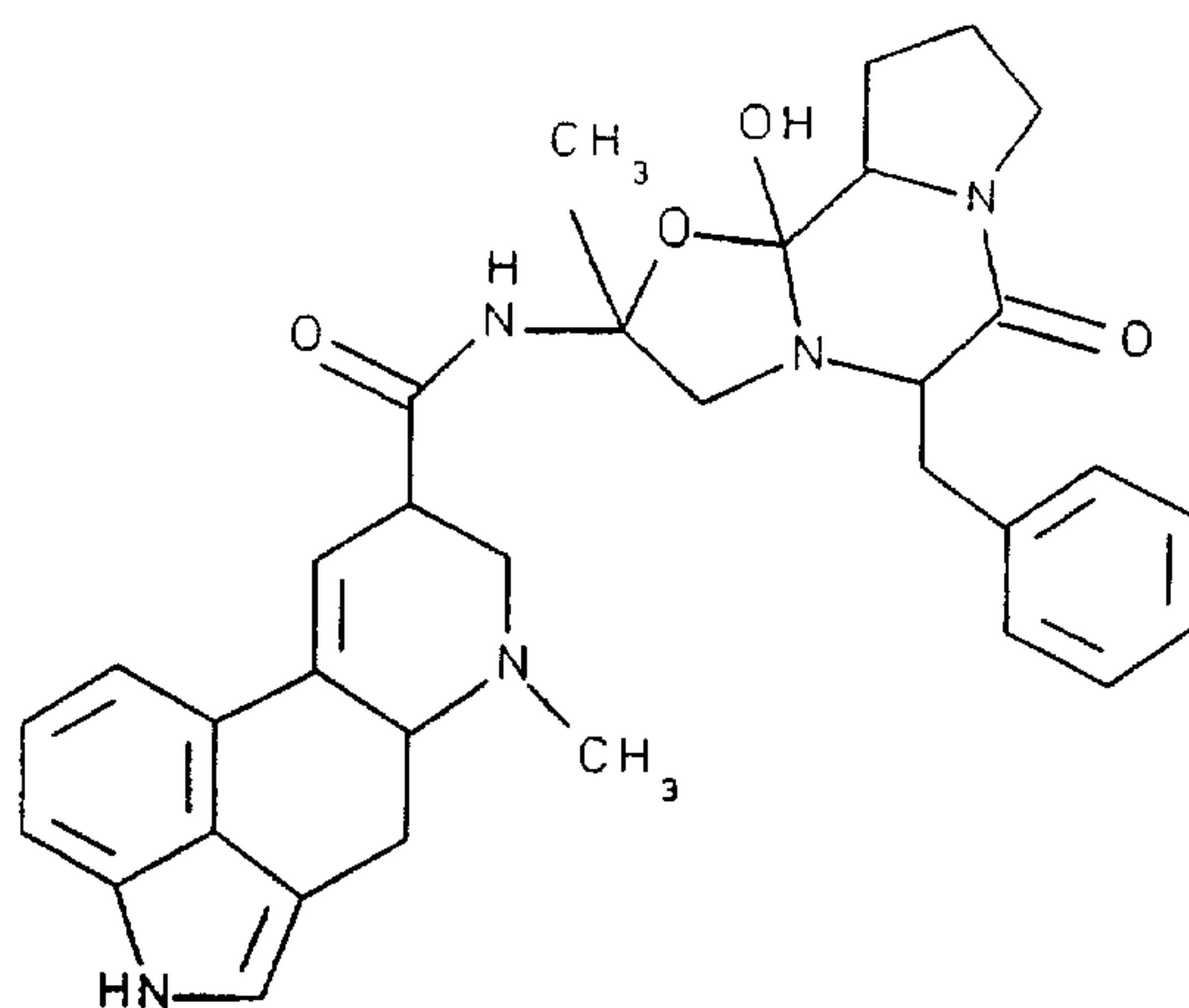
Control 5 - Pyridostigmine Bromide



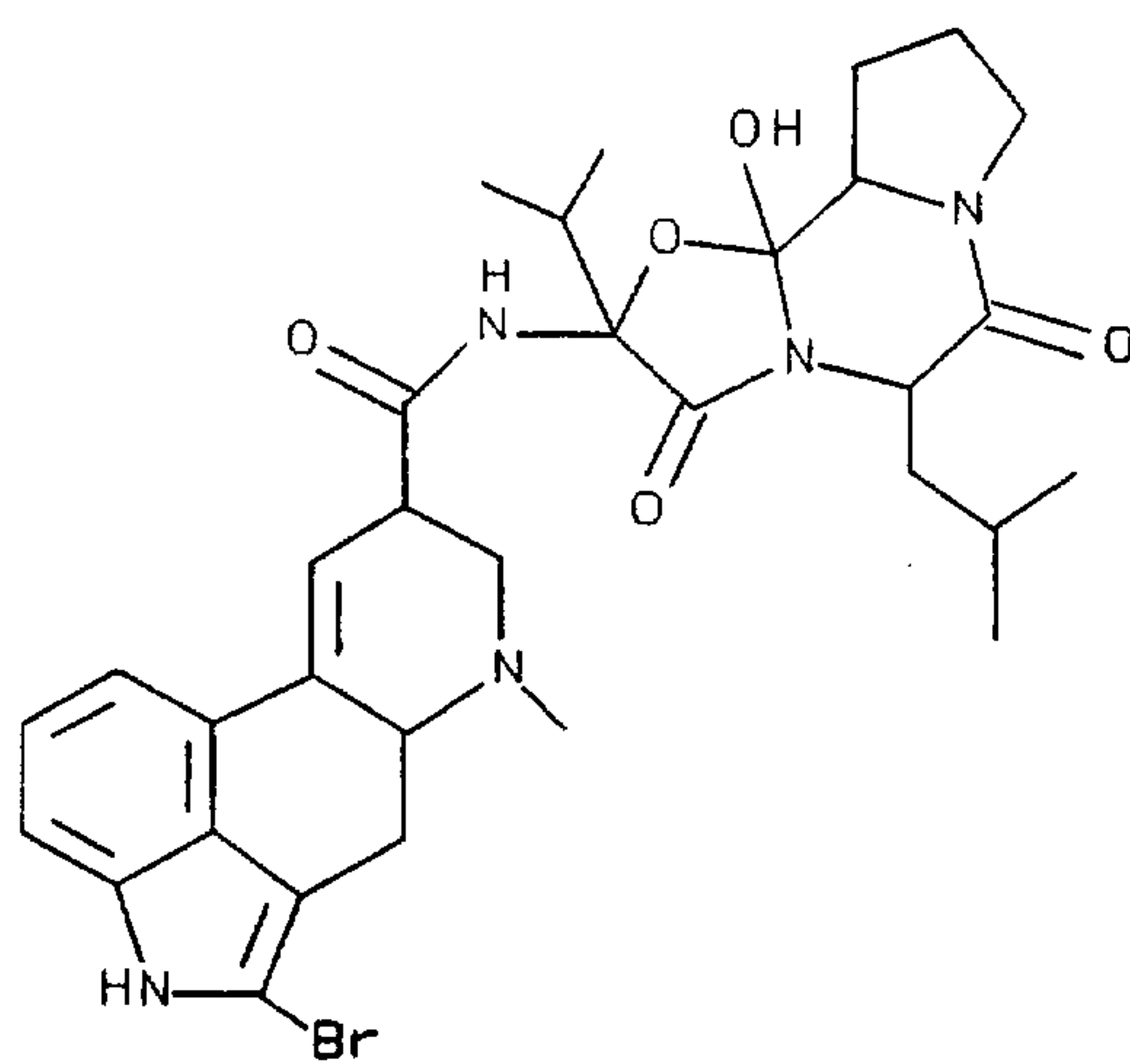
Control 6 - Neostigmine



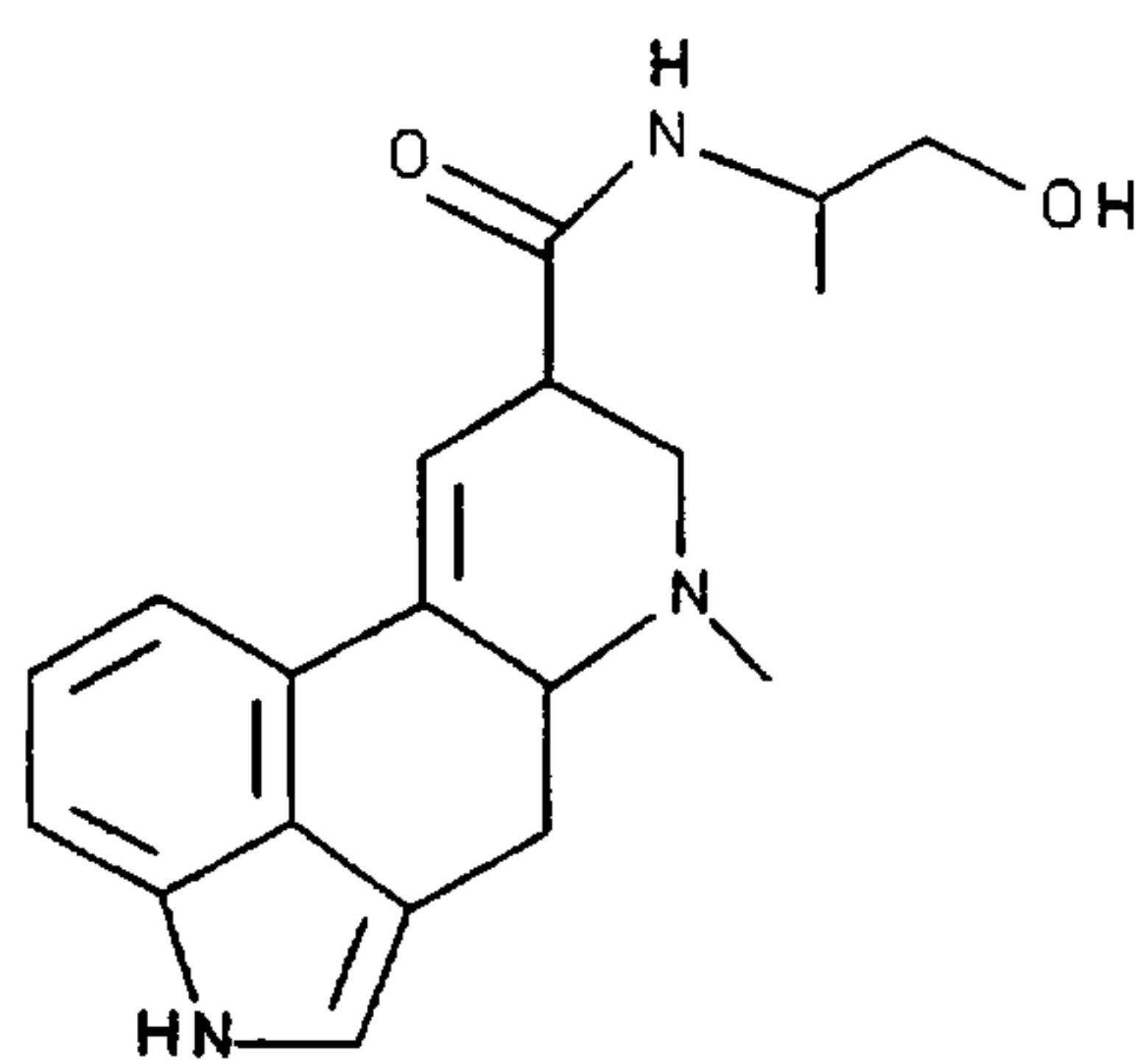
Control 7 - Edrophonium Chloride



Control 9 - Ergotamine



Control 9 - Bromocriptine

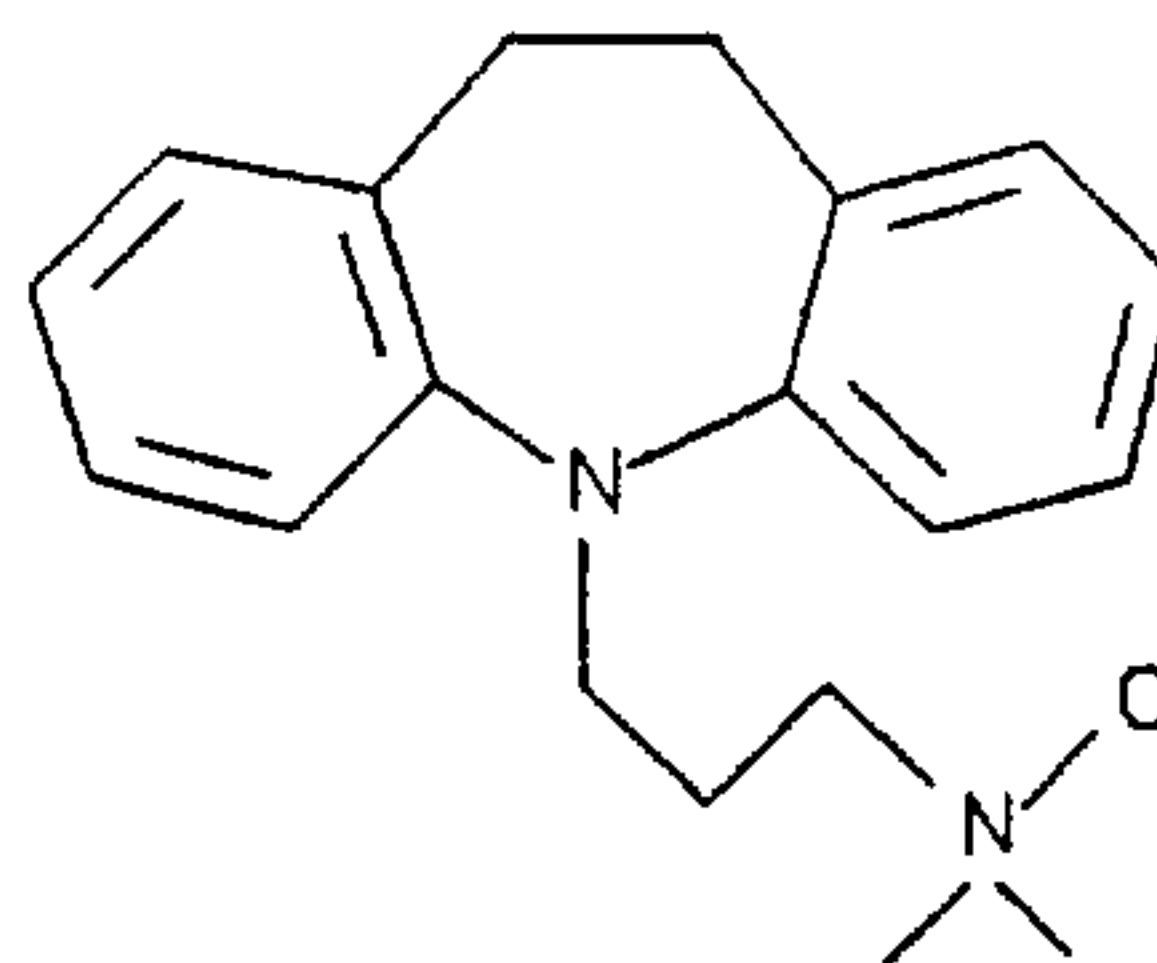
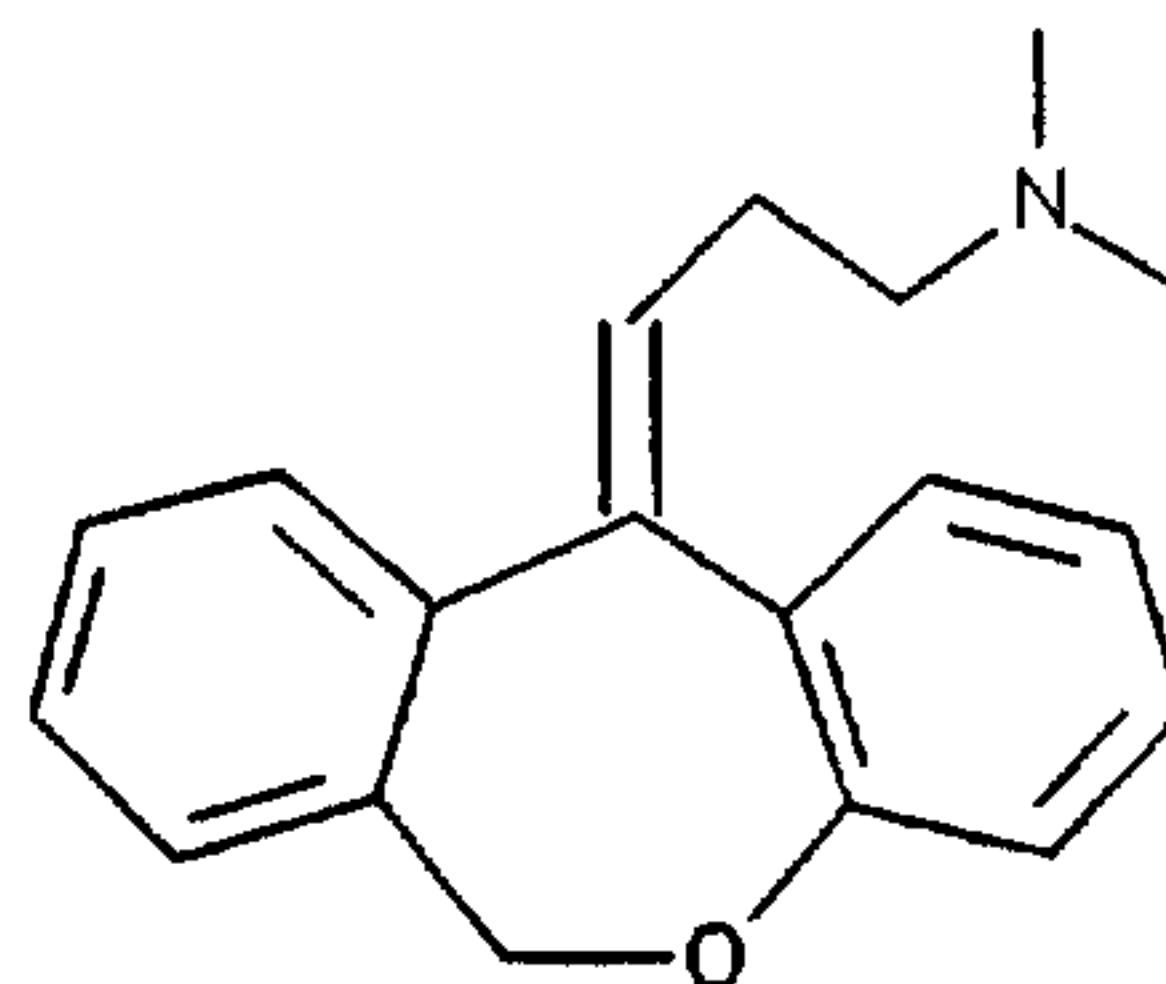
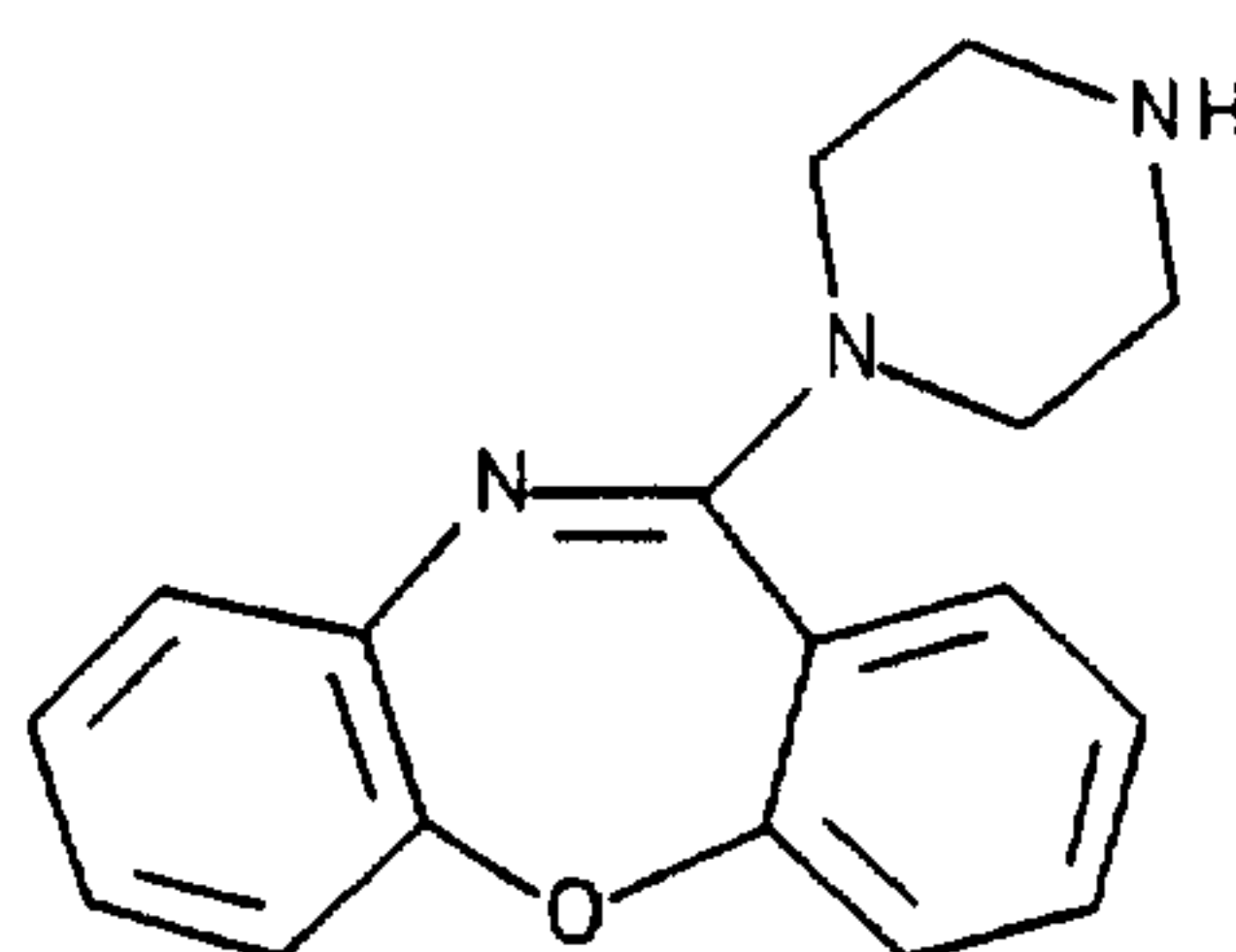
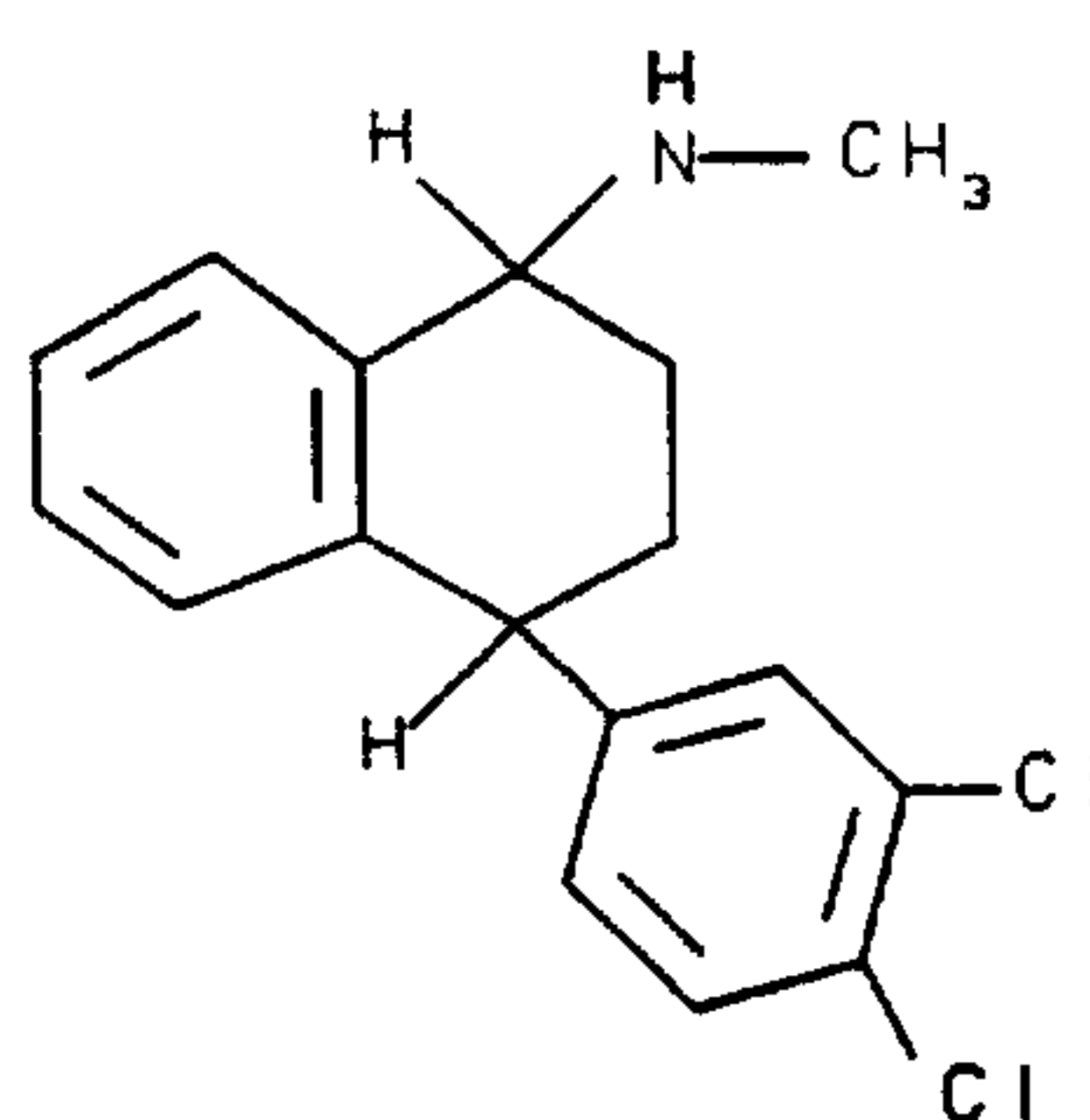


Control 10 - Ergonovine

WO 98/52898

-57-

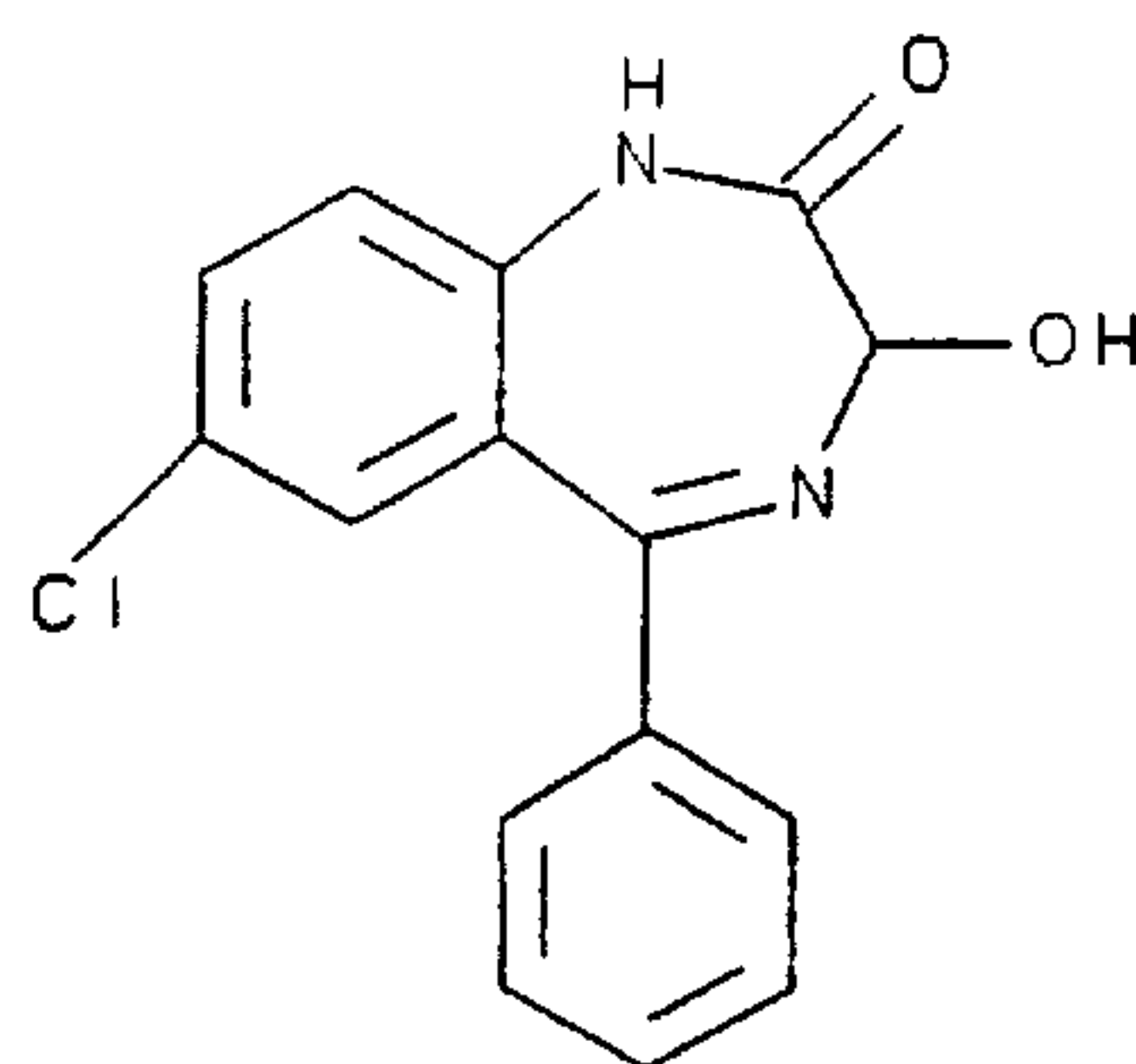
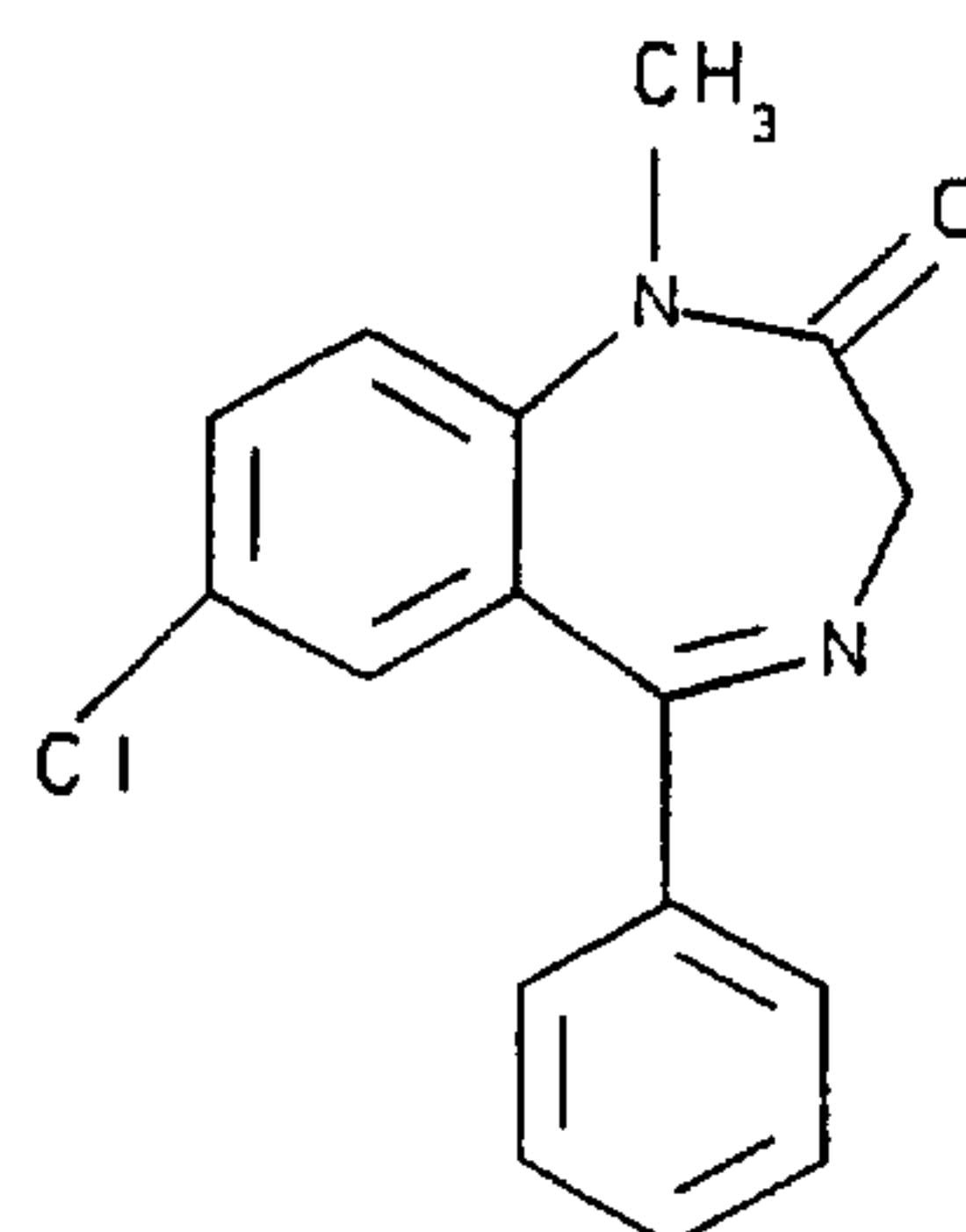
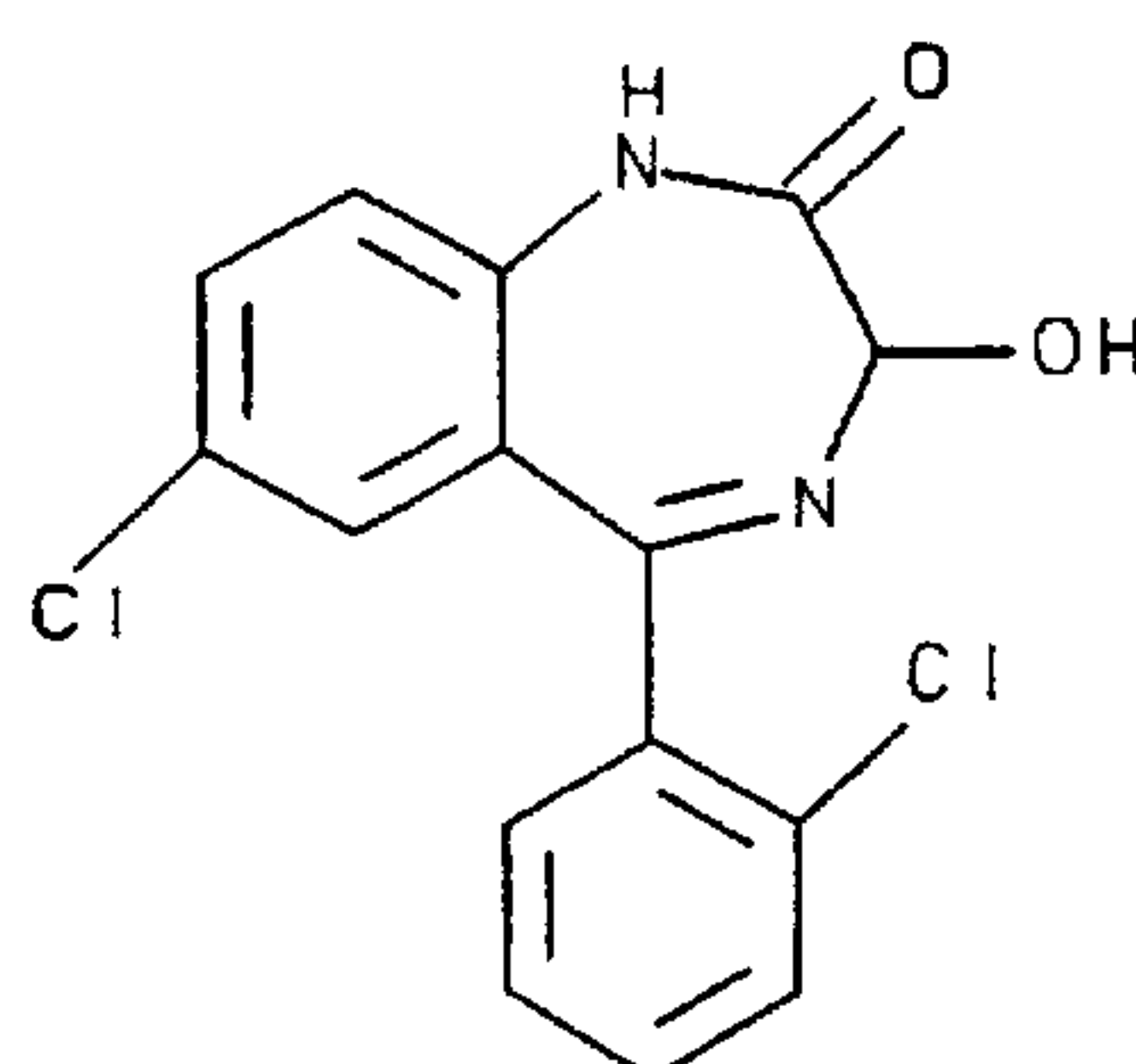
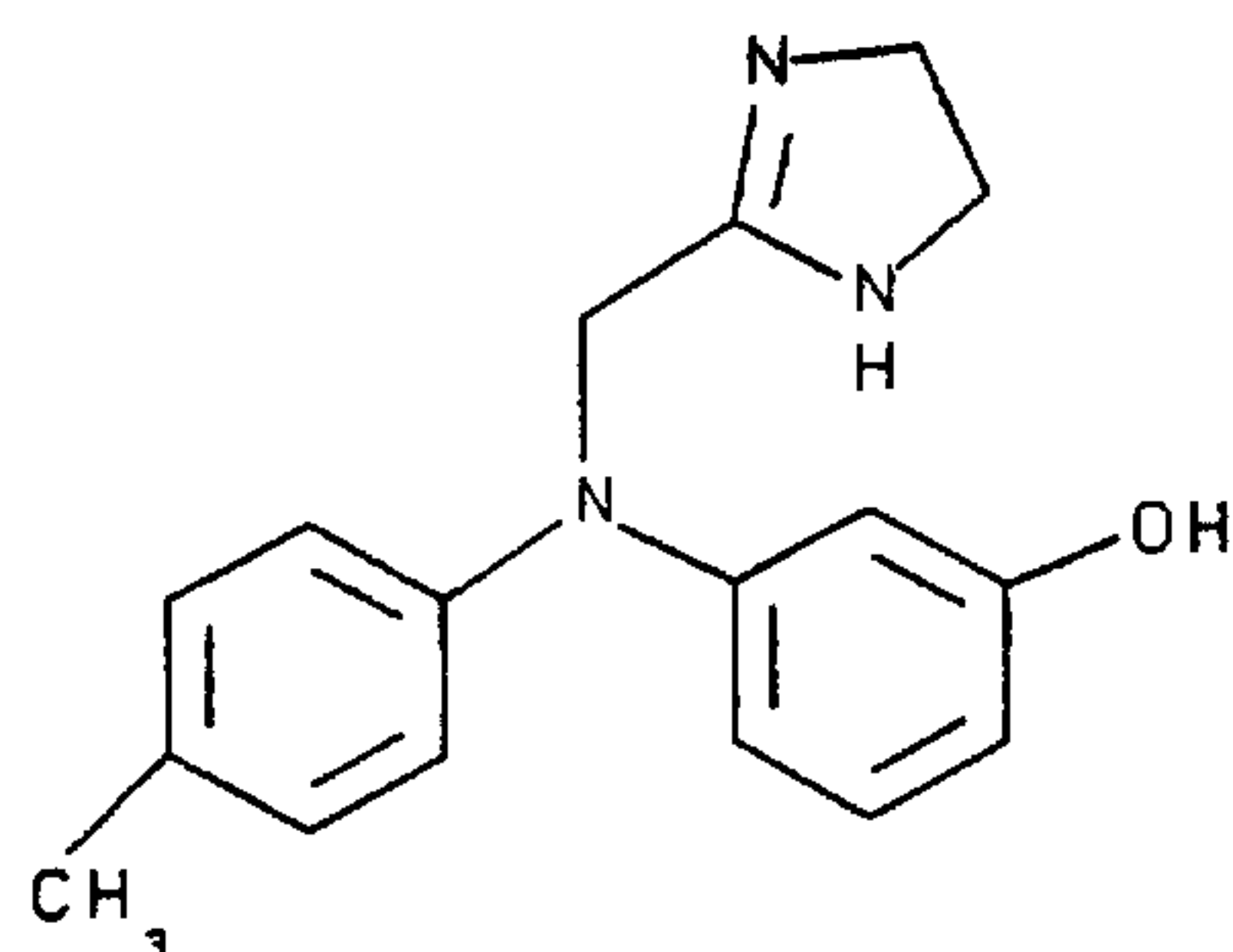
PCT/US98/10218

Control 11 - ImipramineControl 12 - DoxepinControl 13 - AmoxepineControl 14 - Sertraline

WO 98/52898

-58-

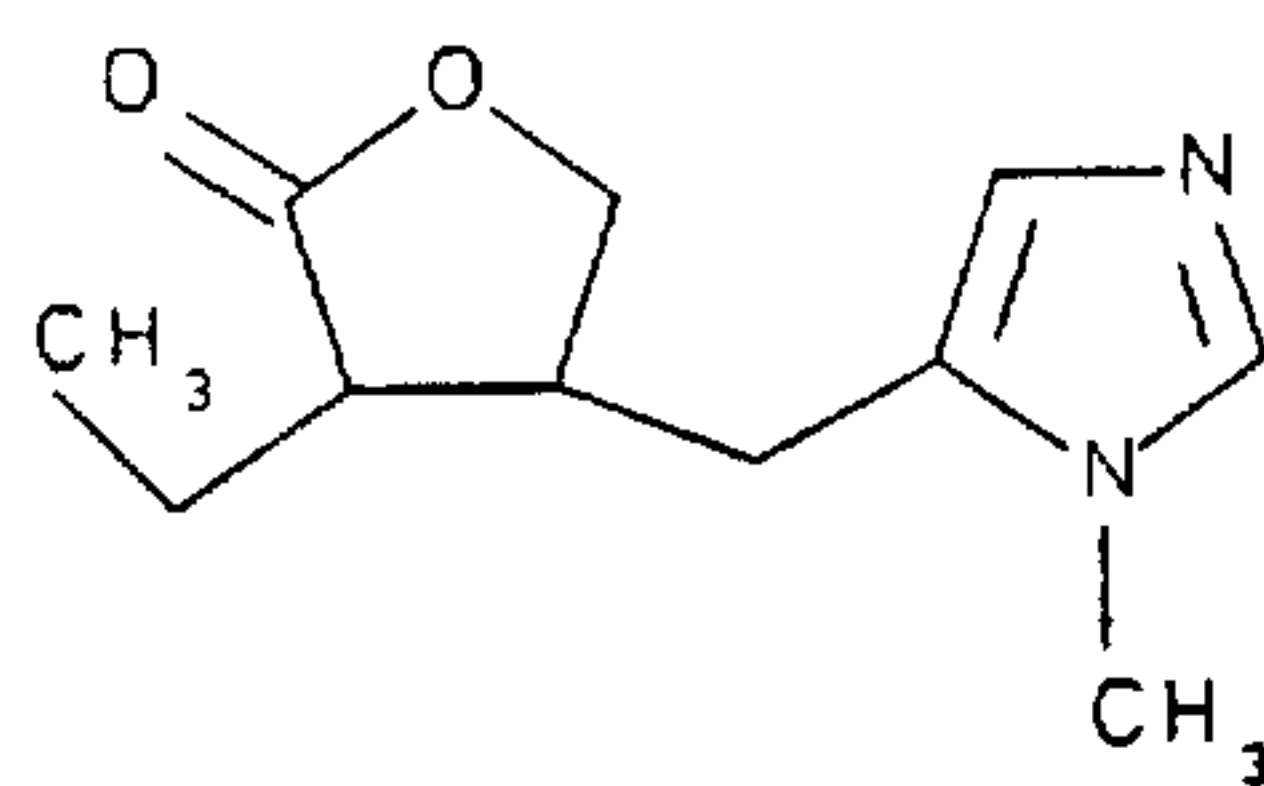
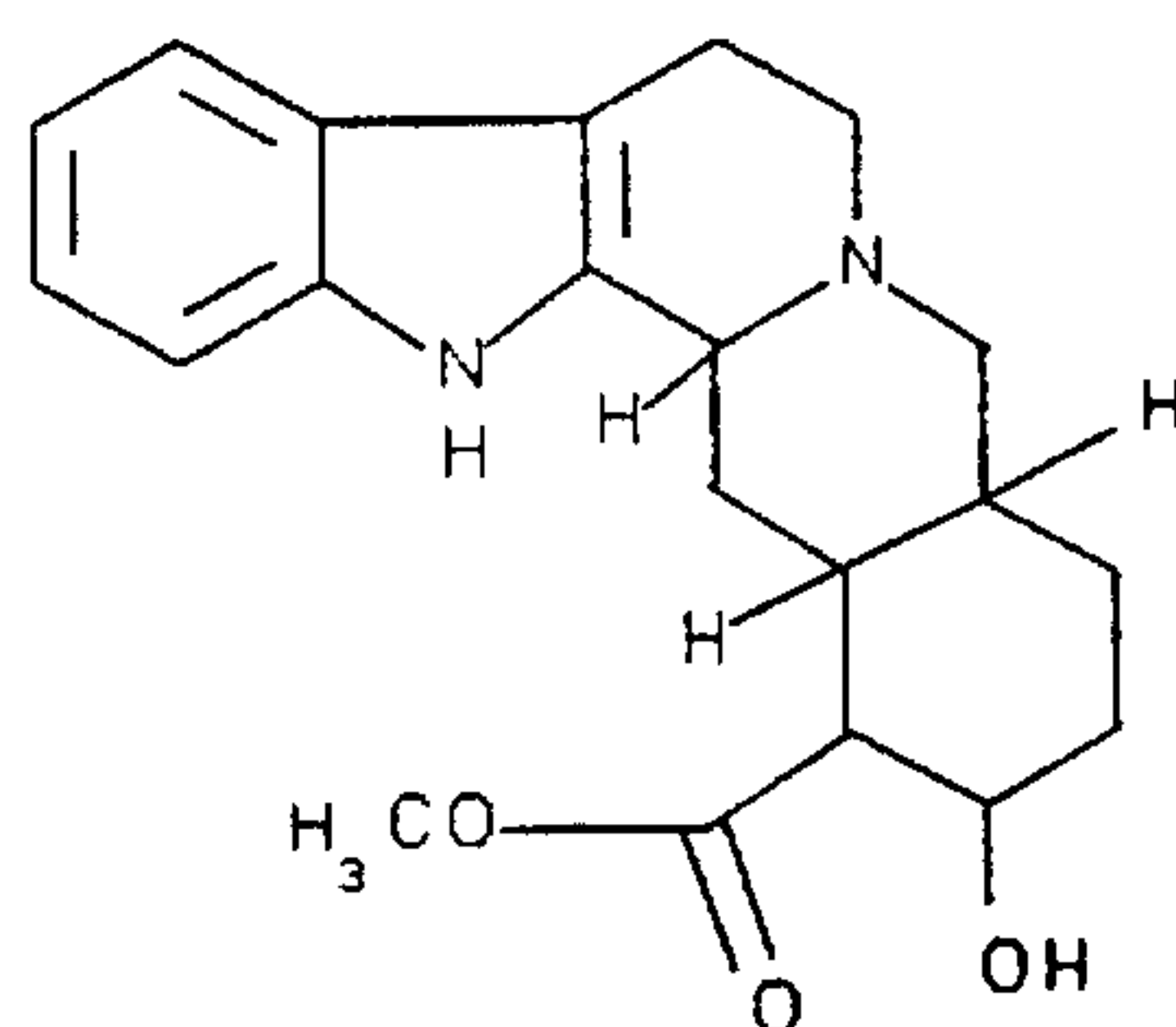
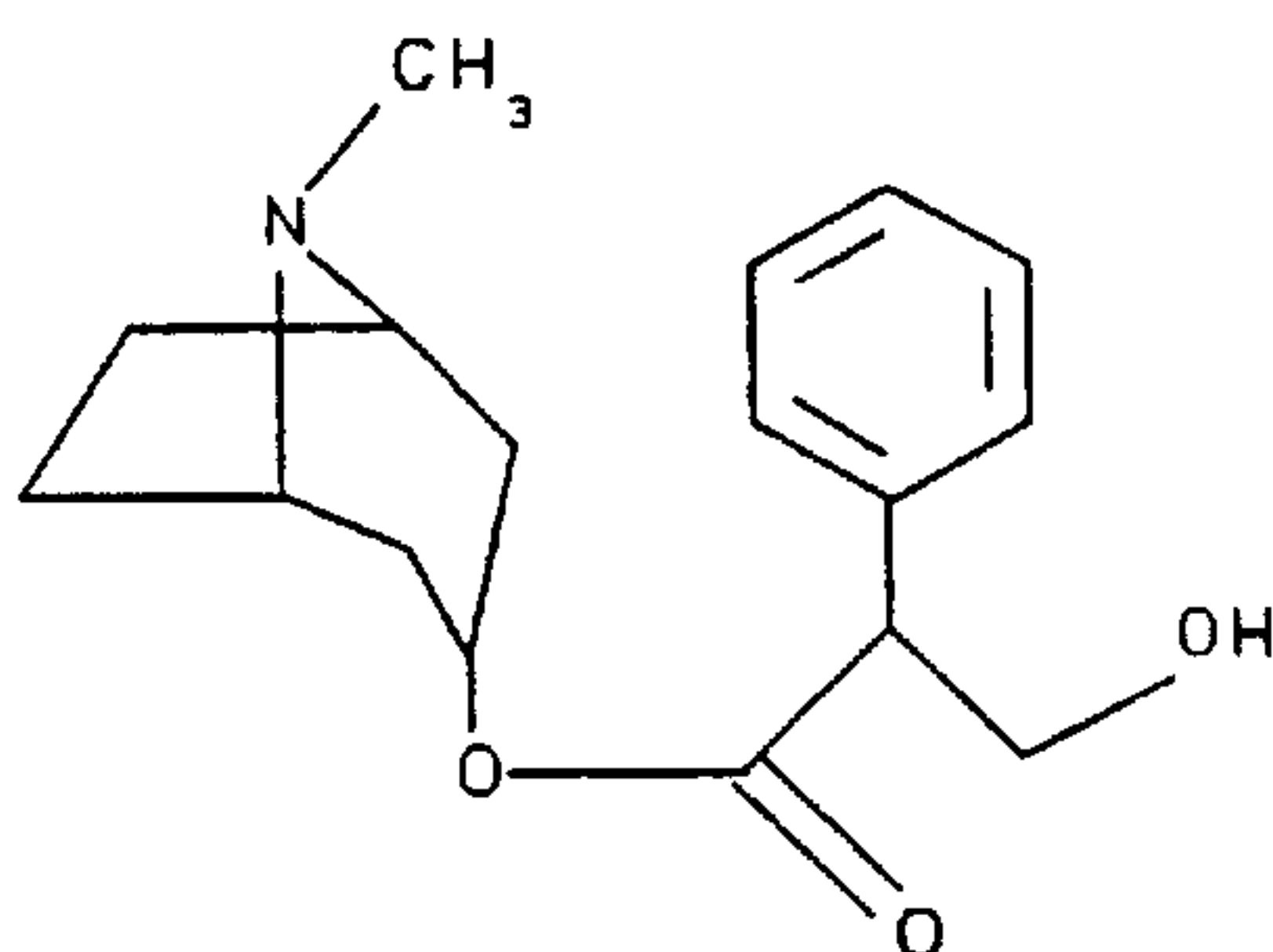
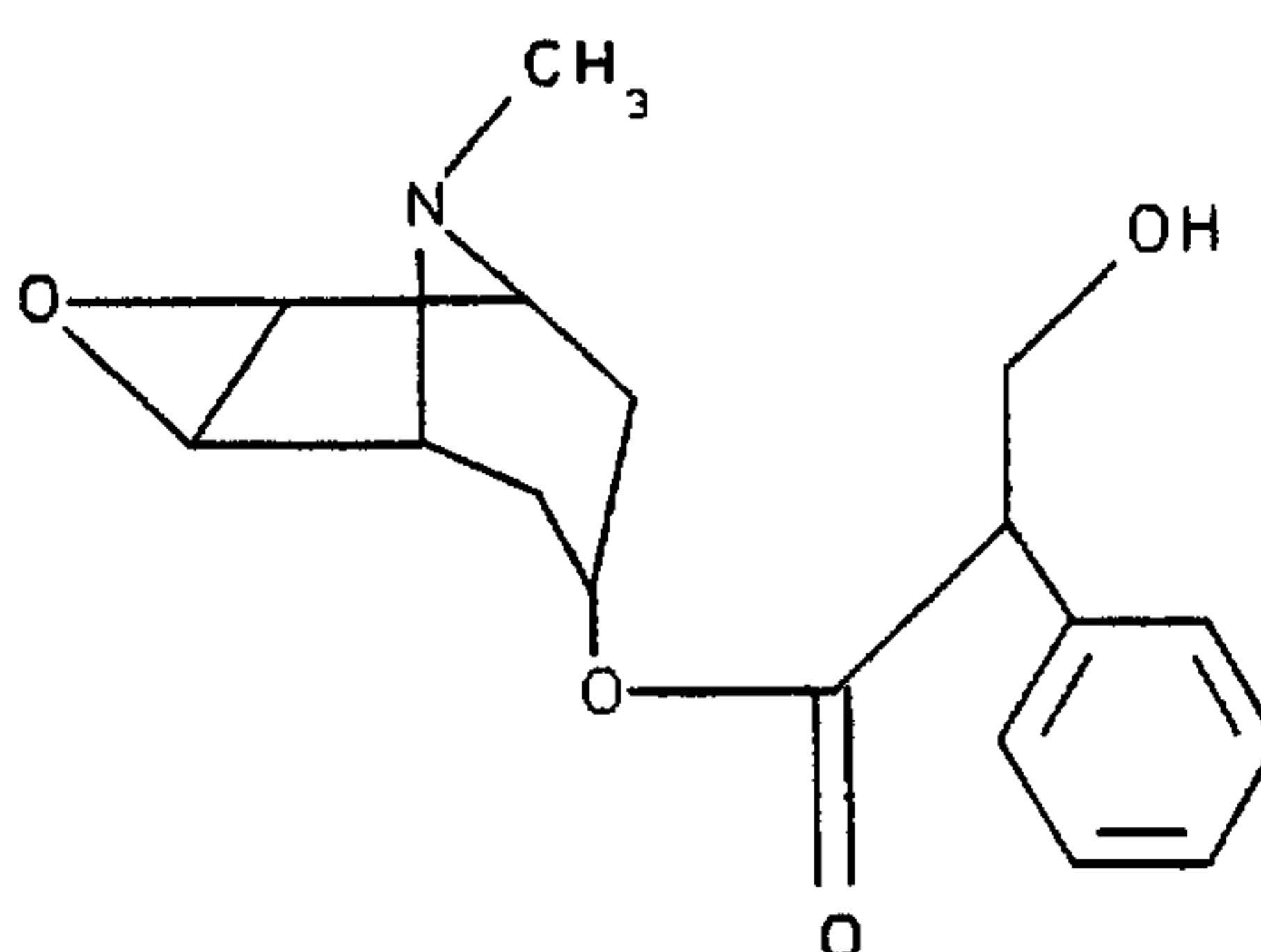
PCT/US98/10218

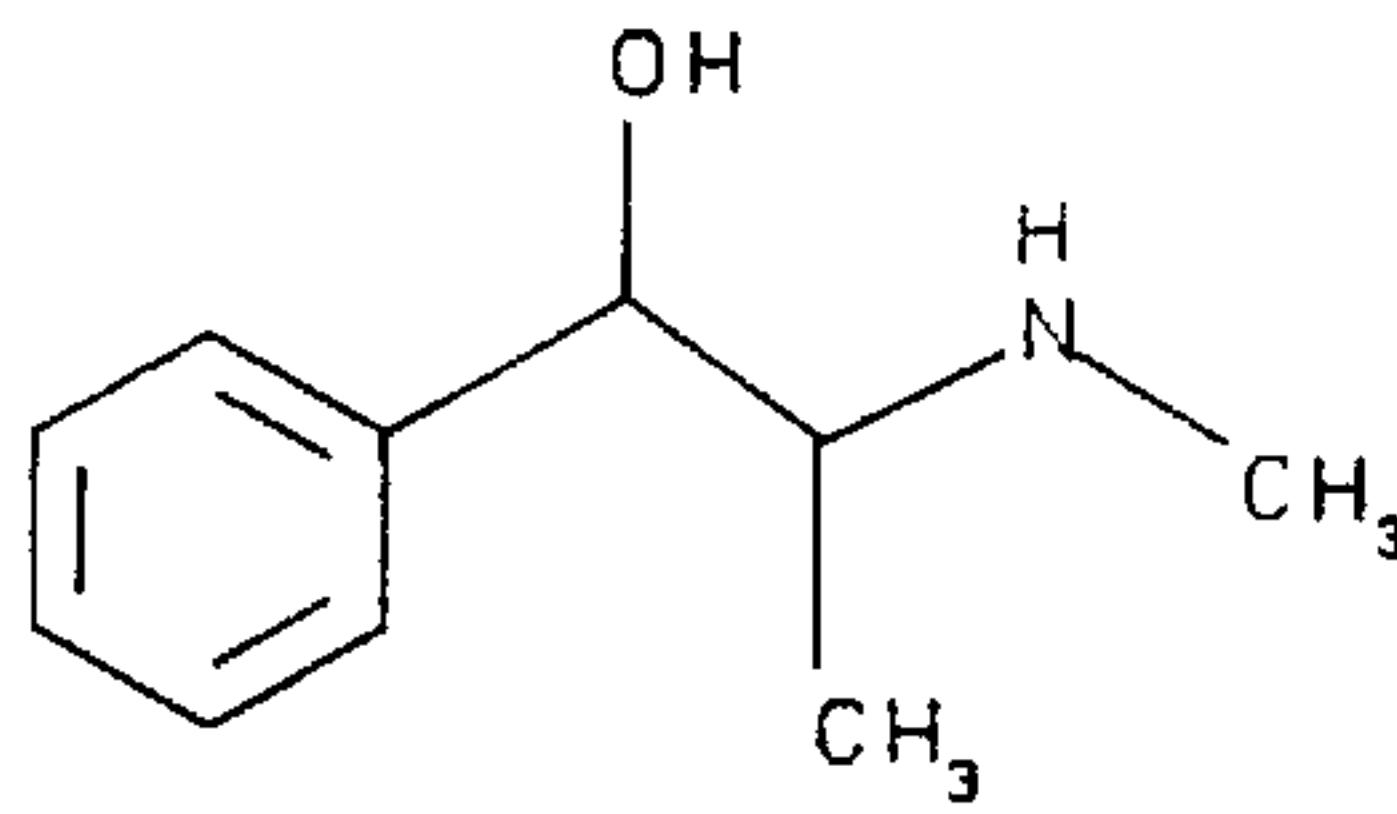
Control 15 - OxazepamControl 16 - Diazepam (Valium)Control 17 - LorazepamControl 18 - Phentolamine

WO 98/52898

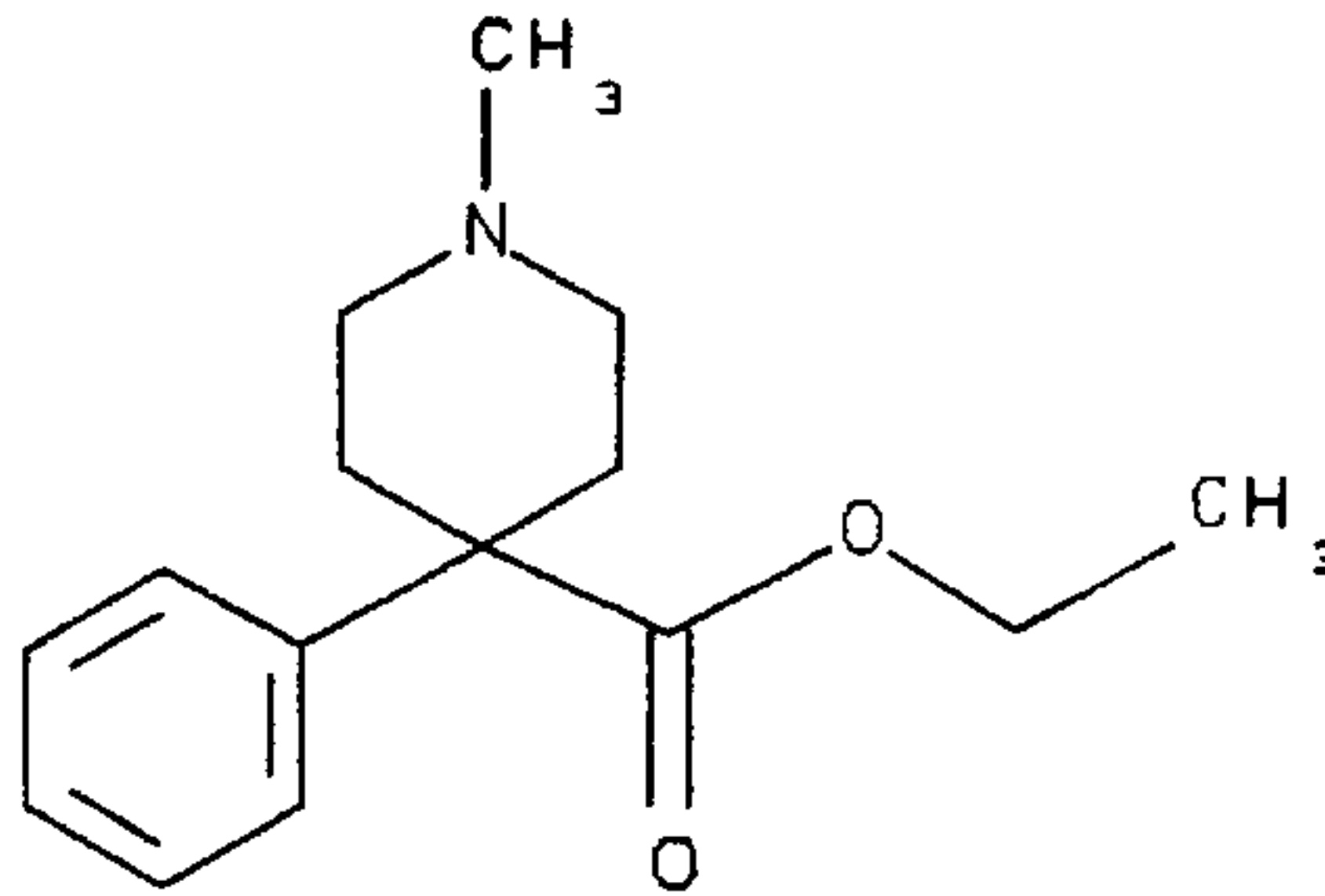
-59-

PCT/US98/10218

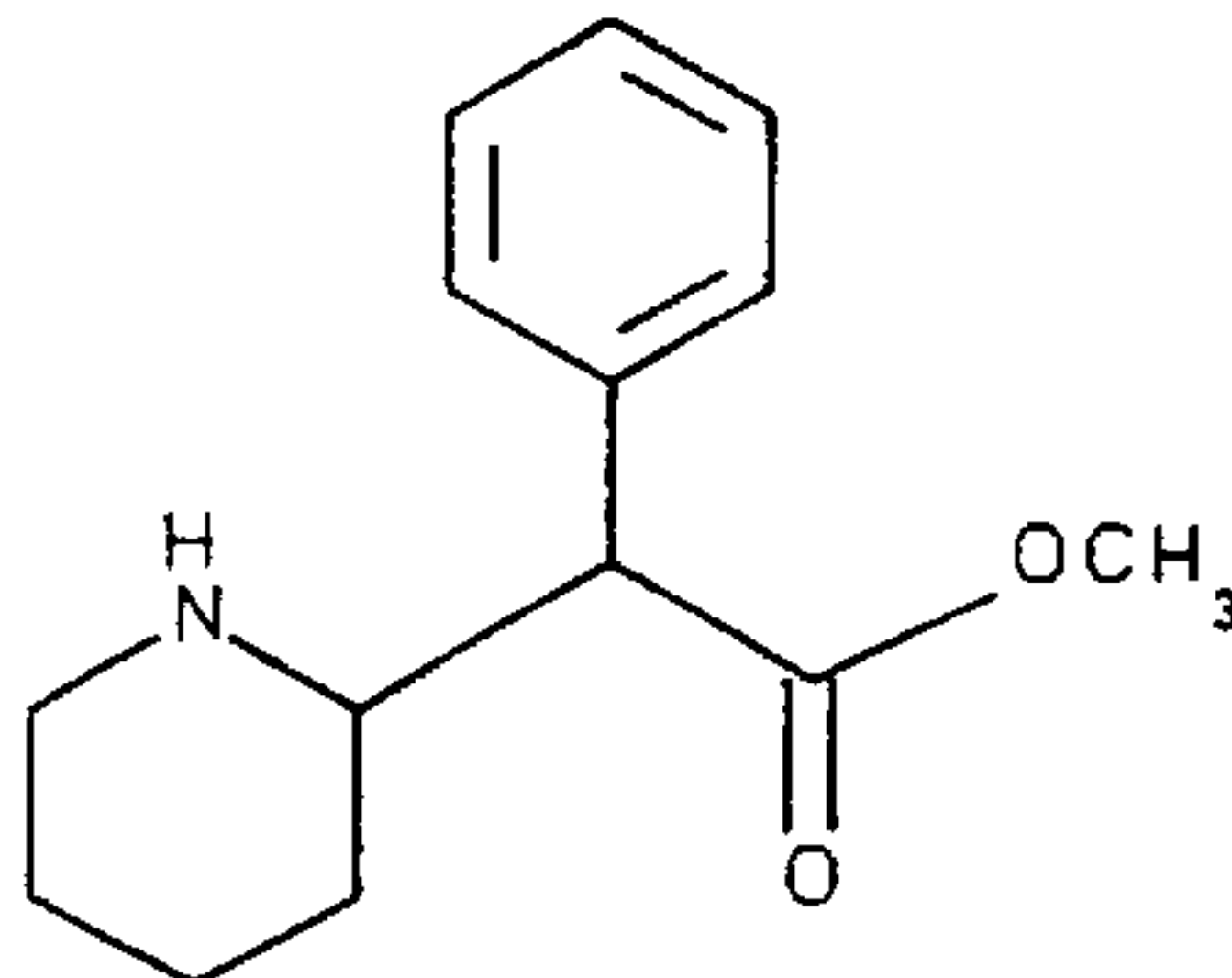
Control 19 - PilocarpineControl 20 - YohimibineControl 21 - AtropineControl 22 - Scopolamine



Control 23 - Ephedrine



Control 24 - Meperidine



Control 25 - Methylphenidate

5 The following tables summarizes the data obtained in accordance with this example. In the tables, the various compounds of the present invention were tested against: control (i), animals that did not receive any injection of DMS or
10 either compounds of the invention or control compounds; control (ii), animals that received an injection of DMS and no compound; and control (iii), animals that received an injection of DMS and an inactive control compound. The initial mean volume of tissue occupied by the DMS, the mean volume of tissue occupied by
disrupted DMS, the ratio of the number of inflammatory cells per DMS (*i.e.*, persistence of inflammatory reaction) and the ratio of the number of macrophages

containing disrupted DMS material (*i.e.*, digestion and removal of DMS) were measured and tabulated.

1. Compounds of Formula (I)

TABLE I

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS ¹ (μ^3)	Persistence of Inflamm. Reaction ²	Digestion and removal of DMS ³
1	1153	1250	< 0.01	> 10
Cont. (i)	1153	0	0	0
Cont. (ii)	1153	1.7x10 ⁵	1.0	1.0
Atropine	1153	1.8x10 ⁵	1.0	1.0
Scopolamine	1153	1.8x10 ⁵	1.0	1.0
Meperidine	1153	2.1x10 ⁵	1.0	1.0

¹ - denotes the mean tissue volume of transformed redistributed disrupted DMS protein per individual DMS;

² - denotes persistence of inflammatory reaction at DMS disruption site: ratio of number of acute inflammatory cells (excluding macrophages) per DMS at the DMS disruption site, compared to control (ii), the latter normalized to 1.0; and

³ - denotes evidence of digestion and removal of DMS material by reticuloendothelial system: ratio of number of macrophages containing intact or digested, altered, proteolyzed or otherwise transformed DMS per DMS at the DMS disruption site, compared to control (ii), the latter normalized to 1.0.

2. Compounds of Formula (II)

TABLE II

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
2	525	560	< 0.01	> 10
4	525	580	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	525	1.5×10^5	1.0	1.0
Physostigmine	525	1.1×10^5	1.0	1.0
Pyridostigmine bromide	525	1.5×10^5	1.1	1.0
Tacrine	525	1.4×10^5	1.1	1.0

TABLE III

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
5	907	940	< 0.01	> 10
6	907	940	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	907	1.05×10^5	1.0	1.0
Pergolide	907	1.2×10^5	1.1	1.0
Bromocriptine	907	1.04×10^5	1.0	1.0
Selegiline	907	1.3×10^5	1.0	1.0

3. Compounds of Formula (III)

TABLE IV

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
9	383	482	< 0.01	> 10
10	383	460	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	383	7.3×10^4	1.0	1.0
Diazepam	383	6.5×10^4	1.1	1.0
Oxazepam	383	6.8×10^4	1.1	1.0
Lorazepam	383	8.2×10^4	1.1	1.0

4. Compounds of Formula (IV)

TABLE V

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
11	907	1260	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	907	1.02×10^5	1.0	1.0
Imipramine	907	1.38×10^5	1.0	1.0
Sertraline	907	1.22×10^5	1.1	1.0
Amoxepine	907	1.36×10^5	1.1	1.0

5. Compounds of Formula (V)

TABLE VI

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
12	525	740	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	525	5.1x10 ⁴	1.0	1.0
Ergotamine	525	5.8x10 ⁴	1.0	1.0
Ergonovine	525	5.2x10 ⁴	1.0	1.0
Methylphenidate	525	5.8x10 ⁴	1.1	1.01

6. Compounds of Formula (VI)

TABLE VII

Compound	Mean Volume DMS (μ^3)	Mean Volume Disrupted DMS (μ^3)	Persistence of Inflamm. Reaction	Digestion and removal of DMS
13	1440	1625	< 0.01	> 10
14	1440	1625	< 0.01	> 10
Cont. (i)	0	0	0	0
Cont. (ii)	1440	1.7x10 ⁵	1.0	1.0
Neostigmine	1440	1.4x10 ⁵	1.1	1.0
Edrophonium Chloride	1440	2.2x10 ⁵	1.0	1.0
Pilocarpine	1440	1.7x10 ⁵	1.1	1.0

As can be seen from the table above, the compounds of the present invention reduced the mean tissue volume of disrupted DMS and associated injury foci *in vivo* by at least 10% when compared to control groups (ii) and (iii). As

can also be seen in the table above, the compounds of the present invention furthermore reduced the ratio of the number of inflammatory cells per DMS at the DMS disruption site by at least 10% when compared to control groups (ii) and (iii). In addition, as can be seen from the table above, the compounds of the present invention furthermore increased the ratio of the number of macrophages containing disrupted DMS material at the DMS disruption site by greater than 10% when compared to control groups (ii) and (iii).

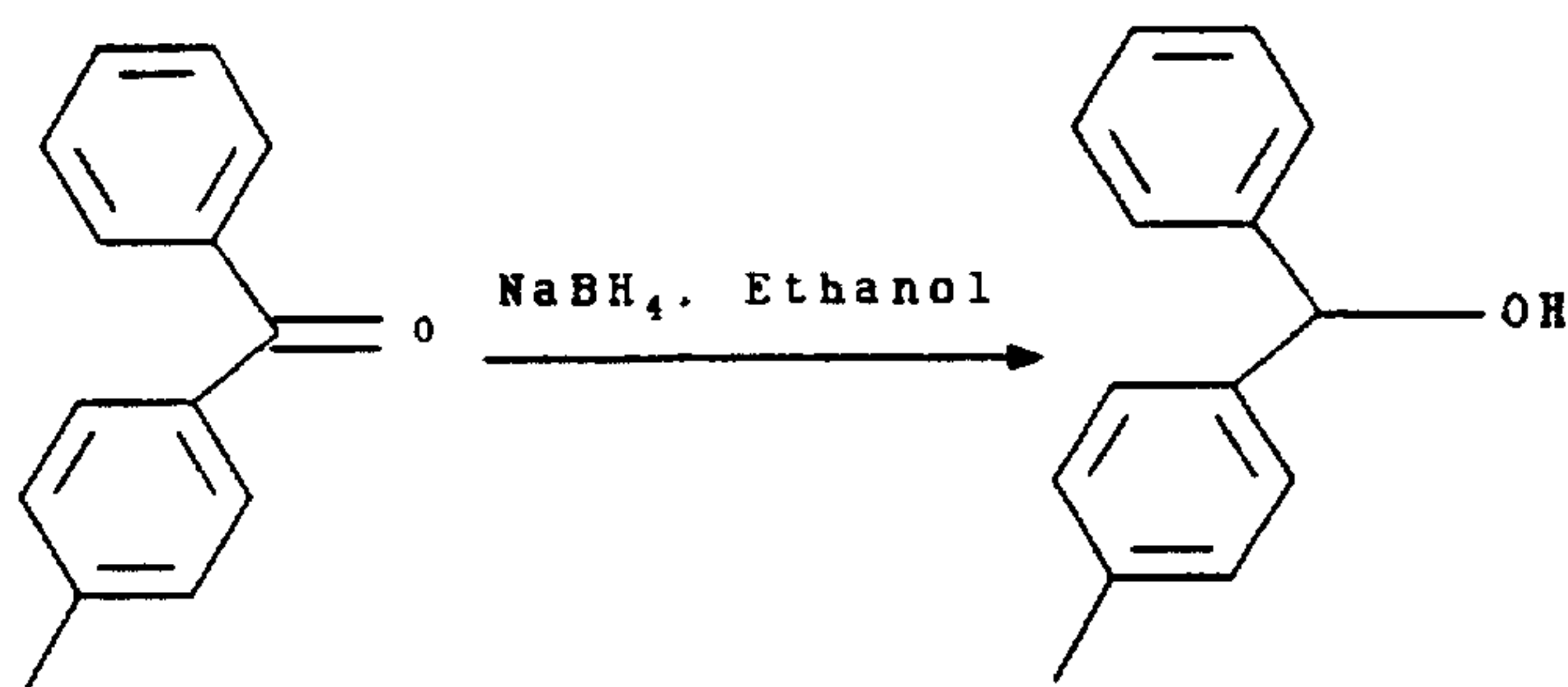
EXAMPLE 6

This example illustrates the synthesis of a number of compounds of general formula A.

Synthesis Example 1 - 1,2-Bis(4-methylphenyl phenylmethoxy)ethane:

Step 1

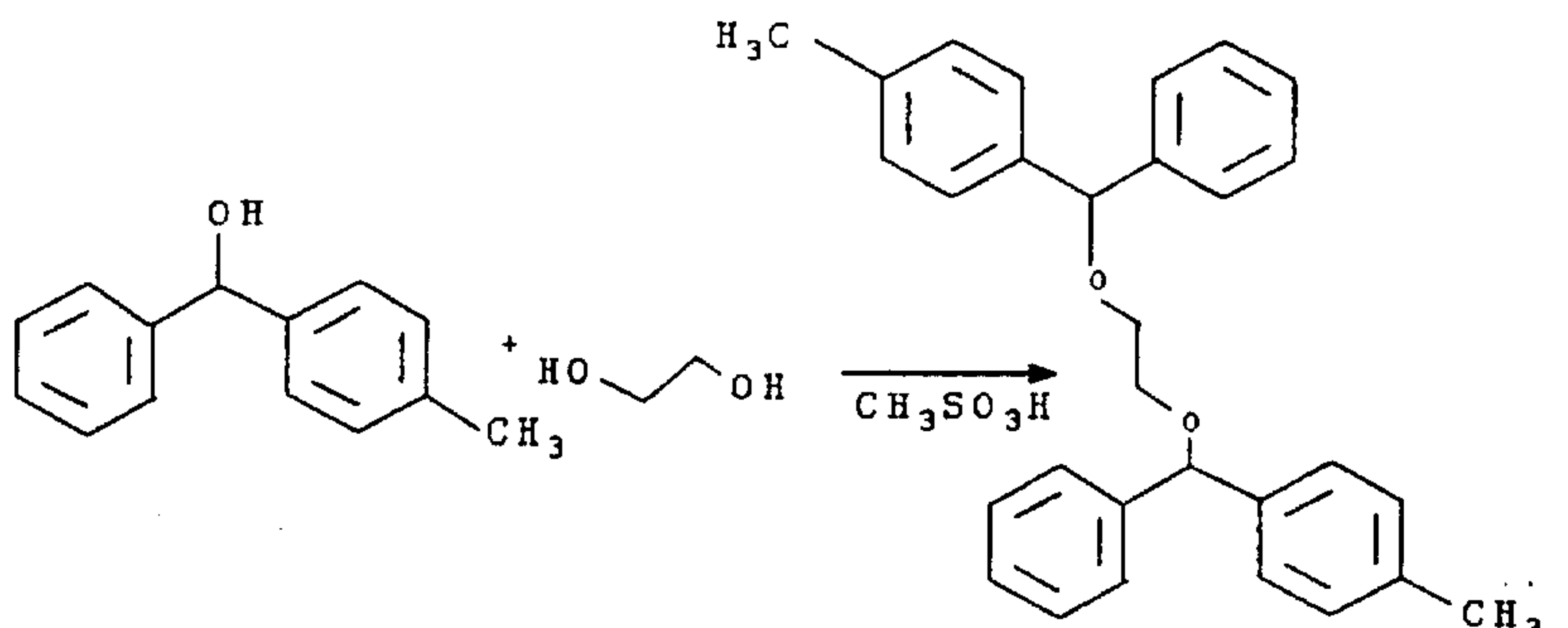
Preparation of 4-Methylbenzohydrol:



Sodium borohydride (1.06 g, 0.028 mol) was added in one portion to a stirred solution of 4-methylbenzophenone (10 g, 0.051 mol) in 25 mL of ethanol. The reaction mixture became warm, and the vessel was immersed in a water bath. After 30 min, the reaction mixture was diluted with 100 mL of water and extracted with 2 X 50 mL of dichloromethane. The combined organic phases were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated at aspirator pressure to yield 8.7 g of 4-methylbenzohydrol. This was used without further purification.

Step 2

Preparation of 1,2-Bis(4-methylphenyl phenylmethoxy)ethane:

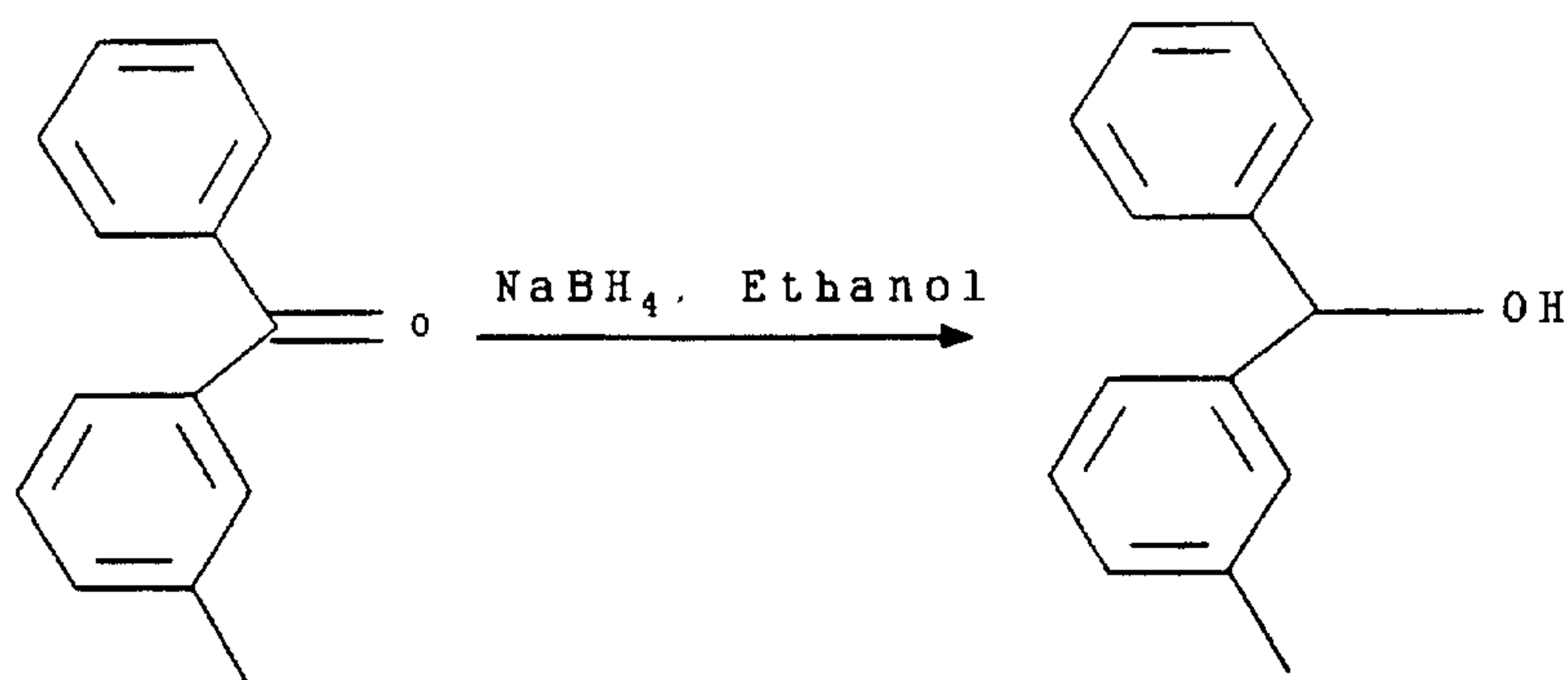


4-Methylbenzohydrol (8.7 g, 0.044 mol) and ethylene glycol (30 mL, 0.53 mol) were combined in a flask equipped with a magnetic stirrer. The mixture was warmed on a steam bath until it became homogeneous, and then methanesulfonic acid (2 mL, 0.030 mol) was added in one portion. The mixture immediately became cloudy, and thin layer chromatographic analysis (TLC) indicated the absence of starting alcohol. The mixture was cooled to room temperature, diluted with 100 mL of water, and extracted with 2 X 50 mL of dichloromethane. The combined organic layers were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The residue was dissolved in hexanes, and the resulting solution was applied to 100 g of flash silica. The column was eluted with 4 X 100 mL each of 5% dichloromethane in hexanes, 10% dichloromethane in hexanes, 20% dichloromethane in hexanes, 50% dichloromethane in hexanes, and dichloromethane. The product was eluted in the 3rd through 8th fractions. The fractions were combined, concentrated, and dissolved in hexanes, from which the product crystallized. The collected solid had a melting point of 107-116°, and weighed 1.9 g (20%).

Synthesis Example 2 - 1,2-Bis[(3-methylphenyl)phenylmethoxy]ethane:

Step 1

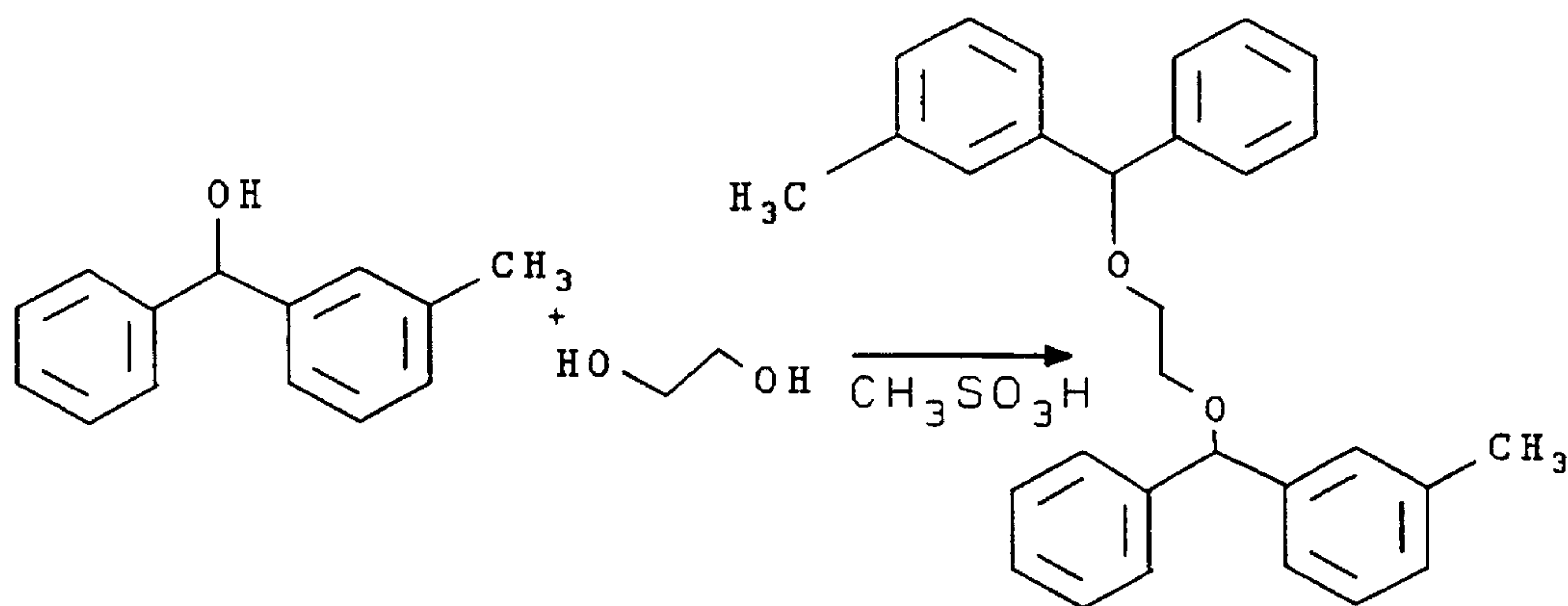
Preparation of 3-Methylbenzohydrol:



Sodium borohydride (1.06 g, 0.028 mol) was added in one portion to a stirred solution of 3-methylbenzophenone (10 g, 0.051 mol) in 25 mL of ethanol. The reaction mixture became warm, and the vessel was immersed in a water bath. After 40 min, the reaction mixture was diluted with 100 mL of water and
 5 extracted with 2 X 50 mL of dichloromethane. The combined organic phases were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated at aspirator pressure to yield 9.1 g of 3-methylbenzohydrol. This was used without further purification.

Step 2

10 Preparation of 1,2-Bis[(2-methylphenyl)phenylmethoxy]ethane:

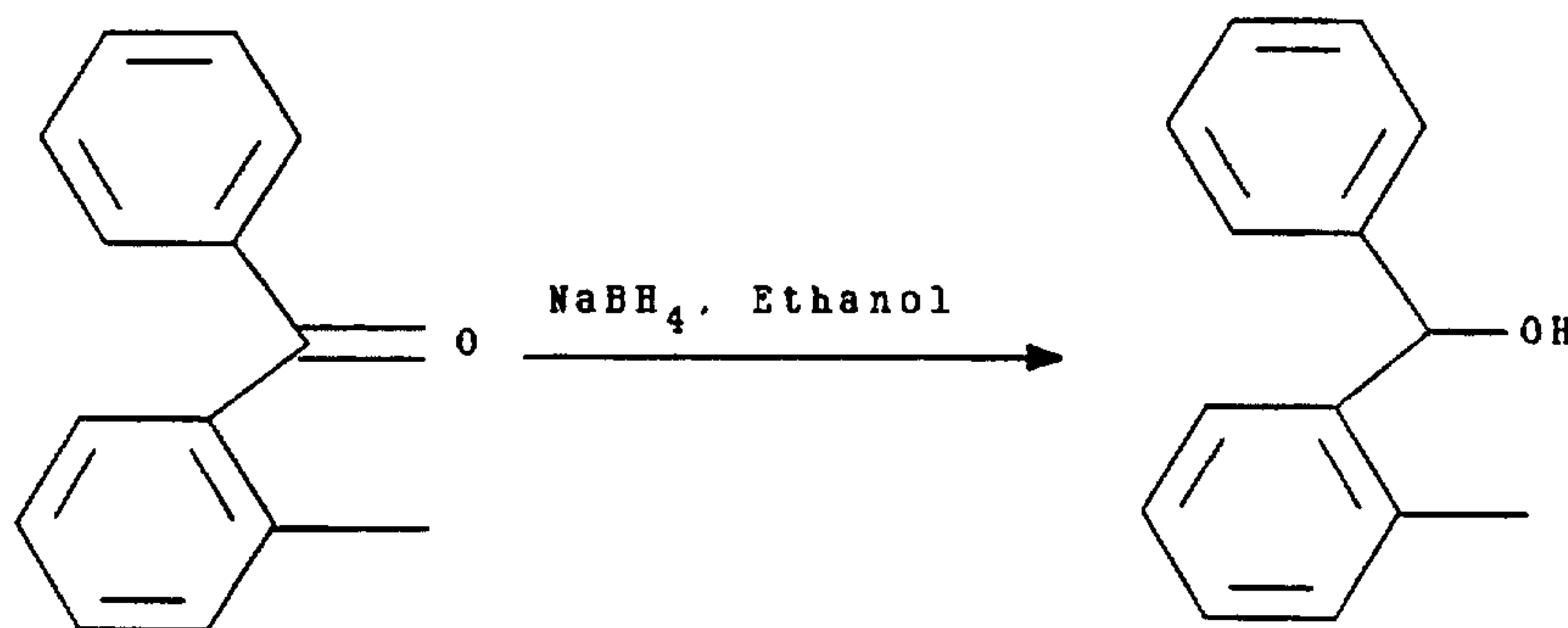


3-Methylbenzohydrol (9.1 g, 0.046 mol) and ethylene glycol (30 mL, 0.53 mol) were combined in a flask equipped with a magnetic stirrer. The mixture was warmed on a steam bath until it became homogeneous, and then methanesulfonic acid (2 mL, 0.030 mol) was added in one portion. The mixture immediately became dark and clouded, and thin layer chromatographic analysis (TLC) indicated the absence of starting alcohol. The mixture was diluted with 200 mL of water and extracted with 2 X 50 mL of dichloromethane. The combined organic layers were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The residue (9.1 g) was dissolved in hexanes, and the resulting solution was applied to 100 g of flash silica. The column was eluted with 4 X 100 mL each of hexanes, 10% dichloromethane in hexanes, 20% dichloromethane in hexanes, 50% dichloromethane in hexanes, and dichloromethane. The product was eluted in the 8th through 15th fractions. The combined fractions were concentrated to yield 3.0 g of an oil.

Synthesis Example 3 - 1,2-Bis[2-methylphenyl(phenyl)methoxy]ethane:

Step 1

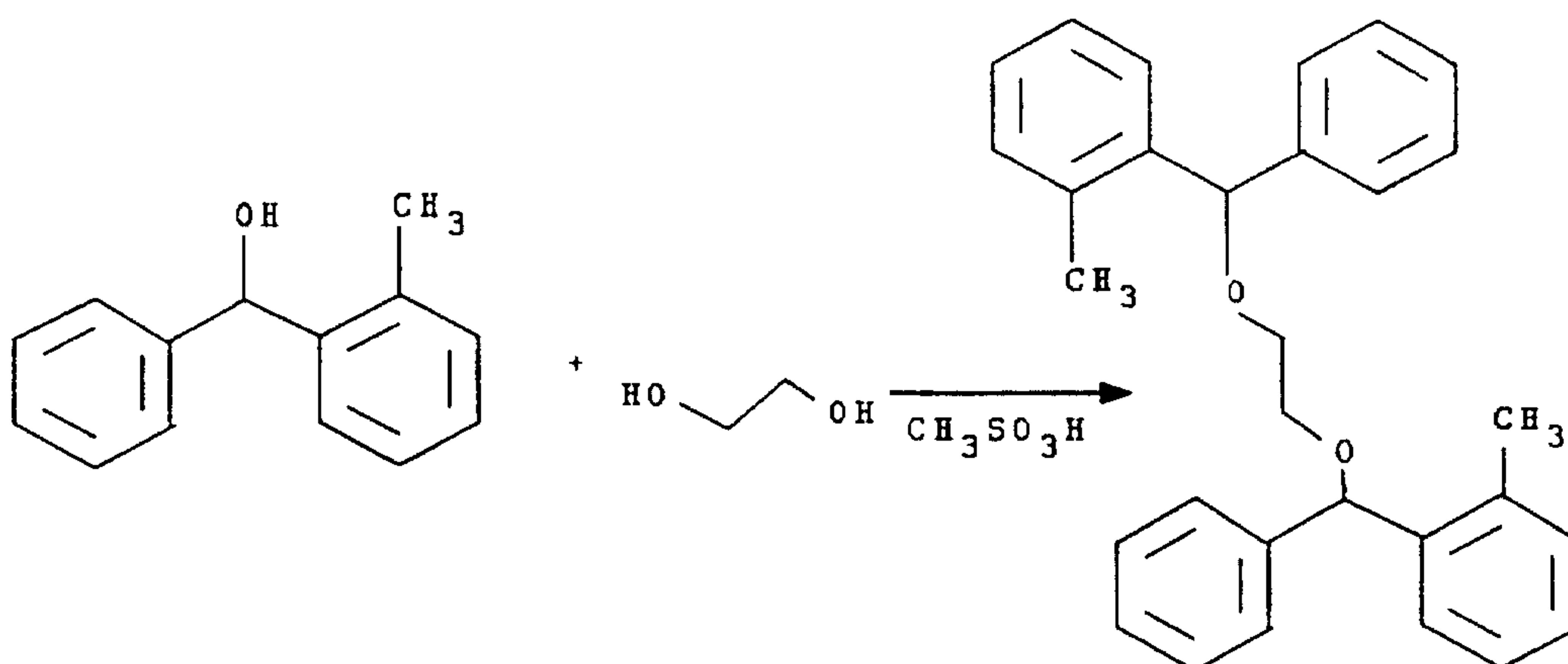
Preparation of 2-Methylbenzohydrol:



Sodium borohydride (2.1 g, 0.056 mol) was added in one portion to a stirred solution of 2-methylbenzophenone (20 g, 0.10 mol) in 50 mL of ethanol. The reaction mixture became warm, and the vessel was immersed in a water bath. After 40 min, the reaction mixture was diluted with 100 mL of water and extracted with 2 X 50 mL of dichloromethane. The combined organic phases were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated at aspirator pressure to yield 19.4 g of 2-methylbenzohydrol. This was used without further purification.

Step 2

Preparation of 1,2-Bis[2-methylphenyl(phenyl)methoxy]ethane:



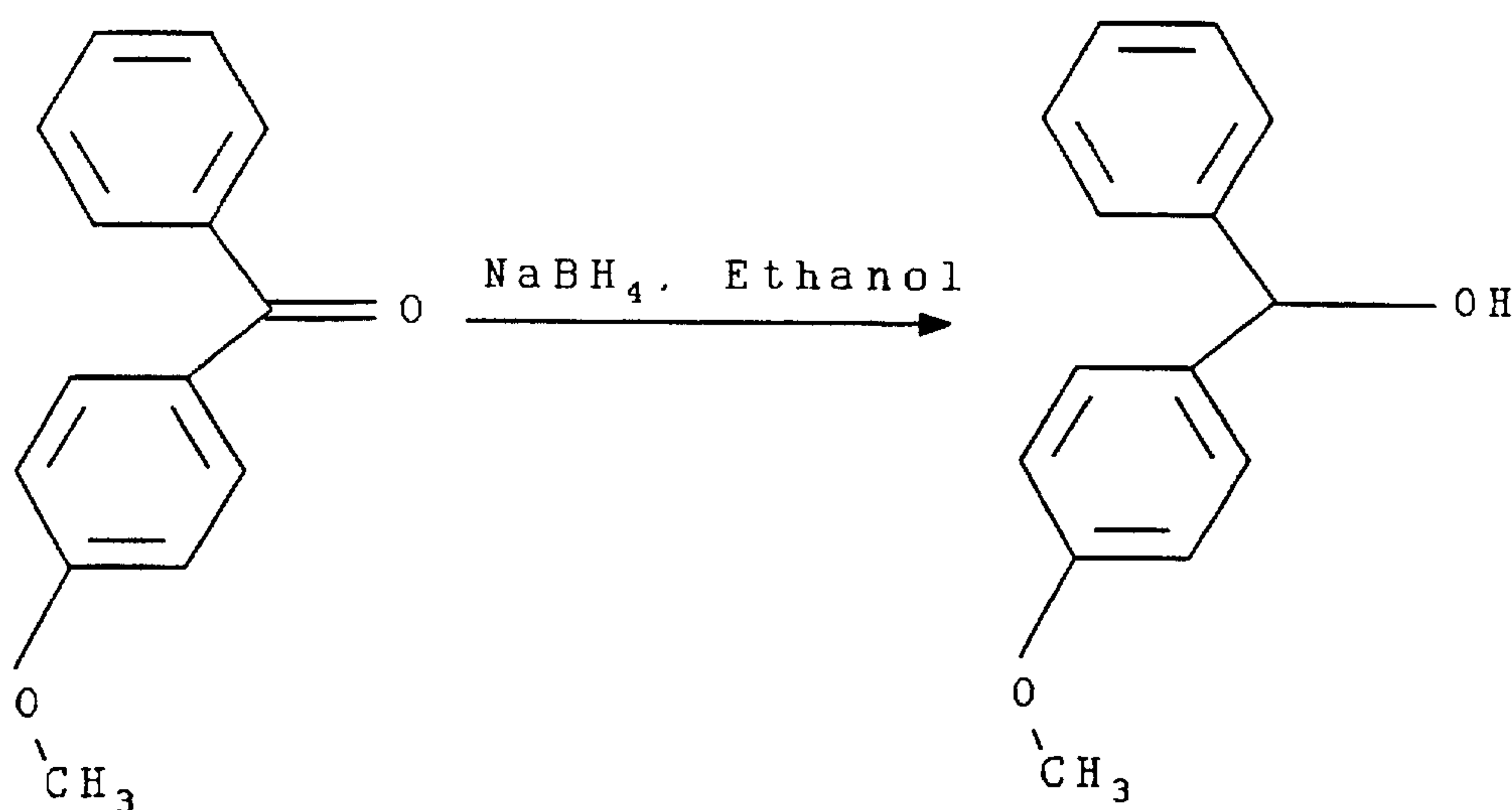
2-Methylbenzohydrol (19.4 g, 0.098 mol) and ethylene glycol 60 mL (1.0 mol) were combined in a flask equipped with a magnetic stirrer, and the mixture was warmed on a steam bath until the starting material dissolved. Methanesulfonic acid (3.9 mL, 0.061 mol) was then added in one portion. Thin layer chromatographic analysis (TLC) indicated the absence of starting alcohol at this point. The mixture was cooled to room temperature, was diluted with 100 mL of water, and was extracted with 2 X 50 mL of dichloromethane. The combined organic layers were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The residue (9.1 g) was dissolved in hexanes, and the resulting solution was applied to 150 g of flash

silica. The column was eluted with 4 X 100 mL each of 5% dichloromethane in hexanes, 10% dichloromethane in hexanes, 20% dichloromethane in hexanes, 50% dichloromethane in hexanes, and dichloromethane. The product was eluted in the 5th through 8th fractions. The combined fractions were concentrated to yield 8 g of material. This was recrystallized from hexanes, yielding 7.35 g (35%) of product having a melting point of 105-109°.

Synthesis Example 4 - 1,2-Bis(4-methoxyphenyl(phenyl)methoxy)ethane:

Step 1

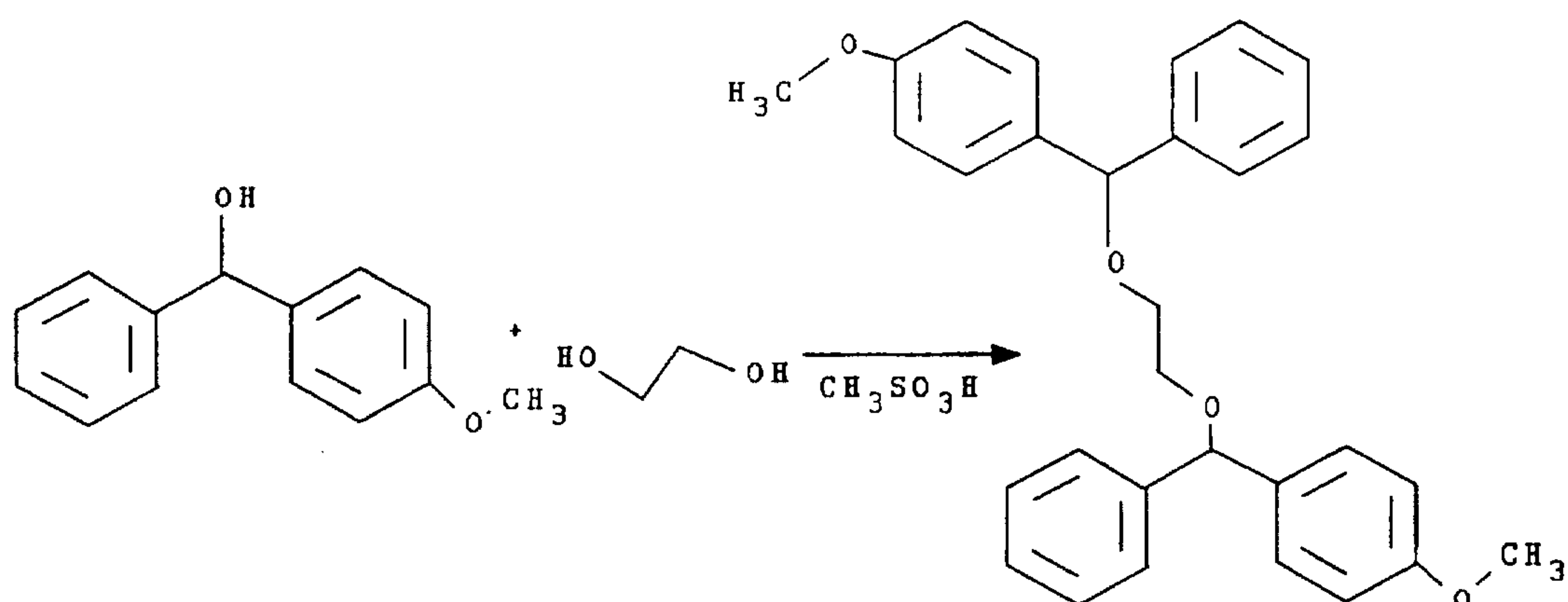
Preparation of 4-Methoxybenzohydrol:



Sodium borohydride (1.96 g, 0.052 mol) was added in one portion to a stirred solution of 4-methoxybenzophenone (20 g, 0.094 mol) in 50 mL of ethanol. The reaction mixture became warm, and the vessel was immersed in a water bath. After 40 min, the reaction mixture was diluted with 100 mL of water and extracted with 2 X 50 mL of dichloromethane. The combined organic phases were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated at aspirator pressure to yield 20 g of 4-methoxybenzohydrol. This was used without further purification.

Step 2

Preparation of 1,2-Bis(4-methoxyphenyl(phenyl)methoxy)ethane:

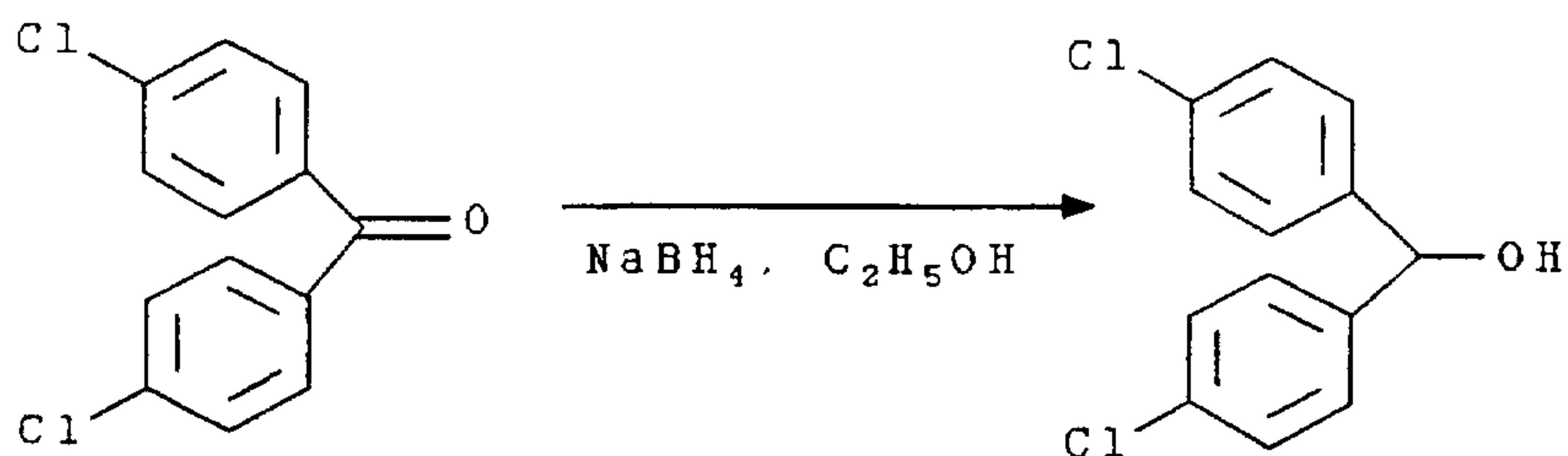


4-Methoxybenzohydrol (20 g, 0.093 mol) and ethylene glycol (60 mL, 1.1 mol) were combined in a flask equipped with a magnetic stirrer, and the mixture was warmed on a steam bath until the starting material dissolved. When methanesulfonic acid (3.9 ml, 0.061 mol) was added in one portion, the reaction mixture rapidly became biphasic. Thin layer chromatographic analysis (TLC) indicated the absence of starting alcohol. The mixture was cooled to room temperature, was then diluted with 100 mL of water, and was extracted with 3 X 50 mL of dichloromethane. The combined organic layers were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The residue was dissolved in hexanes and applied to 120 g of flash silica. The column was eluted with 18 X 100 mL of 1:1 hexanes:dichloromethane. Pure product was eluted in the 3rd through 7th fractions, while a mixture of the product and 2-(4-methoxyphenyl(phenyl)methoxy)ethanol were eluted in the 8th through 12th fractions. The latter were combined and reapplied to a column of the same size. The product was eluted in the first fraction. The combined product fractions were concentrated and maintained at high vacuum with gentle warming, yielding 14 g (66%) of material.

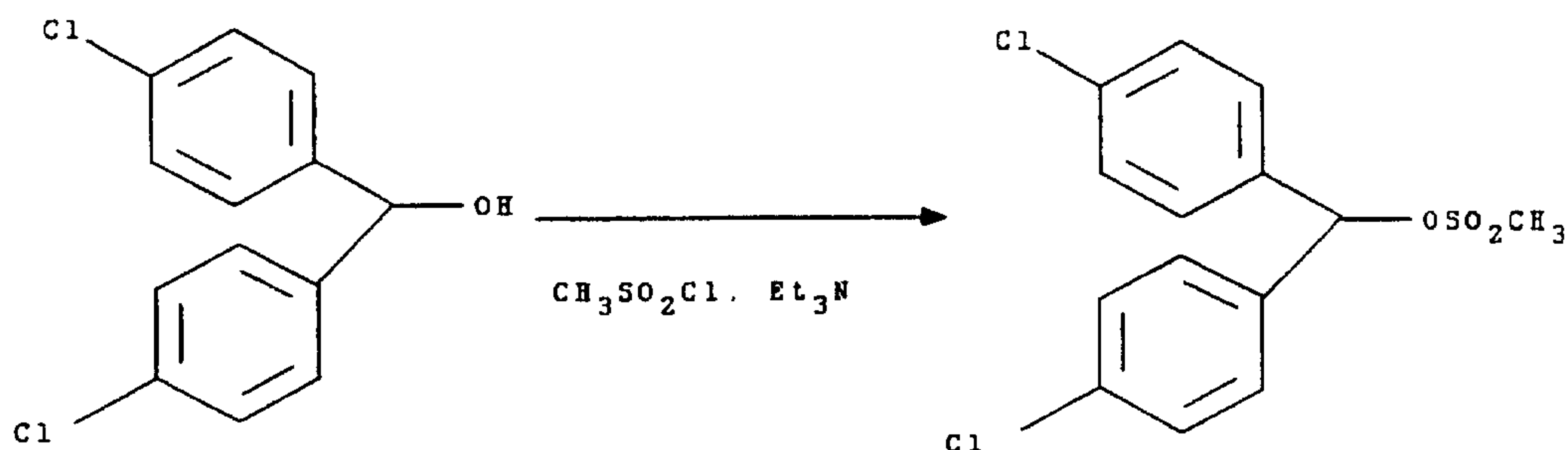
Synthesis Example 5 - 1,2-Bis[di(4-chlorophenyl)methoxy]ethane:

Step 1

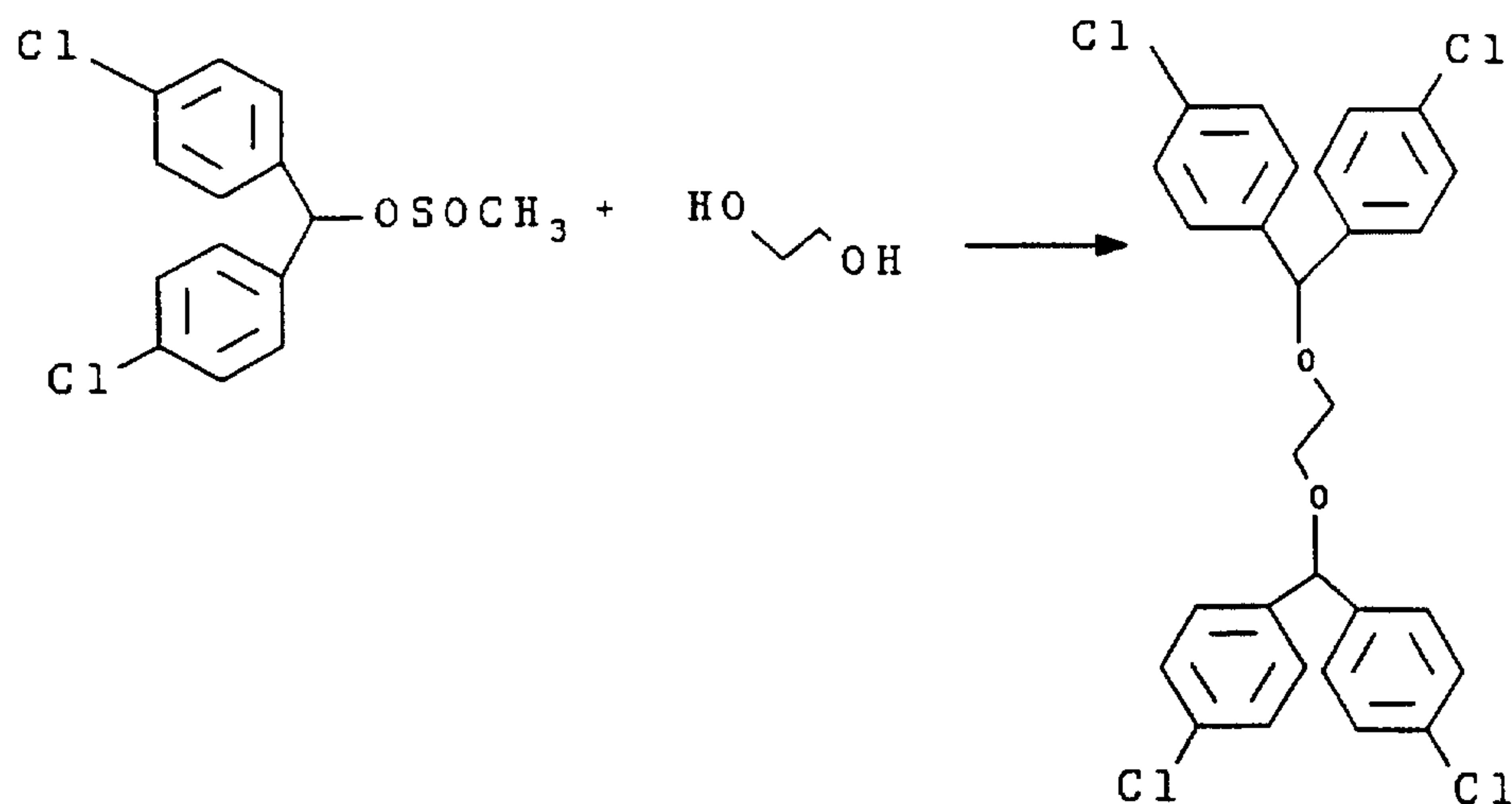
Preparation of 4,4'-Dichlorobenzohydrol:



5 A suspension of 4,4'-dichlorobenzophenone (5.02 g, 20 mmol) in ethanol (15 mL) was treated with sodium borohydride (0.42 g, 11 mmol) added in one portion. The material went into solution, and then a precipitate formed. After 30 min, the reaction was complete by TLC (dichloromethane). The reaction mixture was diluted with 25 mL of water and extracted with dichloromethane (2 x 50 mL). The dichloromethane solution was washed with water (2 x 25 mL) and brine (2 x 25 mL) and then evaporated under reduced pressure to give 5.1 g (100%) of pure 4,4'-dichlorobenzohydrol.

Step 2

5 A stirred solution of 4,4'-dichlorobenzohydrol (5 g, 19.8 mmol) and triethylamine (2.21 g, 21.7 mmol) in ether (50 mL) was cooled in an ice bath, and methanesulfonyl chloride was added over 10 min. The reaction was continued for 1 hr. Two-thirds of the reaction mixture was filtered to remove the triethylamine hydrochloride, and the ether solution was used directly in Step 3.

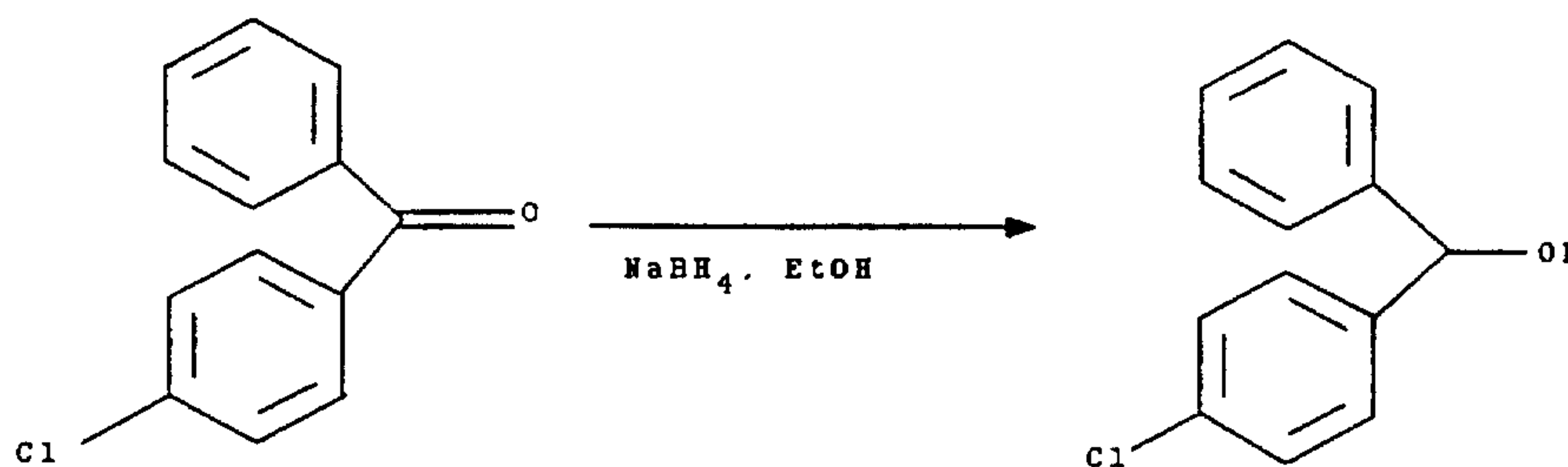
Step 3

The ether solution of 4,4-dichlorobenzohydrol methanesulfonate (about 4.3 g, 13 mmol) from Step 1 was treated with ethylene glycol (0.40 g, 6.5 mmol) and ethyldiisopropylamine (1.7 g, 13 mmol). The solution was evaporated to dryness on the steam bath, and the residue was partitioned between ether and water. The layers were separated and the ether solution was washed successively with 1N citric acid, aqueous sodium bicarbonate, water and brine. The ether was evaporated, and the residue was crystallized from ethanol to give pure 1,2-bis[di(4-chlorophenyl)methoxy]ethane, having a melting point of 98-102°.

Synthesis Example 6 - 1-(4-Chlorophenylphenylmethoxy)-2-(diphenylmethoxy)ethane:

Step 1

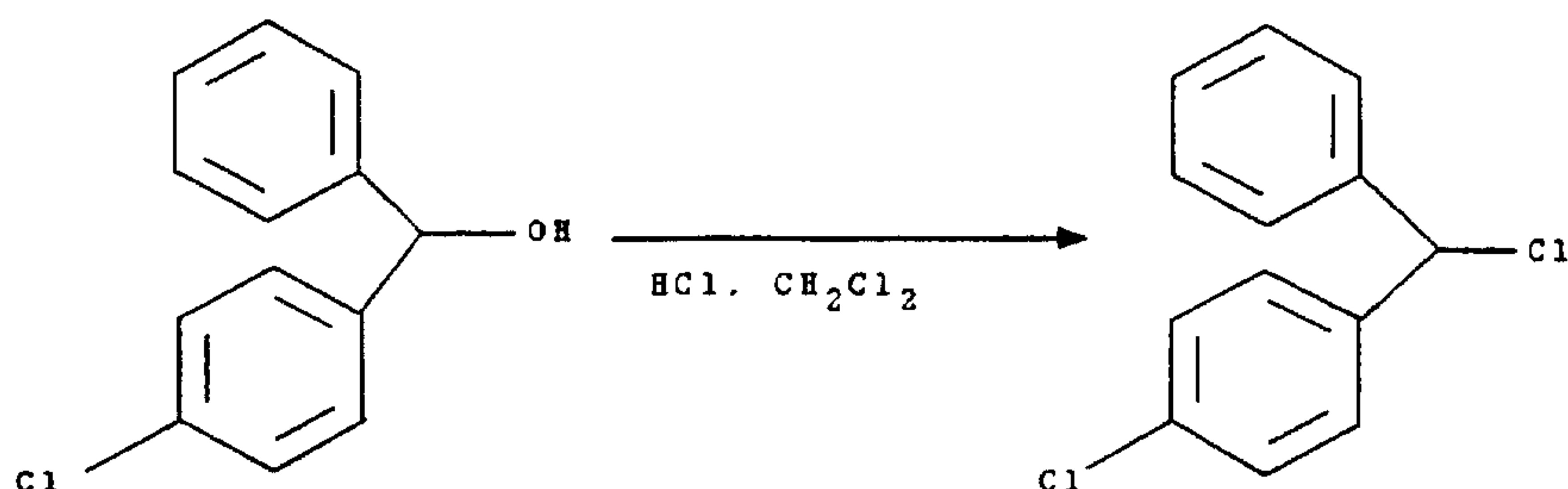
Preparation of 4-Chlorobenzohydrol:



A stirred solution of 4-chlorobenzophenone (20 g, 0.092 mol) in 70 mL of ethanol was treated with sodium borohydride (1.9 g, 0.051 mol) added in portions with cooling in a water bath. The reaction was complete as soon as the addition was complete. The reaction mixture was diluted with water (500 mL) and extracted with dichloromethane (3 x 50 mL). The dichloromethane solution was washed with water (3 x 50 mL), filtered through PS 1 paper and evaporated under reduced pressure to give 20 g (100 %) of 4-chlorobenzohydrol.

Step 2

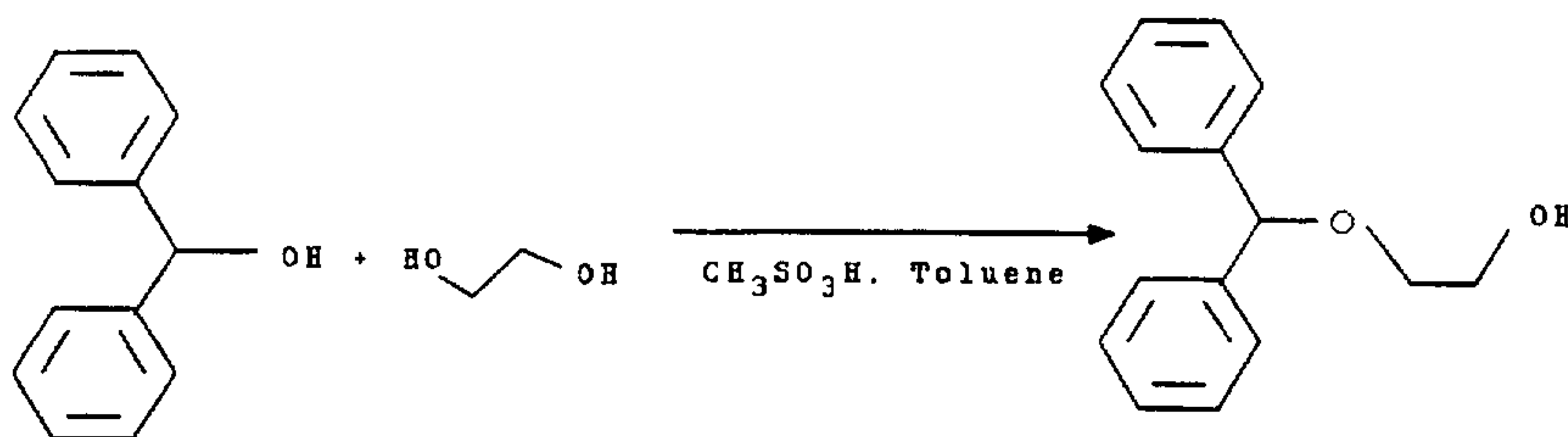
Preparation of 4-Chlorobenzhydryl chloride



5 A solution of 4-chlorobenzhydrol (10 g, 45.8 mmol) in dichloromethane (20 ML) was cooled in an ice bath, and concentrated hydrochloric acid (23 mL, 0.458 mol) was added slowly. The resulting mixture was stirred in the ice bath for 1 hr and then overnight at room temperature. After stirring overnight, a trace of starting material remained as indicated by TLC and the layers were separated and the dichloromethane layer was stirred vigorously for 90 min with fresh concentrated hydrochloric acid (23 mL). The layers were separated and the
10 dichloromethane solution was washed twice with brine (2 x 50 mL). The dichloromethane solution was dried over sodium sulfate and evaporated under reduced pressure to give 9.2 g (85%) of 4-chlorobenzhydryl chloride as an oil.

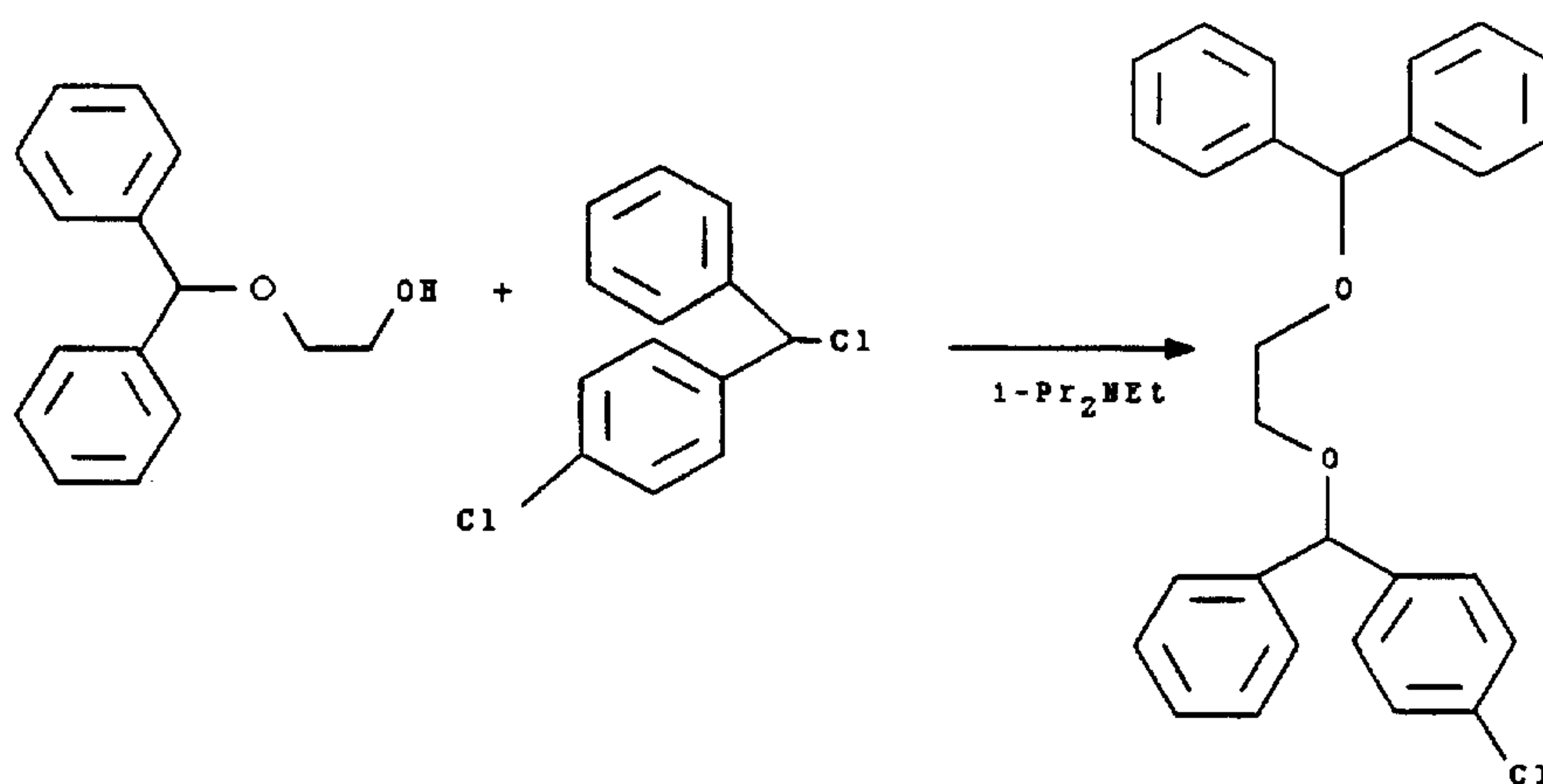
Step 3

Preparation of 2-(Diphenylmethoxy)ethanol:



A 250 mL round bottomed flask was charged with ethylene glycol (60 ml, 1.08 mol), toluene (30 mL), and methanesulfonic acid (5.18 g, 54 mmol). The mixture was heated on the steam bath, and a solution of benzohydrol (10 g, 54 mmol) in toluene (70 mL) was added over 1 hr. The reaction was complete at the end of the addition as indicated by TLC (dichloromethane). The cooled reaction mixture was treated with saturated sodium bicarbonate solution, and the layers were separated. The aqueous layer was extracted with dichloromethane (2 x 50 mL). The combined organic layers were washed with water (3 x 50 mL) and the solvent was evaporated. The crude product was flash chromatographed to give 7.0 g (57%) of 2-(diphenylmethoxy)ethanol.

Step 4



In a 50 mL round bottomed flask were placed 2-diphenylmethoxyethanol (5.4 g, 24 mmol), 4-chlorobenzhydryl chloride (3.76 g, 15.9 mmol) and

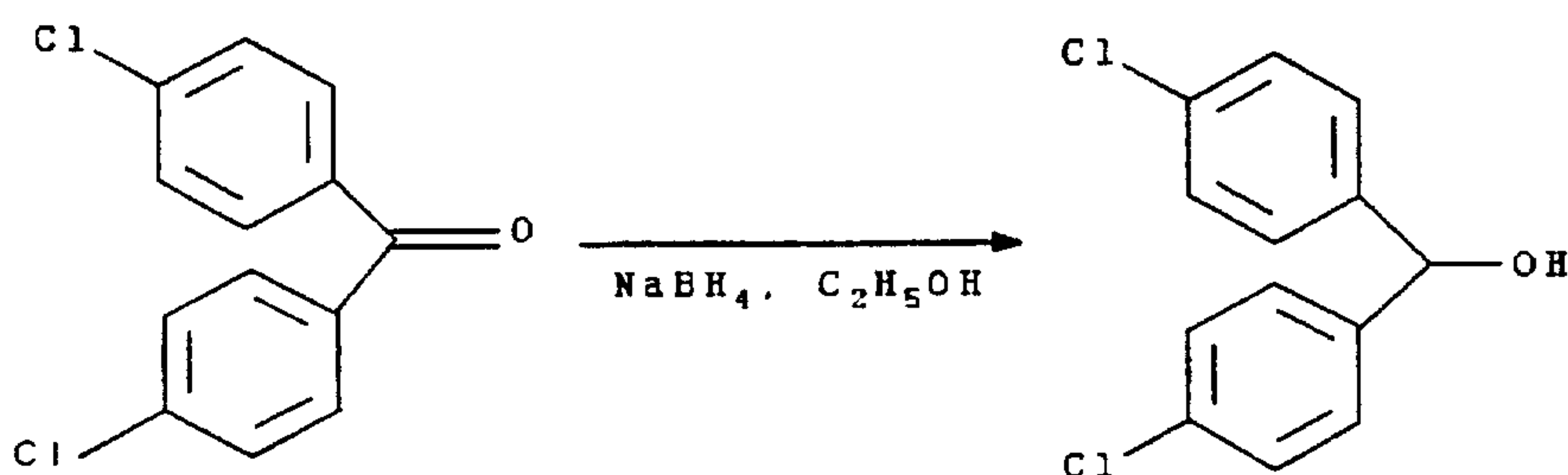
ethyldiisopropylamine (2.05 g, 15.9 mmol). The flask was heated on the steam bath and samples were removed for nmr analysis. When the nmr spectrum of the reaction mixture indicated complete reaction, the mixture was cooled, diluted with dichloromethane, and washed with water (3 x 50 mL). The organic solution was filtered through 1PS filter paper and evaporated to give 9 g of crude product. The material was separated by flash chromatography to give 1.2 g of 1-di(4-chlorophenylphenylmethoxy)-2-(diphenylmethoxy)-ethane.

Synthesis Example 7 - 1-(4,4'-Dichlorophenylmethoxy)-3-(diphenylmethoxy)propane:

10

Step 1

Preparation of 4,4'-Dichlorobenzohydrol:

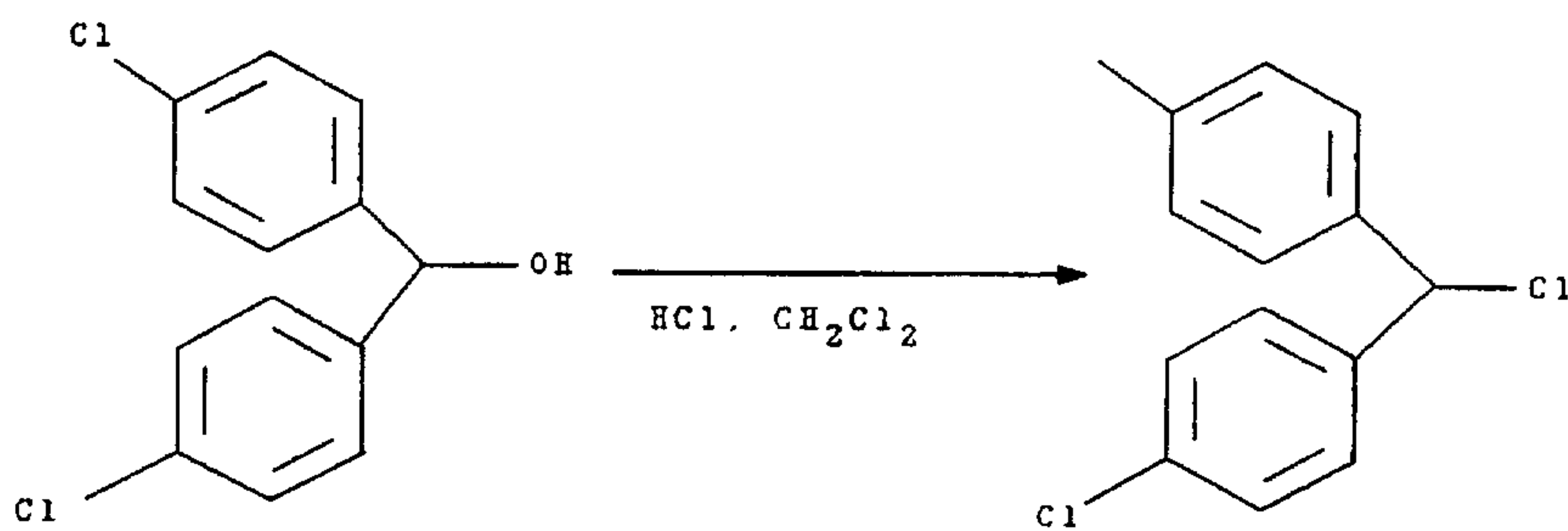


A suspension of 4,4'-dichlorobenzophenone (5.02 g, 20 mmol) in ethanol (15 mL) was treated with sodium borohydride (0.42 g, 11 mmol) added in one portion. The material went into solution, and then a precipitate formed. After 30 min, the reaction was complete by TLC (dichloromethane). The reaction mixture was diluted with 25 mL of water and extracted with dichloromethane (2 x 50 mL). The dichloromethane solution was washed with water (2 x 25 mL) and brine (2 x 25 mL) and then evaporated under reduced pressure to give 5.1 g (100%) of pure 4,4'-dichlorobenzohydrol.

20

Step 2

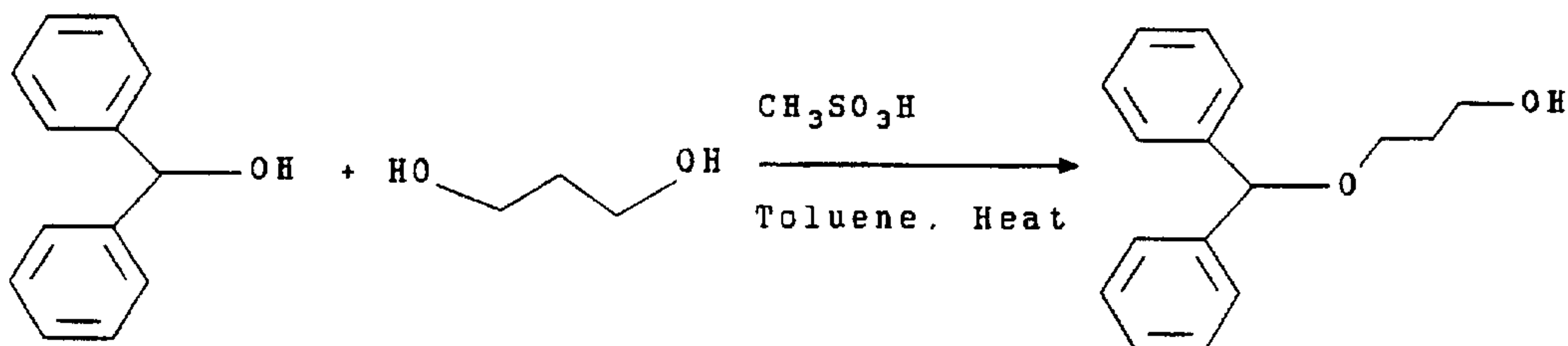
Preparation of 4,4'-Dichlorobenzhydryl chloride:



5 A 125 mL portion of concentrated hydrochloric acid was added dropwise over 15 min to a stirred solution of 4,4'-dichlorobenzohydrol (38 g, 0.15 mol) in 150 mL of dichloro-methane. The mixture was stirred overnight at room temperature. At this point, TLC analysis (dichloromethane) indicated incomplete reaction, and an additional 25 mL of hydrochloric acid were added. After an additional 8 hr, the layers were separated, and the organic layer was washed with 200 mL of water and 200 ml of saturated aqueous sodium chloride, was filtered through Whatman 1PS paper, and was concentrated at aspirator pressure to give 45 g of a solid. The solid was collected with the aid of 100 mL of cold methanol, washed with 25 mL of cold methanol, and air dried. The product weighed 24 g. A second crop of 8.0 g was recovered. The combined crops (32 g) represent a 79% yield.

Step 3

Preparation of 3-Diphenylmethoxypropanol:

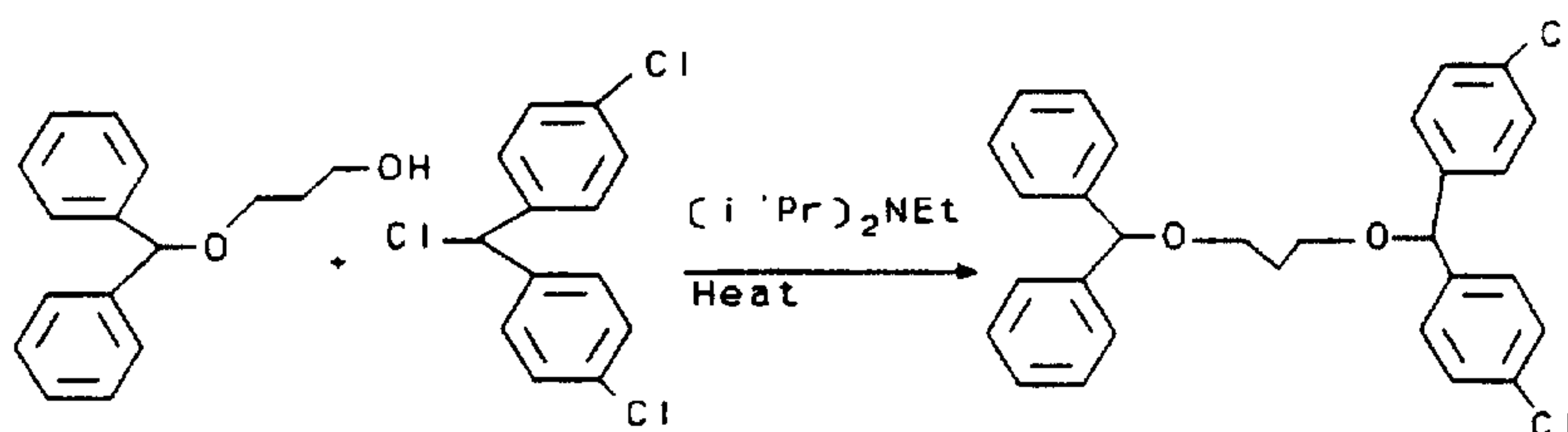


A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,3-propanediol (100 g, 1.31 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene (150 mL). The mixture was immersed in a 130° bath, and benzohydrol (27.6 g, 0.15 mol) dissolved in 250 mL of toluene was added dropwise over 90 min. After an additional 30 min, TLC analysis indicated complete reaction, and the reaction mixture was cooled and diluted with 200 mL of ethyl acetate. The layers were separated, and the diol layer was extracted with an additional 100 mL of ethyl acetate. The combined ethyl acetate layers were washed with 100 mL of water, 100 mL of saturated aqueous sodium bicarbonate, and 100 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 40 g of an oil. The oil was applied to 150 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 3 X 250 mL of 20% ethyl acetate in hexanes, and 5 X 250 mL of 40% ethyl acetate in hexanes. The product was eluted in the 3rd through 8th fractions. The fractions were combined and concentrated to give 30 g of an oil that crystallized upon standing overnight in a refrigerator. The solid was triturated with 100 mL of cold hexanes, collected, washed with 25 mL of cold hexanes, and air dried. The collected product had a melting point of 51-53°, and weighed 25.5 g (70%).

20

Step 4

Preparation of 1-(4,4'-Dichlorophenylmethoxy)-3-(diphenylmethoxy)propane:



25

A mixture of 3-diphenylmethoxypropanol (4.84 g, 0.020 mol), 4,4'-dichlorobenzylchloride (5.43 g, 0.020 mol), and diisopropylethylamine (3.1 g, 0.024 mol) was heated at 130° for 24 hrs. The mixture was then cooled and was diluted with 75 mL of ethyl acetate, washed with 2 X 75 mL of water and 75 mL of saturated aqueous sodium chloride, filtered through Whatman 1PS

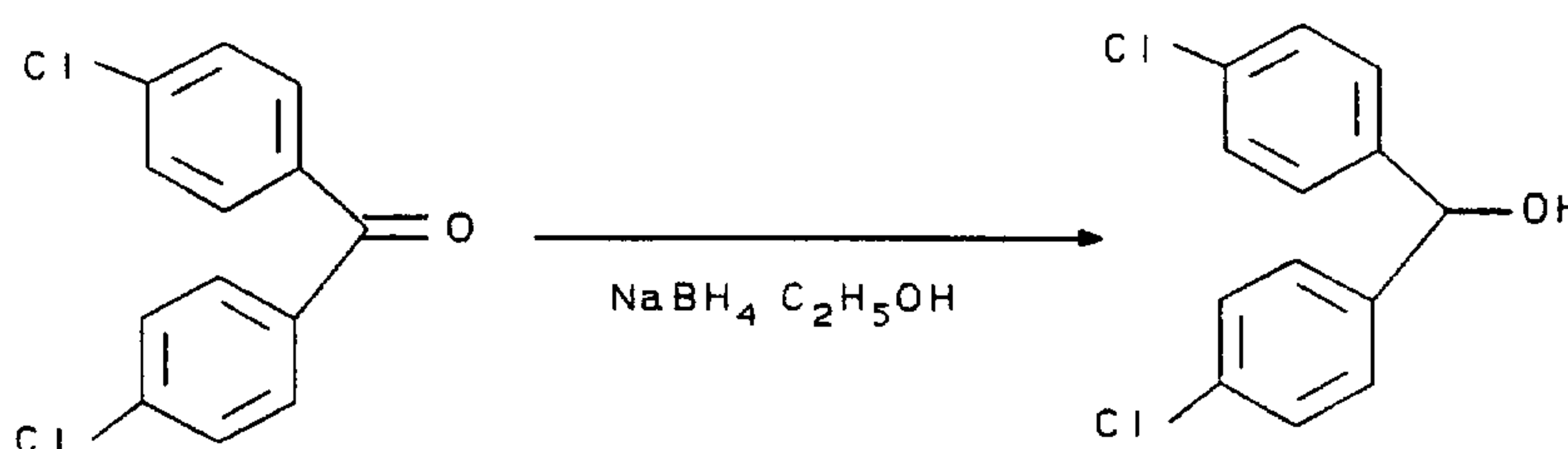
paper, and concentrated at aspirator pressure. The residue was 9.0 g of a brown oil. This was dissolved in hexanes and applied to 75 g of flash silica gel. The column was eluted with 19 X 125 mL of hexanes and 11 X 125 mL of 1% ethyl acetate in hexanes. The product, along with an impurity, was eluted in the 24th and 25th fractions.

The fractions were combined and concentrated to give 4.2 g of an oil. The components could not be separated by elution of a 50 g flash silica gel column with 0.5% ethyl acetate in hexanes. The same sample was applied to a 6 mm "Chromatotron" rotor and eluted with the same solvent. A fraction (2.7 g) enriched in the product was obtained. This was further fractionated on preparative silica gel TLC plates using dichloromethane as eluent. The bands containing the product were scraped from the plate and washed with ethyl acetate. The wash was concentrated to give 2.2 g of the desired product. (The contaminating material was identified as benzophenone). The product was dissolved in hexanes and applied to 25 g of flash silica gel. The column was eluted with 5 X 100 mL of hexanes, 7 X 100 mL of 2% dichloromethane in hexanes, 4 X 125 mL of 5% dichloromethane in hexanes, 4 X 125 mL of 10% dichloromethane in hexanes, and 4 X 125 mL of 25% dichloromethane in hexanes. The product was eluted in the 13th through 22nd fractions. The fractions were combined and concentrated to yield 2.0 g of an oil that displayed only a single spot by TLC analysis.

Synthesis Example 8 - 1-(4,4'-Dichlorophenylmethoxy)-4-(diphenylmethoxy)butane:

Step 1

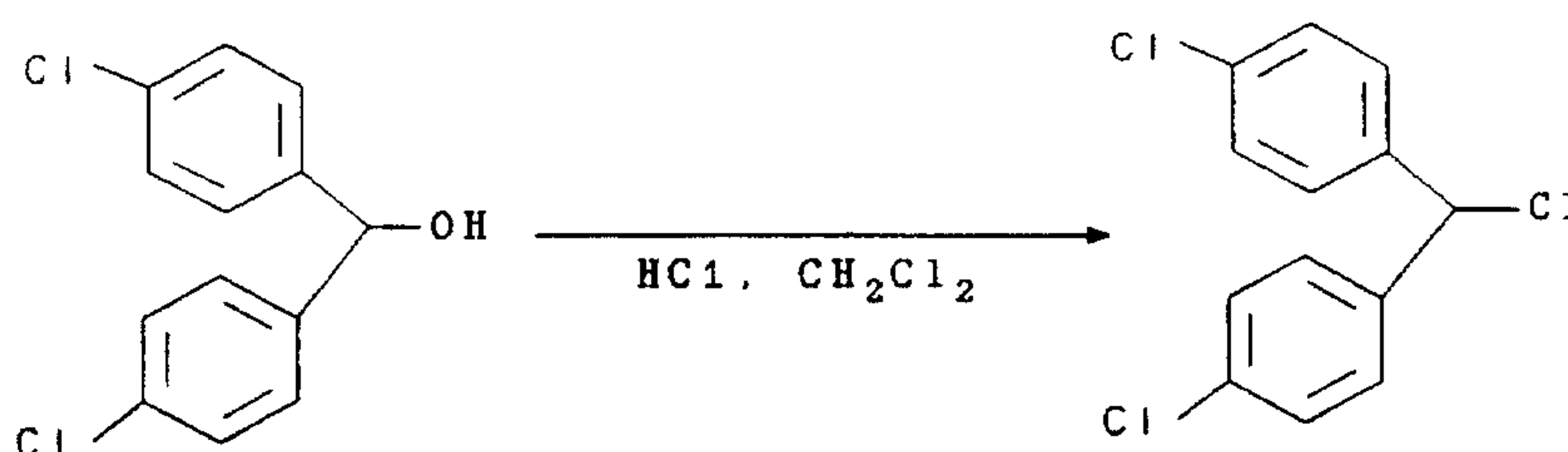
Preparation of 4,4'-Dichlorobenzohydrol:



A suspension of 4,4-dichlorobenzophenone (5.02 g, 20 mmol) in ethanol (15 mL) was treated with sodium borohydride (0.42 g, 11 mmol) added in one portion. The material went into solution and then a precipitate formed. After 30 min, the reaction was complete by TLC (dichloromethane). The reaction mixture
5 was diluted with 25 mL of water and extracted with dichloromethane (2 x 50 mL). The dichloromethane solution was washed with water (2 x 25 mL) and brine (2 x 25 mL) and then evaporated under reduced pressure to give 5.1 g (100%) of pure 4,4'-dichlorobenzohydrol.

Step 2

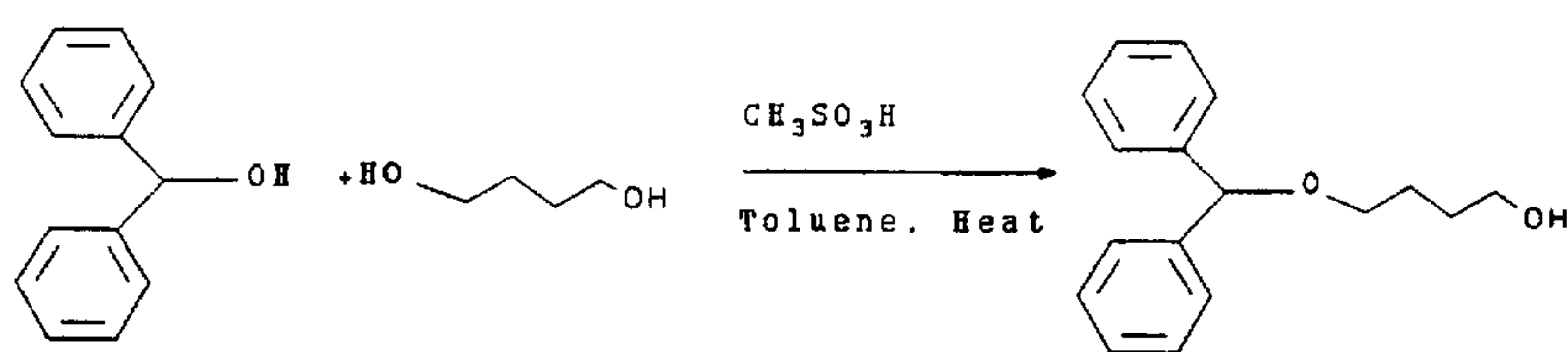
10 Preparation of 4,4'-Dichlorobenzhydryl chloride:



A 125 mL portion of concentrated hydrochloric acid was added dropwise over 15 min to a stirred solution of 4,4'-dichlorobenzohydrol (38 g, 0.15 mol) in 150 mL of dichloromethane. The mixture was stirred overnight at room temperature. At this point, TLC analysis (dichloromethane) indicated incomplete
15 reaction, and an additional 25 mL of hydrochloric acid were added. After an additional 8 hr, the layers were separated, and the organic layer was washed with 200 mL of water and 200 mL of saturated aqueous sodium chloride, was filtered through Whatman 1PS paper, and was concentrated at aspirator pressure to give 45 g of a solid. The solid was collected with the aid of 100 mL of cold
20 methanol, washed with 25 mL of cold methanol, and air dried. The product weighed 24 g. An additional 8.0 g was recovered as a second crop. The combined 32 g represents a 79% yield.

Step 3

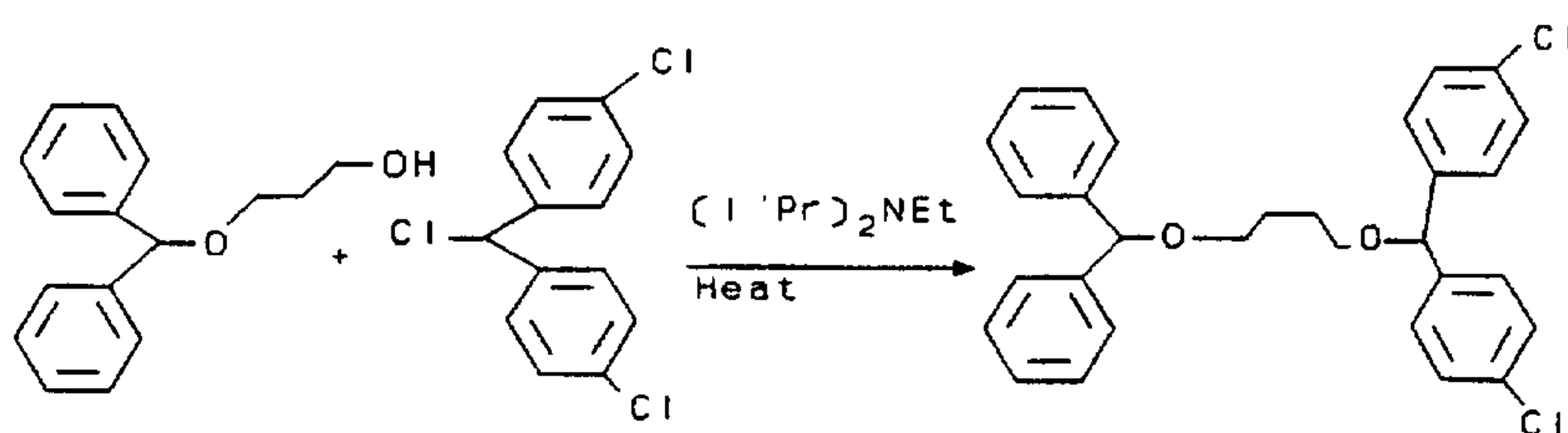
Preparation of 4-Diphenylmethoxybutanol:



A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,4-butanediol (135 g, 1.5 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene 250 mL). The mixture was immersed in a 120° bath, and benzhydrol (27.6 g, 0.15 mol) dissolved in 250 mL of toluene was added dropwise over 75 min. After an additional 1 hr, TLC analysis indicated complete reaction, and the reaction mixture was cooled to room temperature. The layers were separated, and the diol layer was extracted with 100 mL of ethyl acetate. The combined upper phases were washed with 200 mL of water, 200 mL of saturated aqueous sodium bicarbonate, and 200 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 45g of an oil. The oil was applied to 300 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 5 X 250 mL of 20% ethyl acetate in hexanes, and 8 X 250 mL of 40% ethyl acetate in hexanes. The product was eluted in the 8th through 13th fractions. The fractions were combined and concentrated to give 40 g of an oil.

Step 4

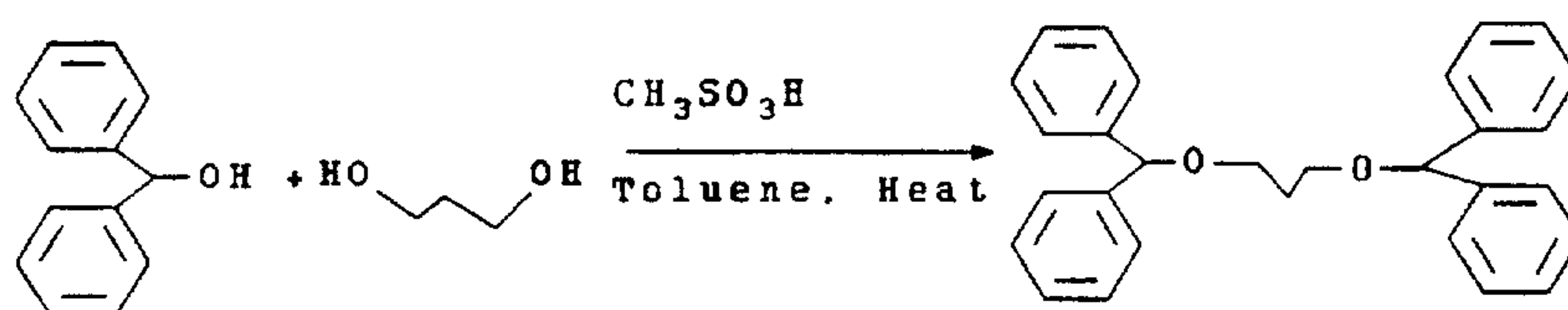
Preparation of 1-(4,4'-Dichlorophenylmethoxy)-4-(diphenylmethoxy)butane:



A mixture of 4-diphenylmethoxybutanol (7.2 g, 0.020 mol), 4,4'-dichlorobenzyl chloride (5.43 g, 0.020 mol), and diisopropylethylamine was heated at 130° for 6 hrs. At this point, TLC analysis (CH₂Cl₂) showed incomplete reaction. Additional alcohol (2.1 g, 0.0082 mol) was added, and the mixture was heated for another 18 hrs. It was then cooled to room temperature

and was diluted with 75 mL of ethyl acetate. This mixture was washed with 2 X 75 mL of water and 75 mL of saturated aqueous sodium chloride, filtered through Whatman 1PS paper, and concentrated *in vacuo* to give 9.9 g of a brown oil. The oil was dissolved in hexanes and applied to 75 g of flash silica gel. The column was eluted with 7 X 125 mL of hexanes and 7 X 125 mL of 5% ethyl acetate in hexanes. The product was eluted in the 9th and 10th fractions. The fractions were combined and concentrated to yield 6.2 g of an oil. This was dissolved in 25 mL of hexanes and 0.10 mL of methanol. Crystallization was initiated by scratching, and the mixture was left in a freezer for 4 hrs. The resulting solid was collected and washed with 10 mL of methanol. After drying, the solid, having a melting point of 57-59°, weighed 4.0 g (41%). An additional 1.2 g (13%) was recovered as a second crop of crystals.

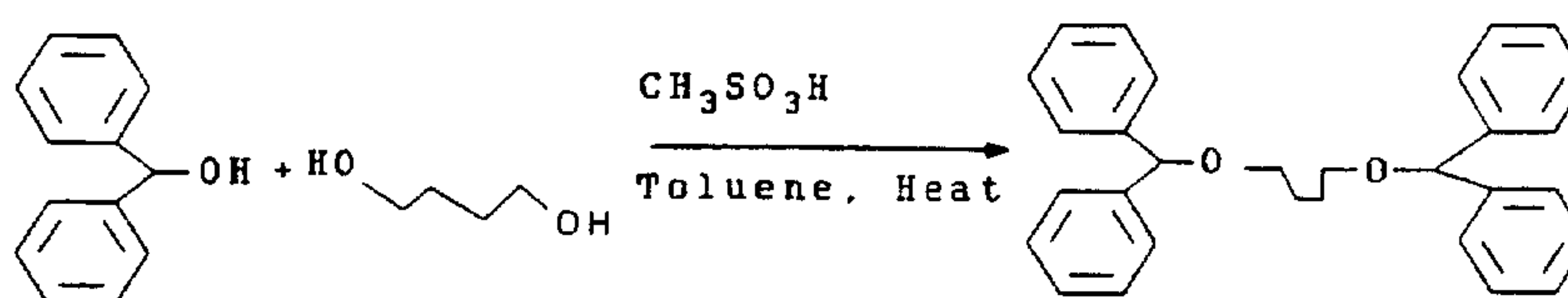
Synthesis Example 9 - 1,3-Bis(diphenylmethoxy)propane:



A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,3-propanediol (100 g, 1.31 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene (150 mL). The mixture was immersed in a 130° bath, and benzhydrol (27.6 g, 0.15 mol) dissolved in 250 mL of toluene was added dropwise over 90 min. After an additional 30 min, TLC analysis indicated complete reaction, and the reaction mixture was cooled and diluted with 200 mL of ethyl acetate. The layers were separated, and the diol layer was extracted with an additional 100 mL of ethyl acetate. The combined ethyl acetate layers were washed with 100 mL of water, 100 mL of saturated aqueous sodium bicarbonate, and 100 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 40 g of an oil. The oil was applied to 150 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 3 X 250 mL of 20% ethyl acetate in hexanes, and 5 X 250 mL of 40% ethyl acetate in hexanes. The product was

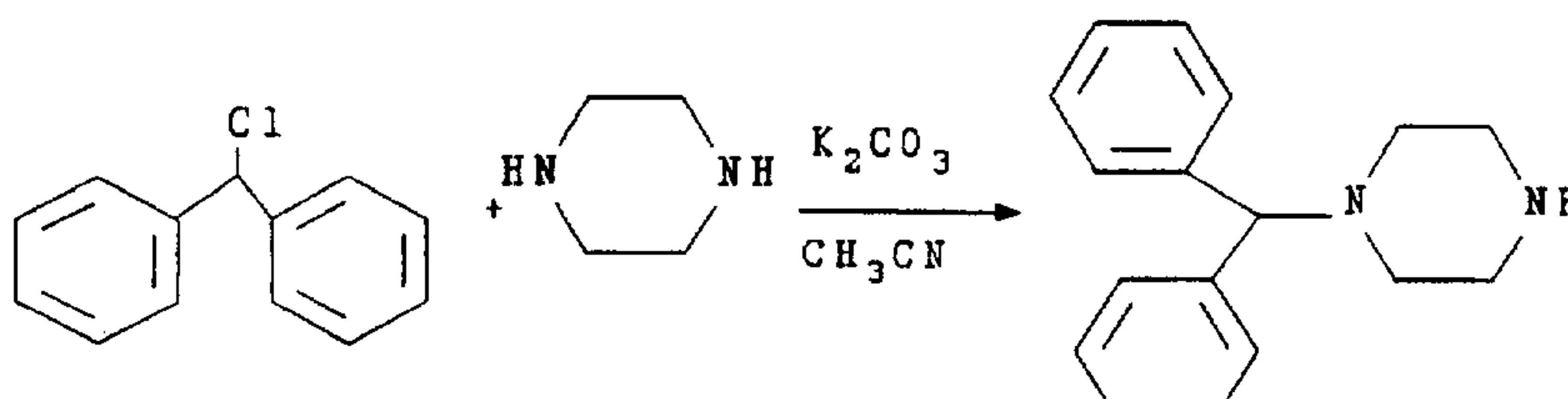
eluted in the 2nd fraction. The 6.2 g of oil that was recovered after concentration was kept under high vacuum for 30 min and was then scratched to initiate crystallization. The resulting solid was collected with the aid of methanol and air dried to give 1, 3-bis(diphenylmethoxy)propane (6.4 g, 10%), having a melting point of 80-82°.

Synthesis Example 10 - 1,4-Bis(diphenylmethoxy)butane:



A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,4-butanediol (135 g, 1.5 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene (250 mL). The mixture was immersed in a 120° bath, and benzhydrol (27.6 g, 0.15 mol) dissolved in 250 mL of toluene was added dropwise over 75 min. After an additional 1 hr, TLC analysis indicated complete reaction, and the reaction mixture was cooled to room temperature. The layers were separated, and the diol layer was extracted with 100 mL of ethyl acetate. The combined ethyl acetate layers were washed with 200 mL of water, 200 mL of saturated aqueous sodium bicarbonate, and 200 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 45g of an oil. The oil was applied to 300 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 5 X 250 mL of 20% ethyl acetate in hexanes, and 8 X 250 mL of 40% ethyl acetate in hexanes. The product was eluted in the 3rd and 4th fractions. The fractions were combined and concentrated to give a crystalline solid that was collected with the aid of some methanol. After drying, the product, having a melting point of 86-89°, weighed 2.0 g (3.1%).

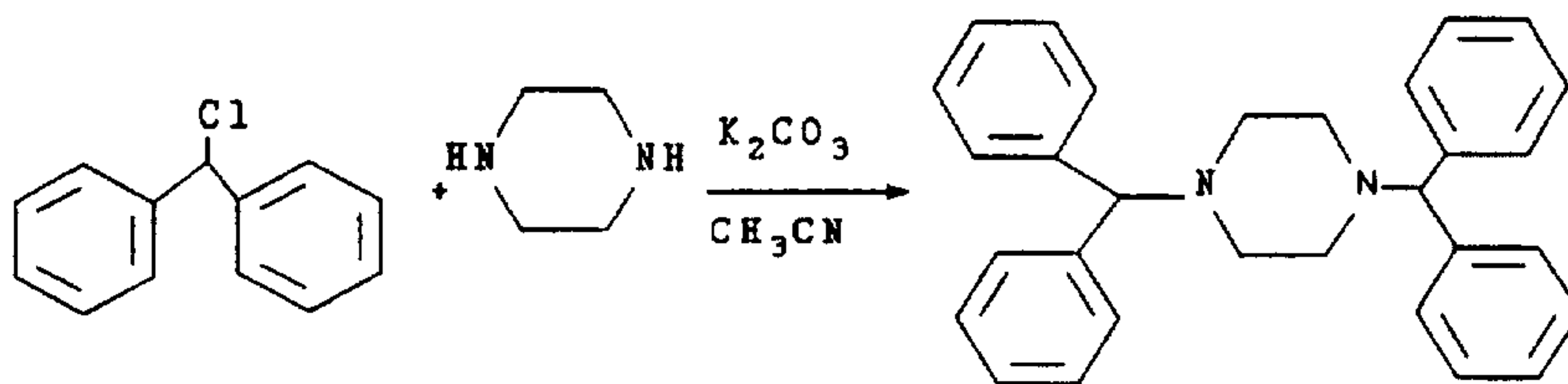
Synthesis Example 11 - 1-Benzhydrylpiperazine:



Benzhydryl chloride (10 g, 0.054 mol) in about 100 mL of acetonitrile was added dropwise to a mixture of piperazine (23.2 g, 0.27 mol) and potassium carbonate (13.8 g, 0.10 mol) in 100 mL of acetonitrile contained in a flask equipped with a magnetic stirrer. The mixture was stirred at room temperature for 20 hrs. At this point, TLC analysis showed no evidence of reaction. After the mixture was heated at reflux for 4 hr, evidence of starting material was absent from the TLC. The reaction mixture was filtered, and the collected solid was washed with about 200 mL of acetonitrile. The combined filtrates were reduced to about 40 mL volume at aspirator pressure and were then mixed with 40 mL of 1 M aqueous sodium hydroxide. The layers were separated, and the aqueous layer was extracted with 3 X 75 mL of dichloro-methane. The combined organic layers were extracted with 3 X 75 mL of 1 M aqueous sodium hydroxide, filtered through Whatman 1PS paper, and concentrated *in vacuo*. After drying for 1 hr at high vacuum, the residue weighed 16 g. This crude product was mixed with diethyl ether, and an insoluble solid was removed by filtration.

The filtrate was concentrated, dissolved in ethyl acetate, and applied to 50 g of flash silica gel. The column was eluted with 5 X 125 mL of 0.5% methanol in ethyl acetate, 4 X 125 mL of 0.5% methanol and 1% triethylamine in ethyl acetate, and 4 X 125 mL of 0.5% methanol and 2% triethylamine in ethyl acetate. The product was eluted in the 3rd through 13th fractions. The combined fractions were concentrated to give 8.5 g of an oil. The oil was mixed with a small volume of hexanes to produce a crystalline solid. The solid was collected with the aid of more hexanes, yielding 7.0 g (52%) of product, having a melting point of 75-77°.

25 *Synthesis Example 12 - 1,4-Bis-benzydrylpiperazine:*

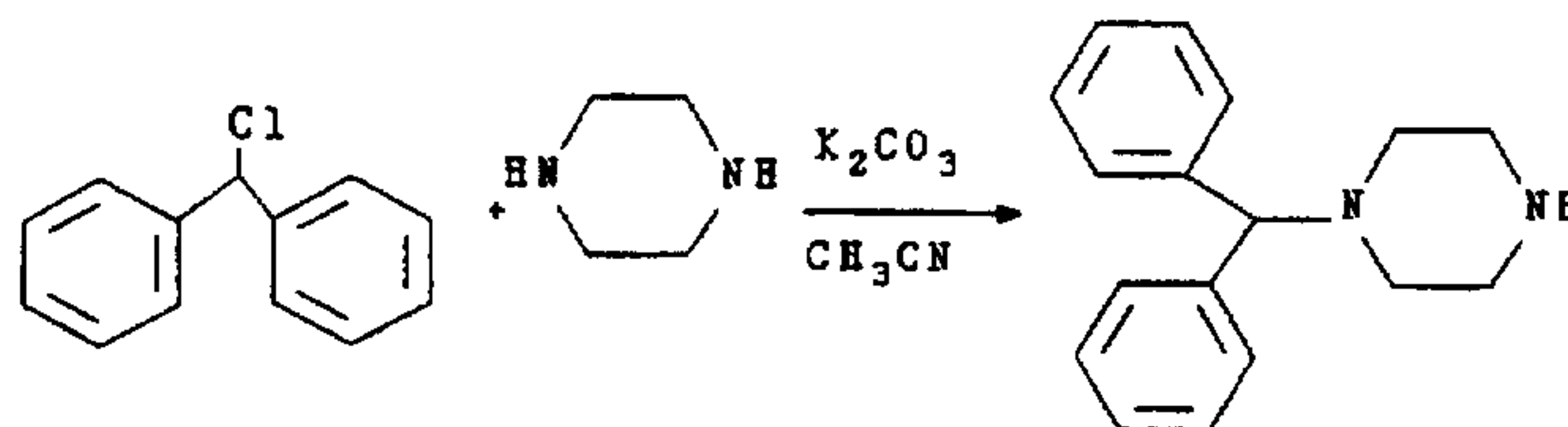


Benzhydryl chloride (10 g, 0.054 mol) in about 100 mL of acetonitrile was added dropwise to a mixture of piperazine (23.2 g, 0.27 mol) and potassium carbonate (13.8 g, 0.10 mol) in 100 mL of acetonitrile contained in a flask equipped with a magnetic stirrer. The mixture was stirred at room temperature for 20 hrs. At this point, TLC analysis showed no evidence of reaction. After the mixture was heated at reflux for 4 hr, evidence of starting material was absent from the TLC. The reaction mixture was filtered, and the collected solid was washed with about 200 mL of acetonitrile. The combined filtrates were reduced to about 40 mL volume at aspirator pressure and were then mixed with 40 mL of 1 M aqueous sodium hydroxide. The layers were separated, and the aqueous layer was extracted with 3 X 75 mL of dichloromethane. The combined organic layers were extracted with 3 X 75 mL of 1 M aqueous sodium hydroxide, filtered through Whatman 1PS paper, and concentrated *in vacuo*. After drying for 1 hr at high vacuum, the residue weighed 16 g. This crude product was mixed with diethyl ether, and an insoluble solid was collected. The solid was recrystallized from tetrahydrofuran to yield 0.10 g (0.44% of theoretical) of product, having a melting point of 255-265°.

Synthesis Example 13 - 1-Benzhydryl-4-[2-(4-trifluoromethylphenyl)phenylmethoxy)ethylpiperazine:

Step 1

Preparation of 1-Benzhydrylpiperazine:



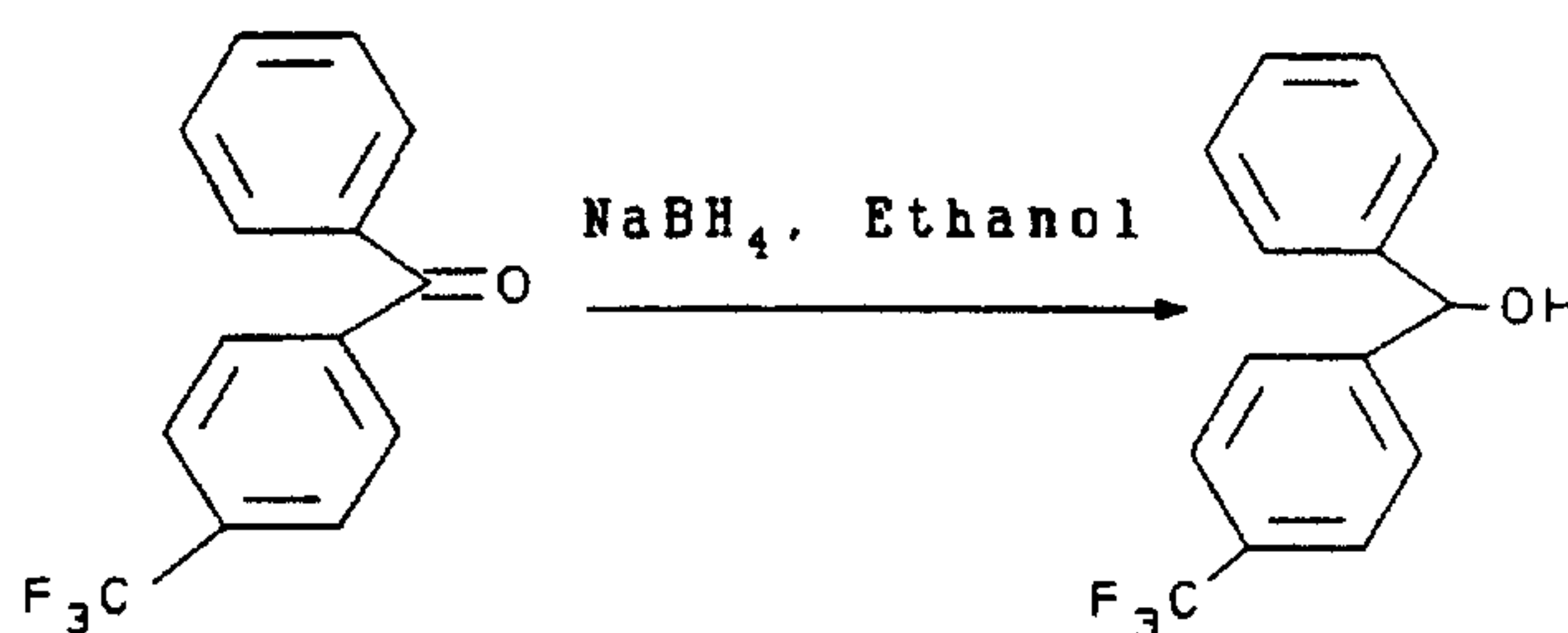
Benzhydryl chloride (10 g, 0.054 mol) in about 100 mL of acetonitrile was added dropwise to a mixture of piperazine (23.2 g, 0.27 mol) and potassium carbonate (13.8 g, 0.10 mol) in 100 mL of acetonitrile contained in a flask equipped with a magnetic stirrer. The mixture was stirred at room temperature for 20 hrs. At this point, TLC analysis showed no evidence of reaction. After the mixture was heated at reflux for 4 hr, evidence of starting material was absent

from the TLC. The reaction mixture was filtered, and the collected solid was washed with about 200 mL of acetonitrile. The combined filtrates were reduced to about 40 mL volume at aspirator pressure and were then mixed with 40 mL of 1 M aqueous sodium hydroxide. The layers were separated, and the aqueous layer was extracted with 3 X 75 mL of dichloromethane. The combined organic layers were extracted with 3 X 75 mL of 1 M aqueous sodium hydroxide, filtered through Whatman 1PS paper, and concentrated *in vacuo*. After drying for 1 hr at high vacuum, the residue weighed 16 g. This crude product was mixed with diethyl ether, and an insoluble solid was removed by filtration.

The filtrate was concentrated, dissolved in ethyl acetate, and applied to 50 g of flash silica gel. The column was eluted with 5 X 125 mL of 0.5% methanol in ethyl acetate, 4 X 125 mL of 0.5% methanol and 1% triethylamine in ethyl acetate, and 4 X 125 mL of 0.5% methanol and 2% triethylamine in ethyl acetate. The product was eluted in the 3rd through 13th fractions. The combined fractions were concentrated to give 8.5 g of an oil. The oil was mixed with a small volume of hexanes to produce a crystalline solid. The solid was collected with the aid of more hexanes, yielding 7.0 g (52%) of product having a melting point of 75-77°.

Step 2

20 Preparation of 4-Trifluoromethylbenzyhydrol:

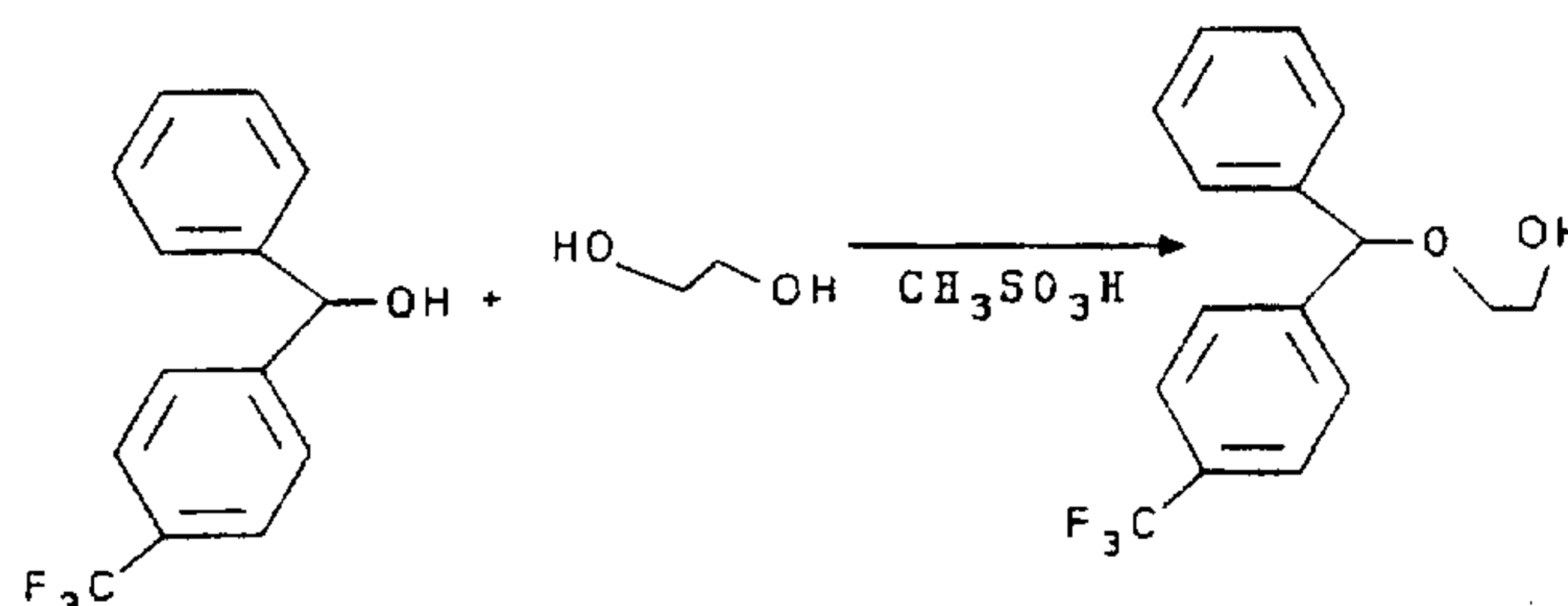


Sodium borohydride (0.42 g, 0.011 mol) was added in 2 equal portions to a stirred solution of 4-trifluoromethylbenzophenone (5 g, 0.020 mol) in 15 mL of ethanol. After 30 min, the reaction mixture was poured into water and extracted with dichloromethane. The organic phase was washed with water, filtered

through Whatman 1PS paper, and concentrated at aspirator pressure to yield 4.5 g of 4--trifluoromethylbenzohydrol. This was used without further purification.

Step 3

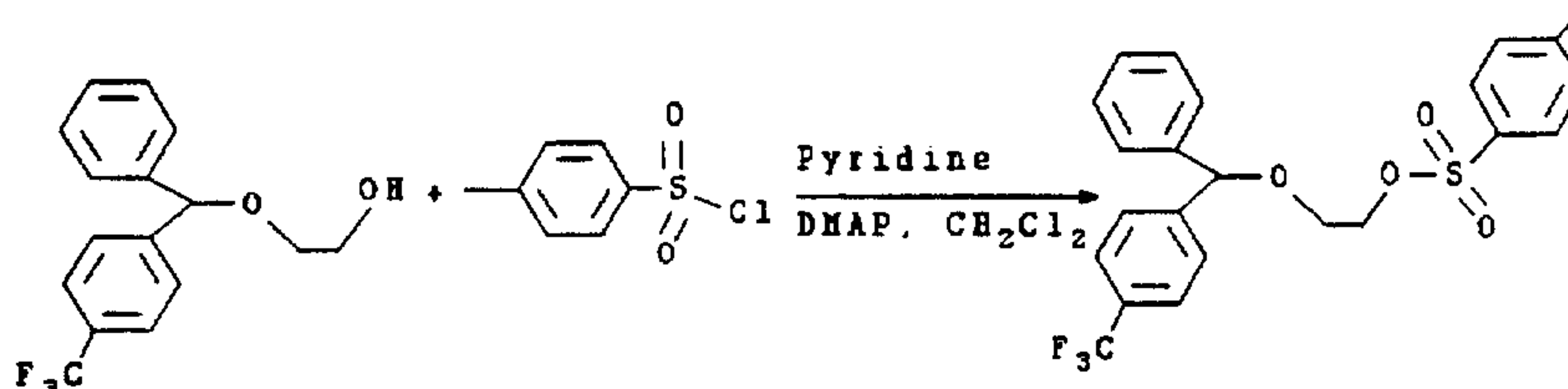
Preparation of 2-(4-Trifluoromethylphenyl(phenyl)methoxy)ethanol:



5 4-Trifluoromethylbenzohydrol (4.5 g, 0.018 mol) was dissolved in ethylene glycol (12 mL 0.21 mol) by warming on a steam bath. Methanesulfonic acid (0.80 mL, 0.012 mol) was then added in one portion. Two phases formed after the mixture had been heated for an additional 10 min. TLC analysis at this point revealed the absence of starting material. The mixture was poured into water and
10 extracted with ethyl acetate. The organic layer was filtered through Whatman 1PS paper and concentrated at reduced pressure. The residue was dissolved in hexanes, and the resulting solution was applied to 30 g of flash silica gel. The column was eluted with 2 X 50 mL of hexanes, 6 X 50 mL of 1:1 hexanes:dichloromethane, and 2 x 50 mL of dichloromethane. Pure product was
15 eluted in the 7th fraction. This was used without further purification.

Step 4

Preparation of 2-(4-Trifluoromethylphenyl(phenyl)methoxy)ethyl p-toluenesulfonate:

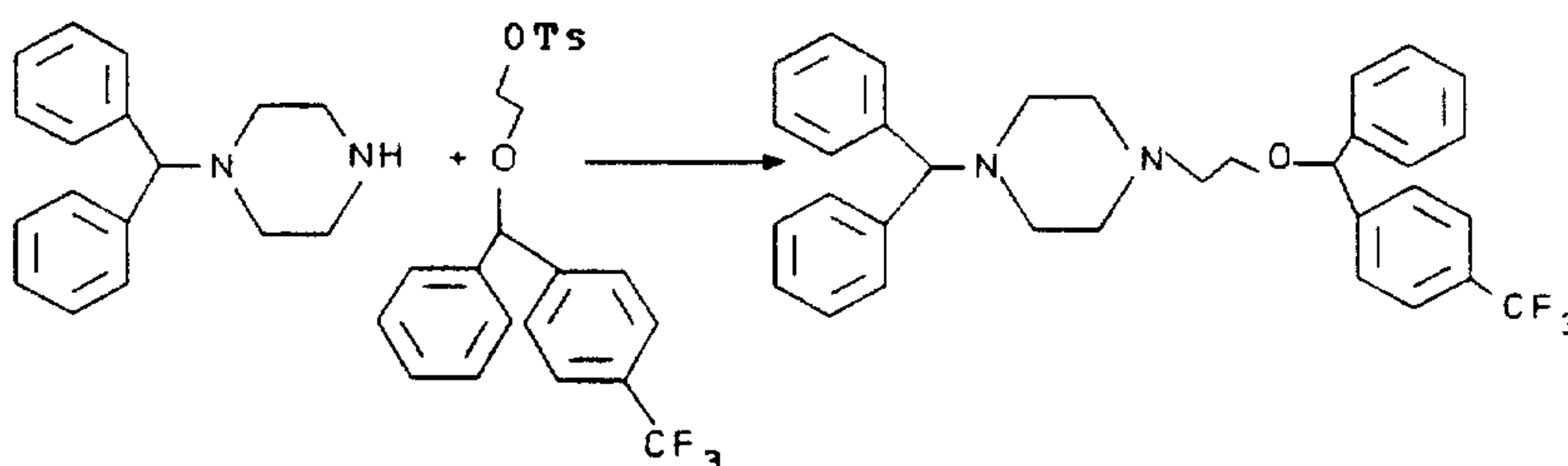


A solution of 2-(4-trifluoromethylphenyl(phenyl)methoxy)ethanol (3.4 g, 0.012 mol) dissolved in about 10 mL of dichloromethane was mixed with 5 mL
20 of pyridine, a trace amount of dimethylaminopyridine, and p-toluenesulfonyl chloride (2.2 g, 0.012 mol) in a flask equipped with a magnetic stirrer and a

drying tube. The mixture was stirred for about 18 hr, and then it was diluted with 100 mL of water and 50 mL of dichloromethane. The organic layer was washed with 100 mL of water, 50 mL of 1 M aqueous citric acid, 100 mL of water, 100 mL of saturated aqueous sodium bicarbonate, 100 mL of water, and 50 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo*. The 4.6 g of product was used without further purification.

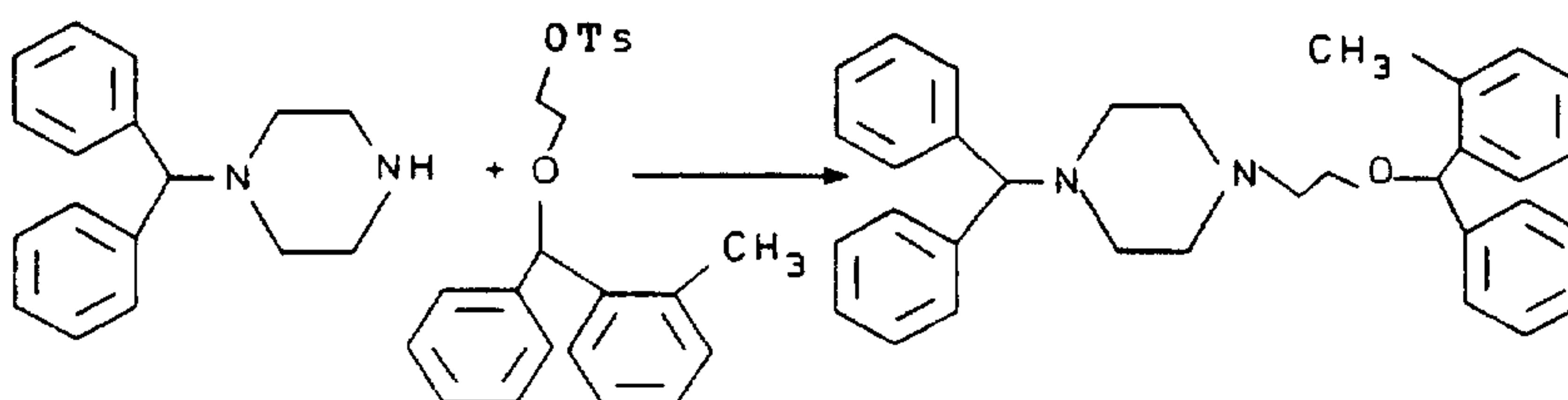
Step 5

Preparation of 1-Benzhydryl-4-[2-(4-trifluoromethylphenyl phenylmethoxy)ethyl]piperazine:



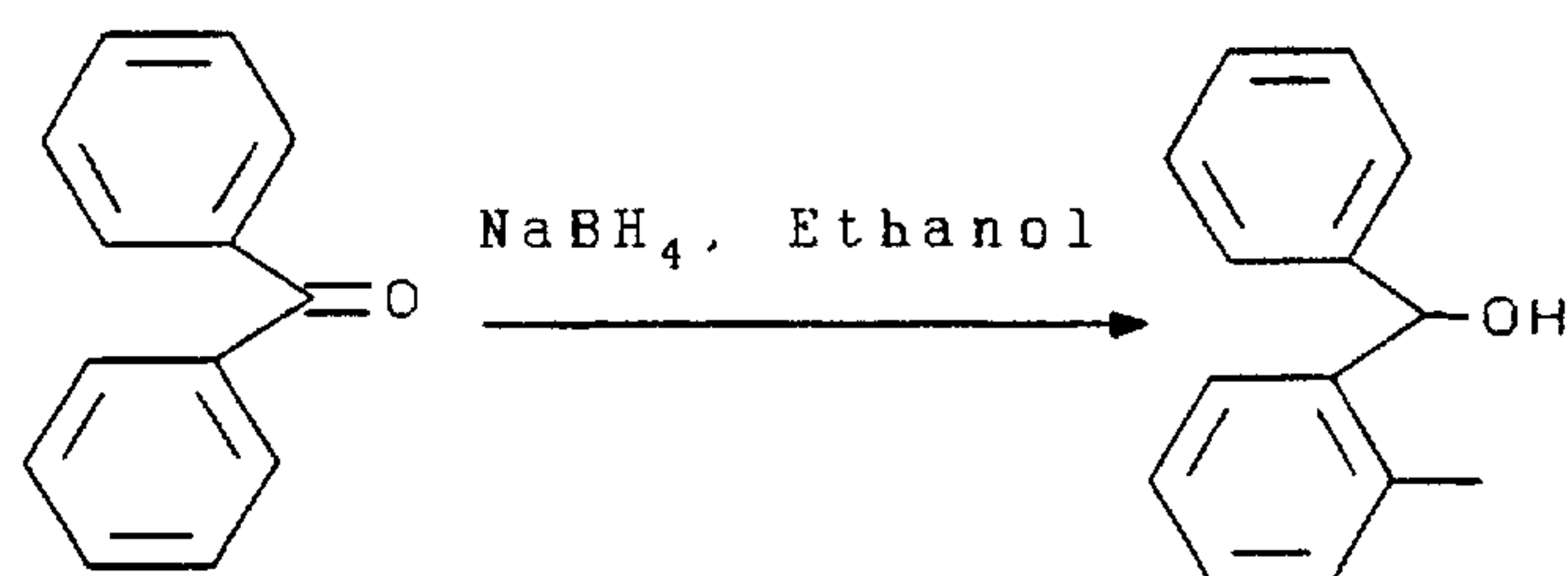
A solution of 1-diphenylmethylnpiperazine (0.5 g, 2.0 mmol) and 4-trifluoromethyldiphenylmethoxyethyl tosylate (0.97 g, 2.0 mmol) was heated under reflux with potassium carbonate (0.41 g, 3 mmol) in methyl isobutyl ketone (10 mL). The reaction was complete after 6 hrs. The reaction mixture was diluted with water and extracted with ethyl acetate. The ethyl acetate solution was evaporated, and the residue was subjected to flash chromatography over silica gel eluting with ethyl acetate/hexane mixtures. The desired 1benzhydryl-4-[2-(4-trifluoromethylphenylphenylmethoxy)ethyl]piperazine (180 mg) was obtained as an oil.

Synthesis Example 14 - 1-Benzhydryl-4-[2-(2-methylphenylphenylmethoxy)ethyl]piperazine:



Step 1

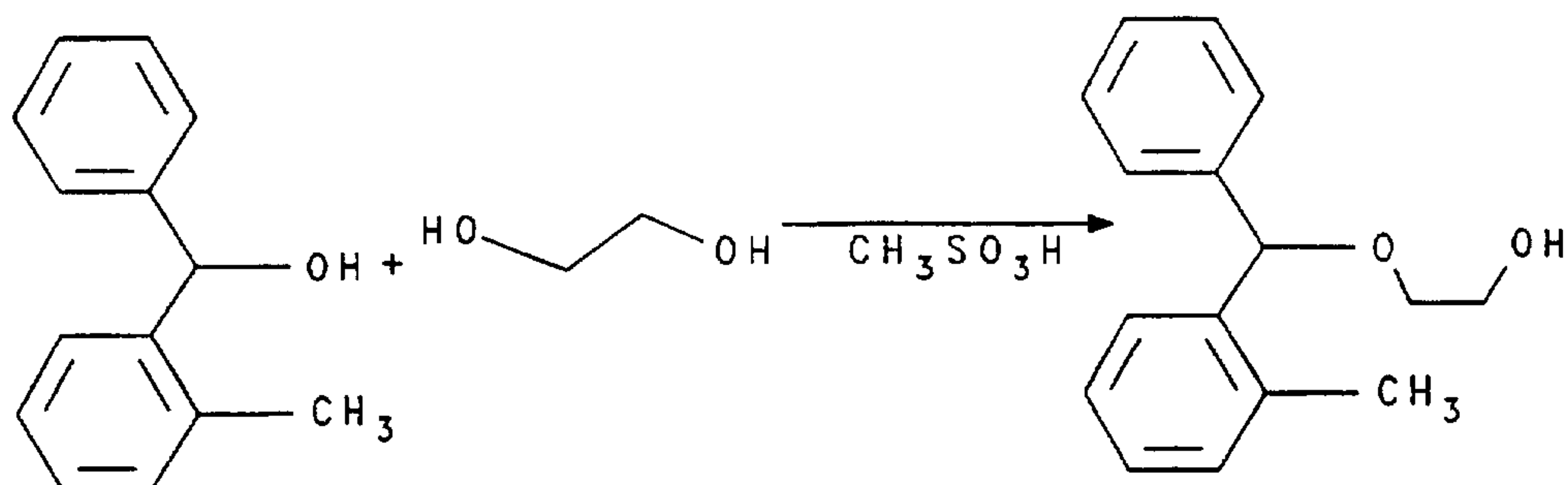
Preparation of 2-Methylbenzohydrol



Sodium borohydride (2.1 g, 0.056 mol) was added in one portion to a stirred solution of 2-methylbenzophenone (20 g, 0.10 mol) in 50 mL of ethanol. The reaction mixture became warm, and the vessel was immersed in a water bath. After 40 min, the reaction mixture was diluted with 100 mL of water and extracted with 2 X 50 mL of dichloromethane. The combined organic phases were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated at aspirator pressure to yield 19.4 g of 2-methylbenzohydrol. This was used without further purification.

Step 2

Preparation of 2- (2-Methylphenyl (phenyl) methoxy) ethanol:

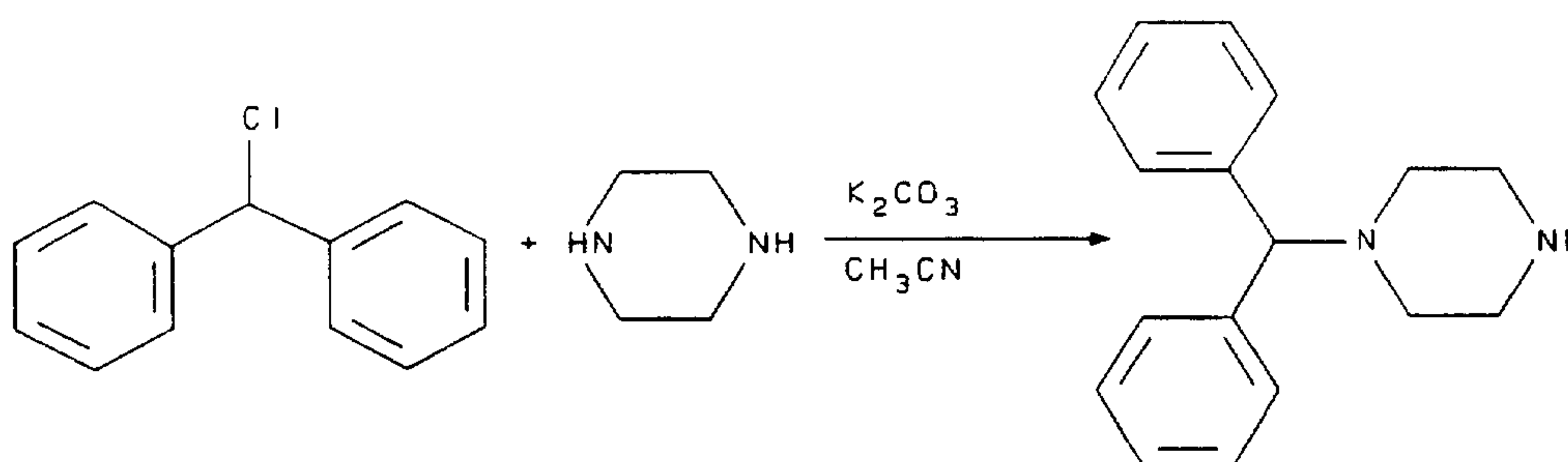


2-Methylbenzohydrol (19.4 g, 0.098 mol) and ethylene glycol (60 mL, 1.0 mol), were combined in a flask equipped with a magnetic stirrer, and the mixture was warmed on a steam bath until the starting material dissolved. Methanesulfonic acid (3.9 mL, 0.061 mol) was then added in one portion. Thin layer chromatographic analysis (TLC) indicated the absence of starting alcohol at this point. The mixture was cooled to room temperature, was diluted with 100 mL of water, and was extracted with 2 X 50 mL of dichloromethane. The combined

organic layers were washed with 3 X 100 mL of water, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The residue (9.1 g) was dissolved in hexanes, and the resulting solution was applied to 150 g of flash silica. The column was eluted with 4 X 100 mL each of 5% dichloromethane in hexanes, 10% dichloromethane in hexanes, 20% dichloromethane in hexanes, 50% dichloromethane in hexanes, and dichloromethane. The product was eluted in the 14th through 21st fractions. The combined fractions were concentrated to yield 7.7 g of material. This was used without further purification.

Step 3

Preparation of 1-Benzhydrylpiperazine:



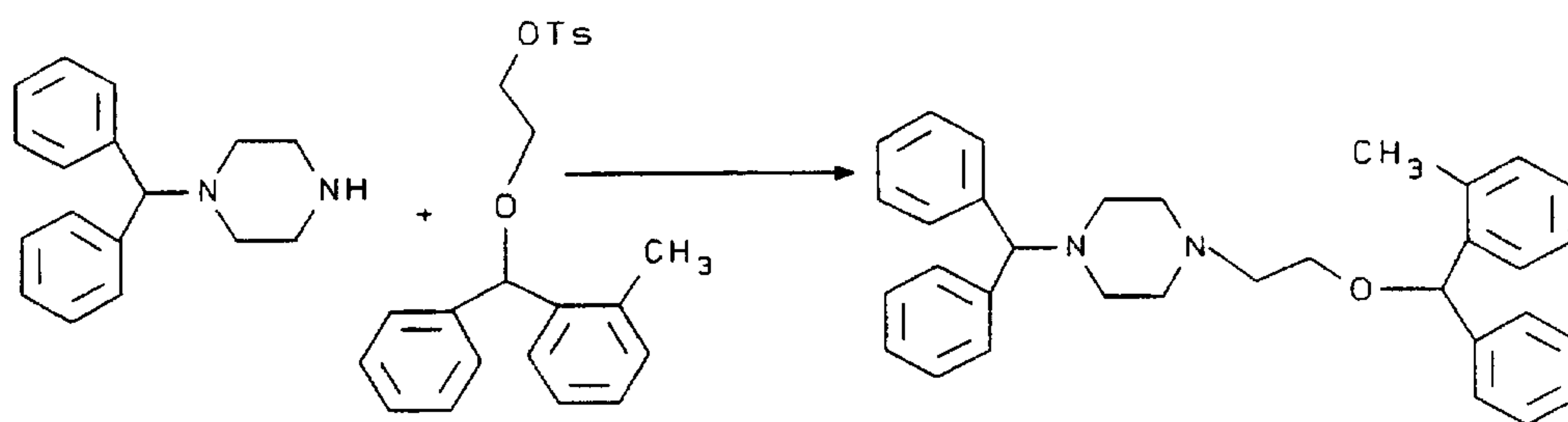
Benzhydryl chloride (10 g, 0.054 mol) in about 100 mL of acetonitrile was added dropwise to a mixture of piperazine (23.2 g, 0.27 mol) and potassium carbonate (13.8 g, 0.10 mol) in 100 mL of acetonitrile contained in a flask equipped with a magnetic stirrer. The mixture was stirred at room temperature for 20 hrs. At this point, TLC analysis showed no evidence of reaction. After the mixture was heated at reflux for 4 hr, evidence of starting material was absent from the TLC. The reaction mixture was filtered, and the collected solid was washed with about 200 mL of acetonitrile. The combined filtrates were reduced to about 40 mL volume at aspirator pressure and were then mixed with 40 mL of 1 M aqueous sodium hydroxide. The layers were separated, and the aqueous layer was extracted with 3 X 75 mL of dichloromethane. The combined organic layers were extracted with 3 X 75 mL of 1 M aqueous sodium hydroxide, filtered through Whatman 1PS paper, and concentrated *in vacuo*. After drying for 1 hr at high vacuum, the residue weighed 16 g. This crude product was mixed with diethyl ether, and an insoluble solid was removed by filtration.

The filtrate was concentrated, dissolved in ethyl acetate, and applied to 50 g of flash silica gel. The column was eluted with 5 x 125 mL of 0.5% methanol in ethyl acetate, 4 X 125 mL of 0.5% methanol and 1% triethylamine in ethyl acetate, and 4 X 125 mL of 0.5% methanol and 2% triethylamine in ethyl acetate.

5 The product was eluted in the 3rd through 13th fractions. The combined fractions were concentrated to give 8.5 g of an oil. The oil was mixed with a small volume of hexanes to produce a crystalline solid. The solid was collected with the aid of more hexanes, yielding 7.0 g (52%) of product, having a melting point of 75-77°.

Step 4

10 Preparation of 1-Benzhydryl-4-[2-(2-methylphenylphenylmethoxy) ethyl-piperazine:

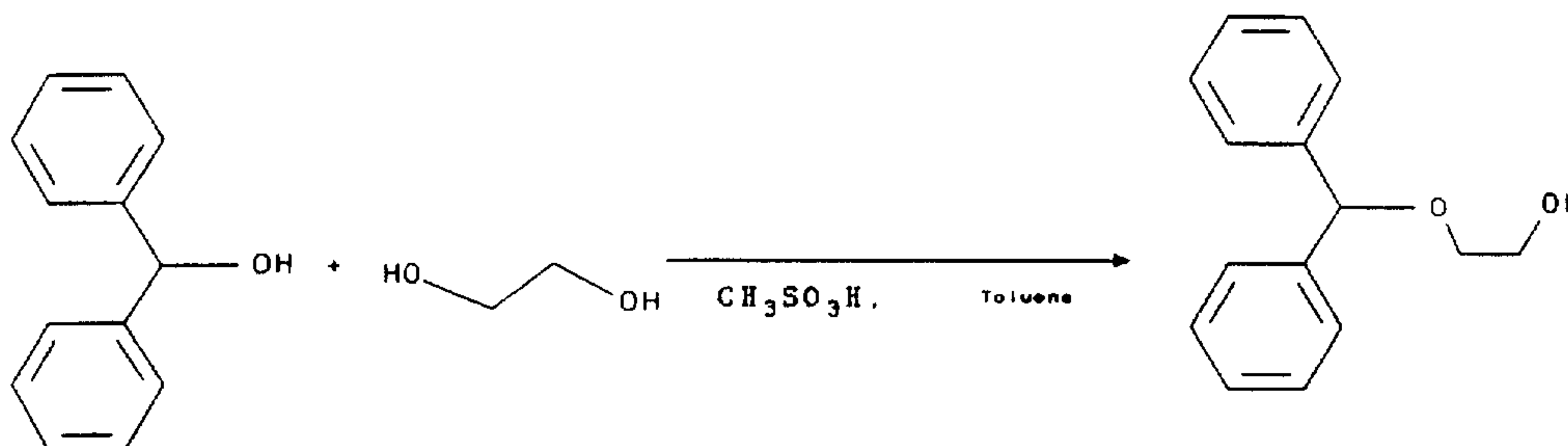


A solution of 1-benzhydrylpiperazine (0.5 g, 2 mmol), 2—(2-methylbenzhydryloxy)ethyl tosylate (0.86 g, 2 mmol) and potassium carbonate (0.41 g, 3 mmol) and 10 mL of methyl isobutyl ketone was heated under reflux
 15 with stirring for 6 hr when the reaction appeared to be complete. The reaction was diluted with water (25 mL) and extracted with ethyl acetate (3 x 25 mL). The ethyl acetate solution was washed with saturated brine (25 mL) and filtered through 1PS filter paper. Evaporation of the ethyl acetate afforded the crude product which was flash chromatographed over 25 g of flash chromatography
 20 silica gel. The 1-benzhydryl—4-[2—(2- methylphenylphenylmethoxy)ethyl-piperazine (250 mg, 25%) was obtained as an oil which showed $R_f=0.6$ on TLC (dichloromethane).

Synthesis Example 15 - 1-Benzhydryl-4-[2-(diphenylmethoxy) ethyl] piperazine:

Step 1

Preparation of 2-(Diphenylmethoxy) ethanol:

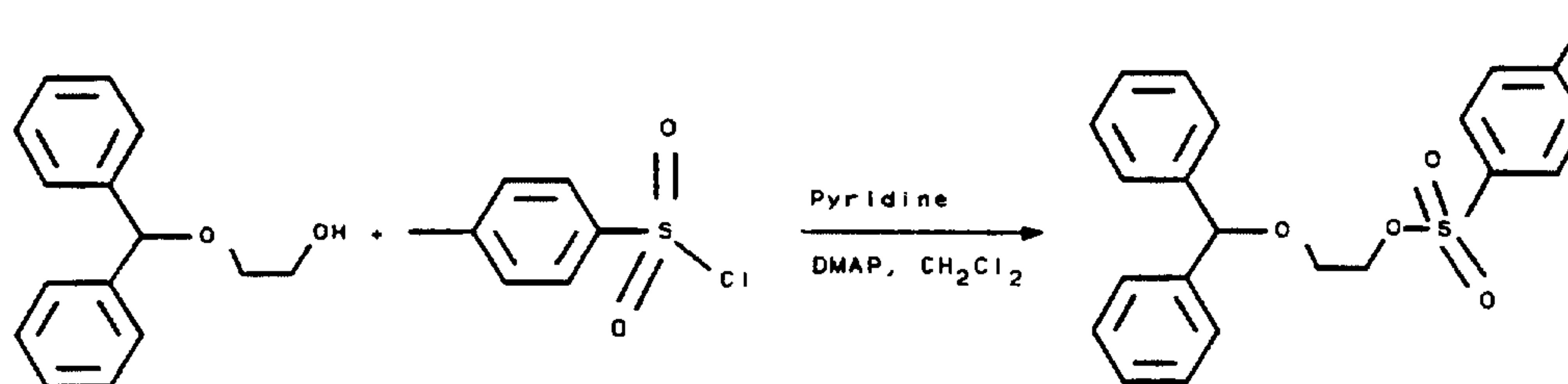


- 5 A 250 mL round bottomed flask was charged with ethylene glycol (60 ml, 1.08 mol), toluene (30 mL) and methanesulfonic acid (5.18 g, 54 mmol). The mixture was heated on the steam bath and a solution of benzhydrol (10 g, 54 mmol) in toluene (70 mL) was added over 1 hr. The reaction was complete at the end of the addition as indicated by TLC (dichloromethane). The cooled
- 10 reaction mixture was treated with saturated sodium bicarbonate solutions, and the layers were separated. The aqueous layer was extracted with dichloromethane (2 x 50 mL). The combined organic layers were washed with water (3 x 50 mL) and the solvent was evaporated. The crude product was flash chromatographed to give 7.0g (57%) of 2-diphenylmethoxyethanol.

15

Step 2

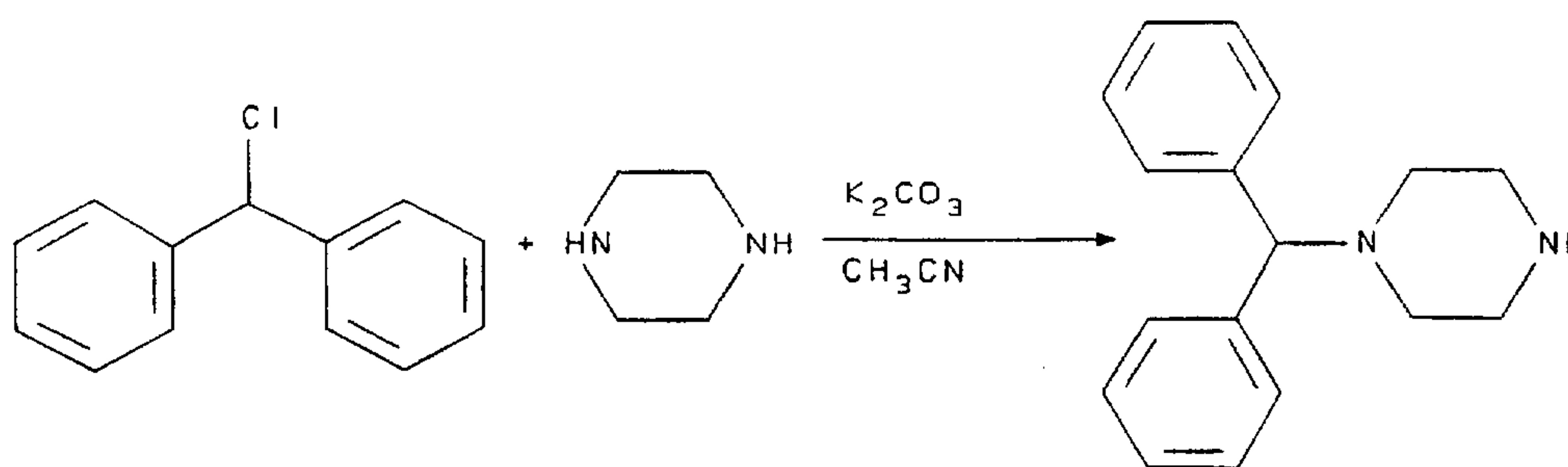
Preparation of 2-Diphenylmethoxyethyl p-toluenesulfonate:



A sample of 2—diphenylmethoxyethanol (5.7 g, 0.025 mol) was dissolved in about 25 mL of dichloromethane. This solution was placed over 3A molecular sieves to dry. It was then decanted away from the sieves and mixed with p-toluenesulfonyl chloride (4.76 g, 0.025 mol), pyridine (8.2 mL), and a minute amount of dimethylaminopyridine. The mixture was stirred for 64 hr, and then it was washed, in order, with water, 1 M hydrochloric acid, water, saturated aqueous sodium bicarbonate, and water. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 6.1 g of product.

Step 3

10 Preparation of 1-Benzhydrylpiperazine:



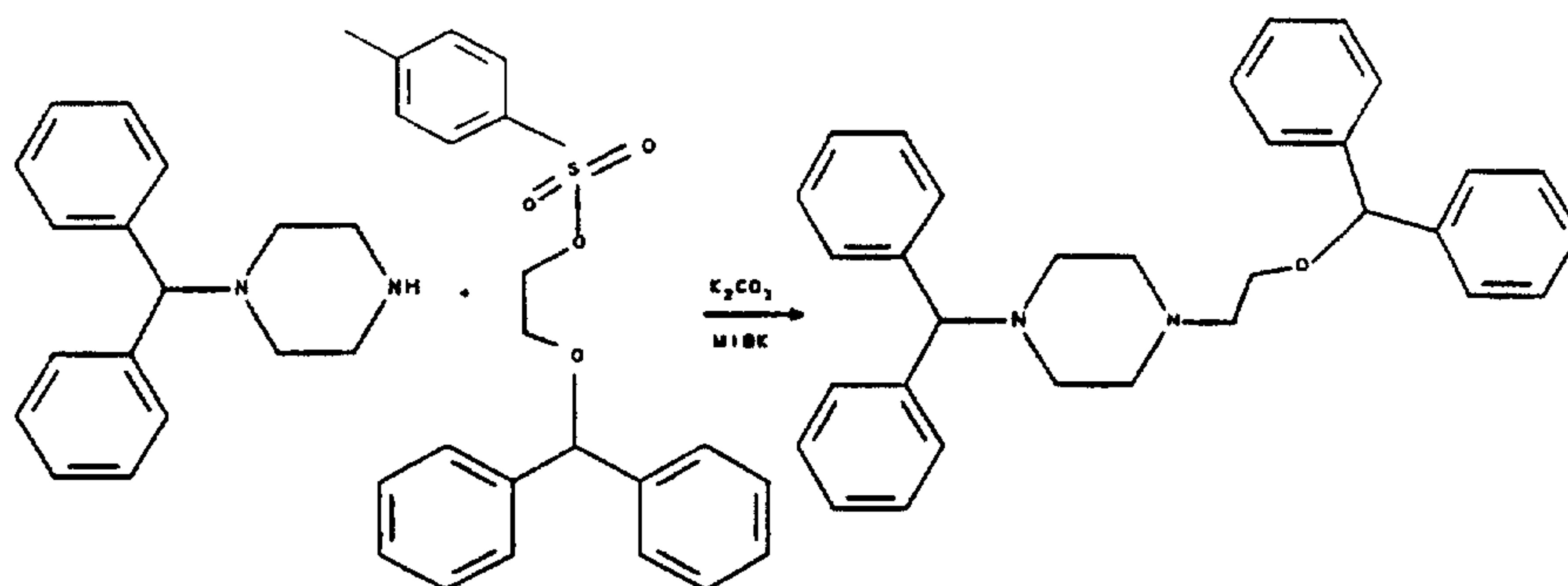
Benzhydryl chloride (10 g, 0.054 mol) in about 100 mL of acetonitrile was added dropwise to a mixture of piperazine (23.2 g, 0.27 mol) and potassium carbonate (13.8 g, 0.10 mol) in 100 mL of acetonitrile contained in a flask equipped with a magnetic stirrer. The mixture was stirred at room temperature for 20 hrs. At this point, TLC analysis showed no evidence of reaction. After the mixture was heated at reflux for 4 hr, evidence of starting material was absent from the TLC. The reaction mixture was filtered, and the collected solid was washed with about 200 mL of acetonitrile. The combined filtrates were reduced to about 40 mL volume at aspirator pressure and were then mixed with 40 mL of 1 M aqueous sodium hydroxide. The layers were separated, and the aqueous layer was extracted with 3 X 75 mL of dichloromethane. The combined organic layers were extracted with 3 X 75 mL of 1 M aqueous sodium hydroxide, filtered through Whatman 1PS paper, and concentrated *in vacuo*. After drying for 1 hr at high vacuum, the residue weighed 16 g. This crude product was mixed with diethyl ether, and an insoluble solid was removed by filtration.

The filtrate was concentrated, dissolved in ethyl acetate, and applied to 50 g of flash silica gel. The column was eluted with 5 x 125 mL of 0.5% methanol in ethyl acetate, 4 X 125 mL of 0.5% methanol and 1% triethylamine in ethyl acetate, and 4 X 125 mL of 0.5% methanol and 2% triethylamine in ethyl acetate.

5 The product was eluted in the 3rd through 13th fractions. The combined fractions were concentrated to give 8.5 g of an oil. The oil was mixed with a small volume of hexanes to produce a crystalline solid. The solid was collected with the aid of more hexanes, yielding 7.0 g (52%) of product, having a melting point of 75-77°.

10 Step 4

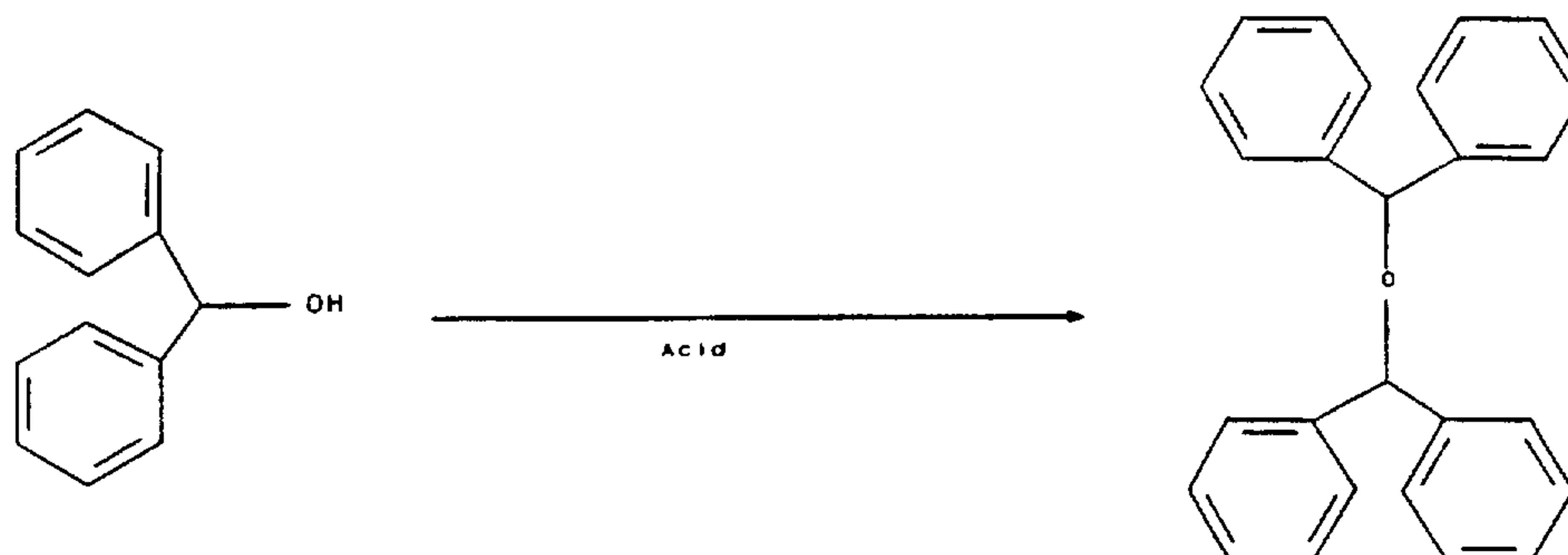
Preparation of 1—Benzhydryl-4- [2-(diphenylmethoxy) ethyl]piperazine:



A mixture of 2-diphenylmethoxyethyl p-toluenesulfonate (0.83 g, 0.0020 mol), 1—benzhydrylpiperazine (0.50 g, 0.0020 mol), and potassium carbonate (0.41 g, 0.0030 mol) in 10 mL of methyl isobutyl ketone was heated at reflux for 18 hrs. At this point, TLC analysis indicated complete reaction. The mixture was diluted with 25 ml of water and extracted with 2 X 25 mL of ethyl acetate. The combined organic layers were washed with 25 mL of saturated aqueous sodium chloride, filtered through Whatman 1PS paper, and concentrated *in vacuo*. The 1.2 g of residue was applied to 20 g of flash silica gel and eluted with 5 X 50 mL of hexanes, 7 X 100 mL of dichloromethane, and 3 X 50 mL of ethyl acetate. The product appeared in the 13th and 14th fractions. These were combined and concentrated, and the oily residue was triturated with 25 mL of hexanes. The supernatant was separated from a gummy residue and concentrated to give 0.75 g of an oil. This crystallized from about 3 mL of pentane. After the

crystals were collected and dried under high vacuum over refluxing acetone, they weighed 0.45 g. (49% of theoretical), having a melting point of 77-79°.

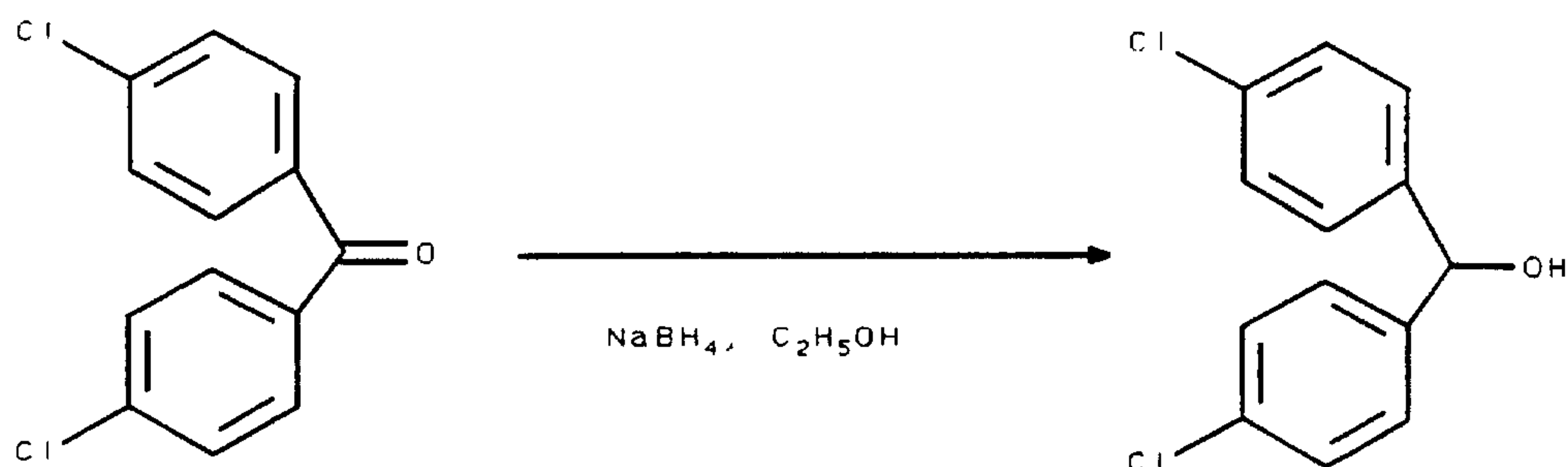
Synthesis Example 16 - Dibenzhydryl ether:



To a stirred solution of benzhydrol (500 g, 2.7 mol) in ethylene glycol
5 (101.06 g, 1.63 mol) and 1,2-dimethoxy-ethane (1.5 L) was added
methanesulfonic acid (100 mL) over 10 min. The reaction mixture was heated
on a steam bath for 30 min., when TLC (dichloromethane) indicated that no
starting material remained. The mixture was cooled and poured into ice water (6
L) containing sodium hydroxide (60 g, 1.5 mol). The mixture was extracted with
10 ethyl acetate (2 x 1L), and the ethyl acetate was filtered to give 100 g of solid.
The ethyl acetate solution was evaporated under reduced pressure. The solid was
recrystallized from ethanol to give 50 g of impure 1,2-bis(diphenyl-
methoxy)ethane, sample 1. The residue from the ethyl acetate solution was
successively recrystallized from ethanol and then ethyl acetate to give 150 g of
15 impure 1,2-bis(diphenyl-methoxy)ethane. This material was recrystallized from
100 mL of toluene to give 100 g of impure 1,2-bis(diphenylmethoxy)ethane. The
mother liquors from the recrystallization from toluene were evaporated under
reduced pressure. The residue was crystallized successively from ethanol and
then toluene. The toluene mother liquors were evaporated under reduced pressure
20 and the residue was crystallized successively from ethanol and twice from toluene
to give pure bis(benzhydryl)ether (13 g, 2%).

Synthesis Example 17 - Di (4,4-dichlorobenzhydryl) ether:

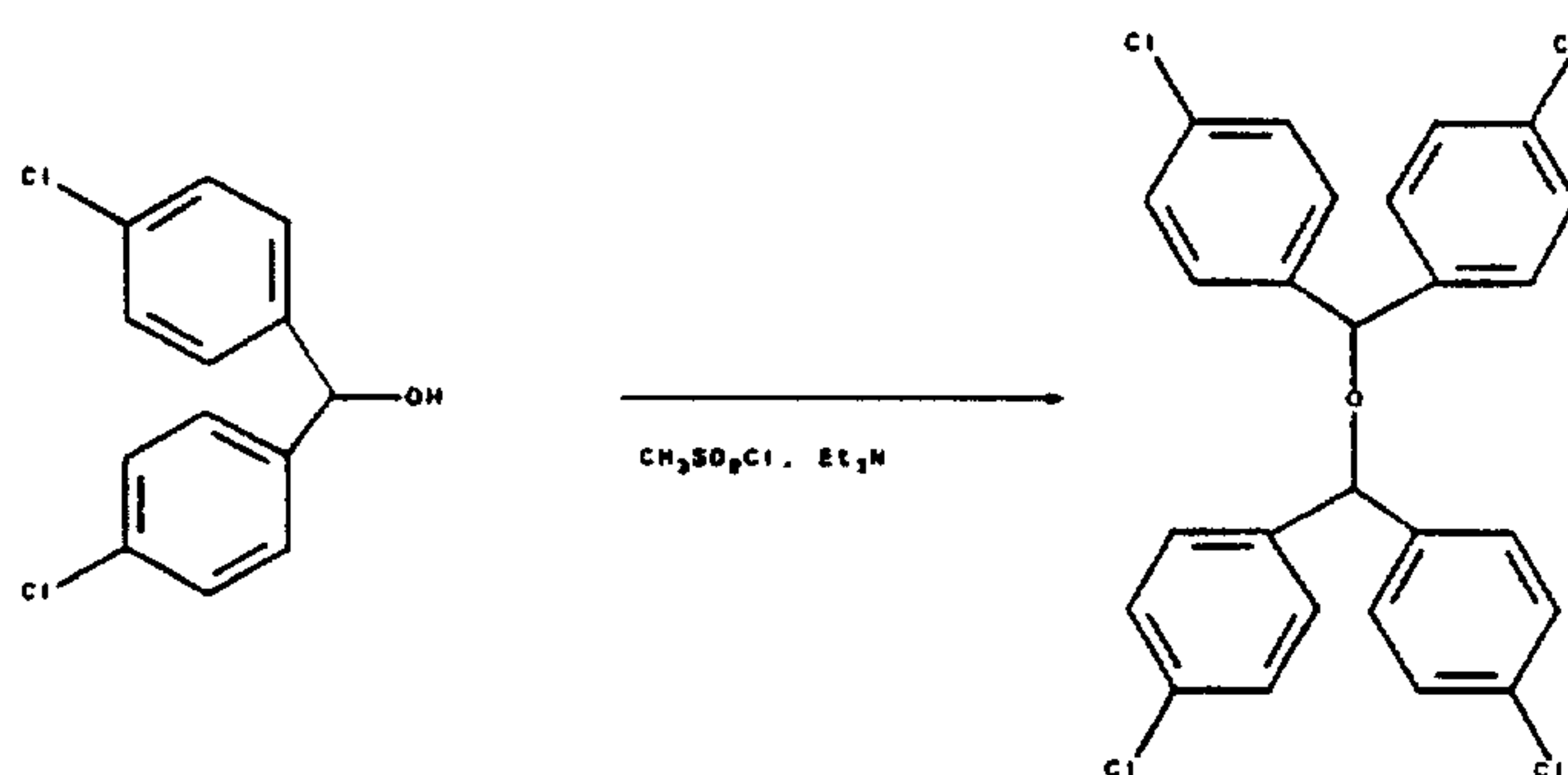
Step 1



A suspension of 4,4'-dichlorobenzophenone (5.02 g, 20 mmol) in ethanol (15 mL) was treated with sodium borohydride (0.42 g, 11 mmol) added in one portion. The material went into solution and then a precipitate formed. After 30 min., the reaction was complete by TLC (dichloromethane). The reaction mixture was diluted with 25 mL of water and extracted with dichloromethane (2 x 50 mL). The dichloromethane solution was washed with water (2 x 25 mL) and brine (2 x 25 mL) and then evaporated under reduced pressure to give 5.1 g (100%) of pure 4,4'-dichlorobenzohydrol.

Step 2

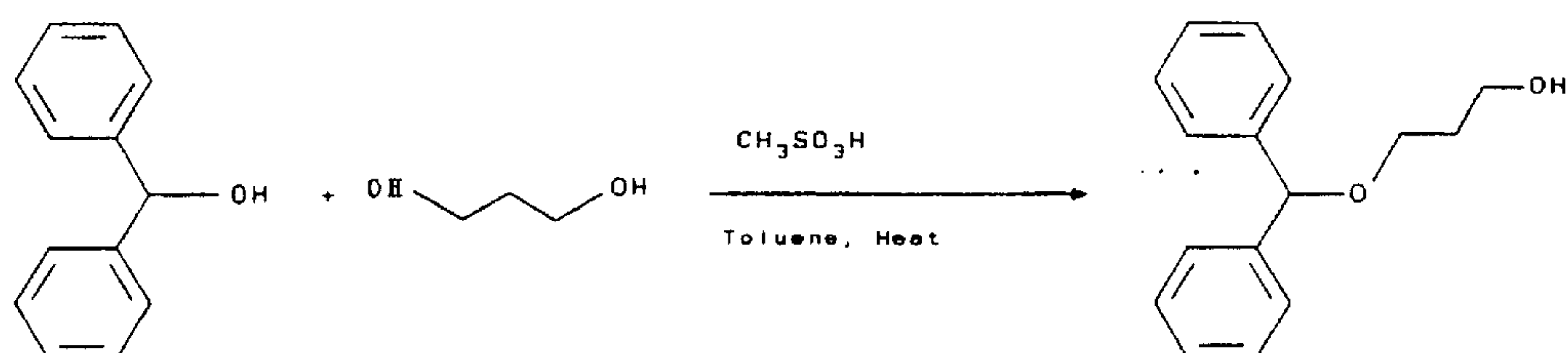
10 Preparation of Di (4, 4'-dichlorobenzhydryl) ether:



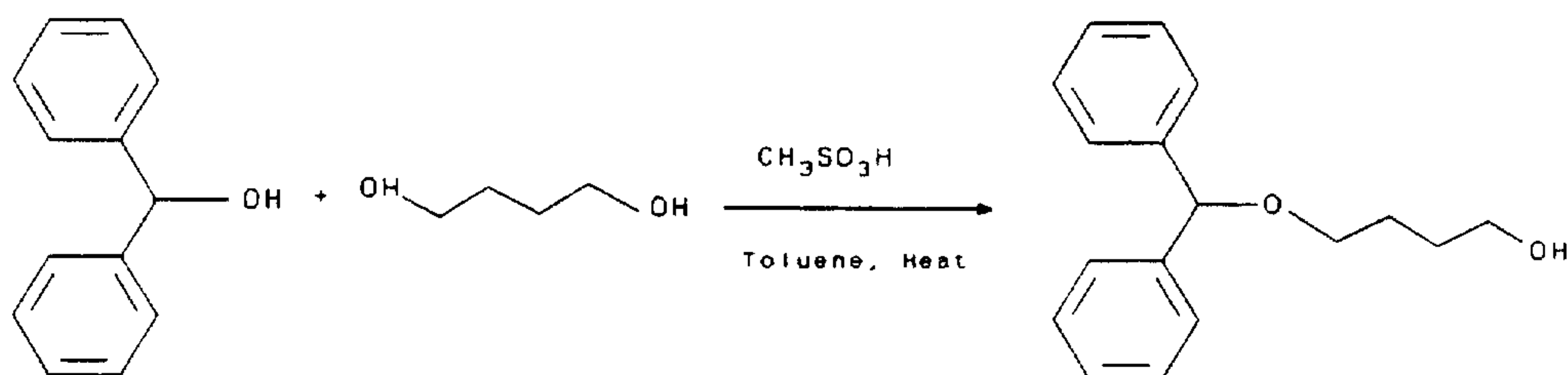
A stirred solution of 4,4'-dichlorobenzohydrol (5.0 g, 19.8 mmol) in ether (50 mL) was cooled in an ice bath and treated with methanesulfonyl chloride (2.48 g, 21.7 mmol) added in one portion. A solution of triethylamine (2.21 g, 21.7 mmol) in ether (10 mL) was added dropwise over 10 min. Stirring and cooling was continued for 30 min. and the reaction mixture was filtered. The crude product in ether solution was washed successively with sodium bicarbonate solution, 1 N citric acid, water and brine. The ether was evaporated under

reduced pressure, and the residue was crystallized from ethanol to give pure di(4,4'- dichlorobenzhydryl)ether, having a melting point of 126-129°.

Synthesis Example 18 - 3-Diphenylmethoxypropanol:



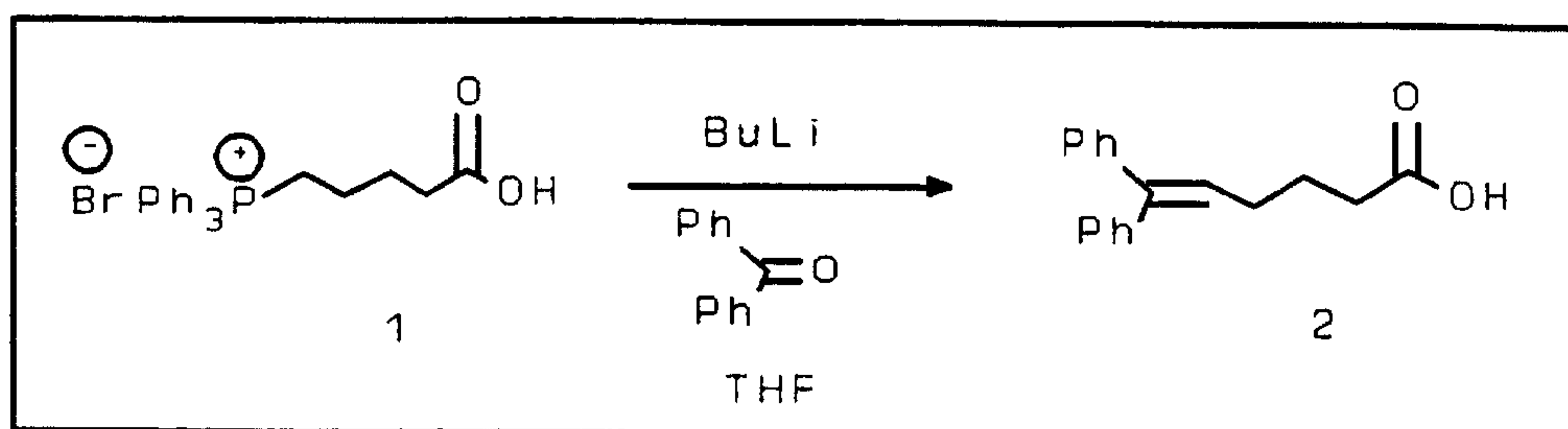
A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,3-propanediol (100 g, 1.31 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene (150 mL). The mixture was immersed in a 130° bath, and benzohydrol (27.6 g, 0.15 mol) dissolved in 250 mL of toluene was added dropwise over 90 min. After an additional 30 min., TLC analysis indicated complete reaction, and the reaction mixture was cooled and diluted with 200 mL of ethyl acetate. The layers were separated, and the diol layer was extracted with an additional 100 mL of ethyl acetate. The combined ethyl acetate phases were washed with 100 mL of water, 100 mL of saturated aqueous sodium bicarbonate, and 100 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 40 g of an oil. The oil was applied to 150 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 3 X 250 mL of 20% ethyl acetate in hexanes, and 5 X 250 mL of 40% ethyl acetate in hexanes. The product was eluted in the 3rd through 8th fractions. The fractions were combined and concentrated to give 30 g of an oil that crystallized upon standing overnight in a refrigerator. The solid was triturated with 100 mL of cold hexanes, collected, washed with 25 mL of cold hexanes, and air dried. The collected product, which had a melting point of 51–53°, weighed 25.5 g (70%).

Synthesis Example 19 - 4 -Diphenylmethoxybutanol

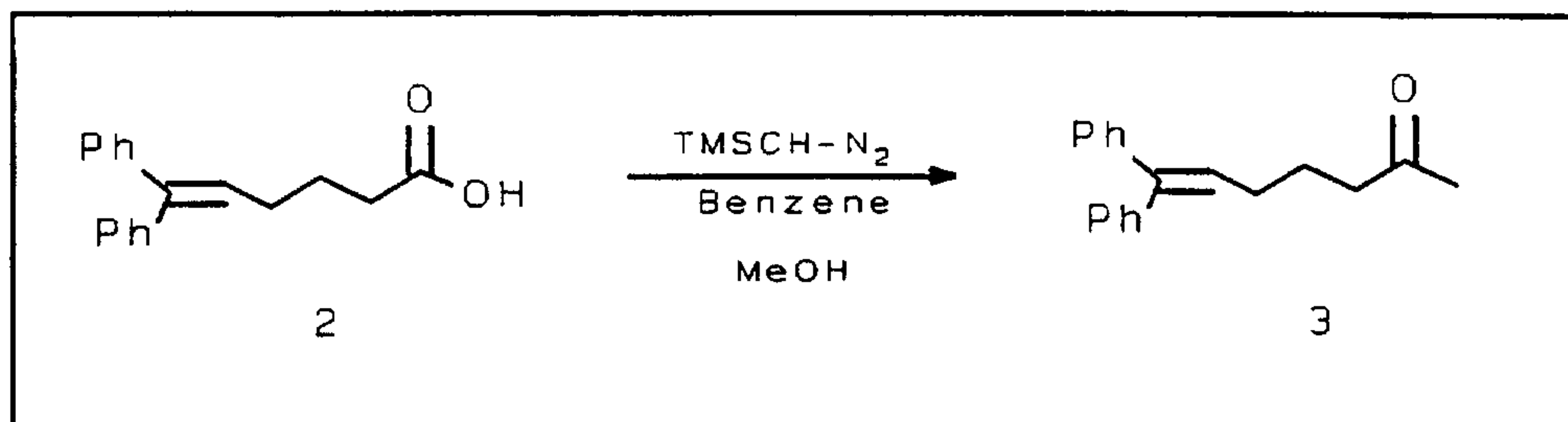
A flask equipped with a condenser, a dropping funnel, and a magnetic stirrer was charged with 1,4-butanediol (135 g, 1.5 mol), methanesulfonic acid (7.2 g, 0.075 mol), and toluene 250 mL). The mixture was immersed in a 120° bath, and benzohydrol (27.6 g, 0.15 mol) dissolved in 250 ml of toluene was added dropwise over 75 min. After an additional 1 hr, TLC analysis indicated complete reaction, and the reaction mixture was cooled to room temperature. The layers were separated, and the diol layer was extracted with 100 mL of ethyl acetate. The combined upperphases were washed with 200 mL of water, 200 mL of saturated aqueous sodium bicarbonate, and 200 mL of saturated aqueous sodium chloride. The solution was filtered through Whatman 1PS paper and concentrated *in vacuo* to yield 45g of an oil. The oil was applied to 300 g of flash silica gel. The column was eluted with 2 X 250 mL of 10% ethyl acetate in hexanes, 5 X 250 mL of 20% ethyl acetate in hexanes, and 8 X 250 mL of 40% ethyl acetate in hexanes. The product was eluted in the 8th through 13th fractions. The fractions were combined and concentrated to give 40 g of an oil.

Synthesis Example 20

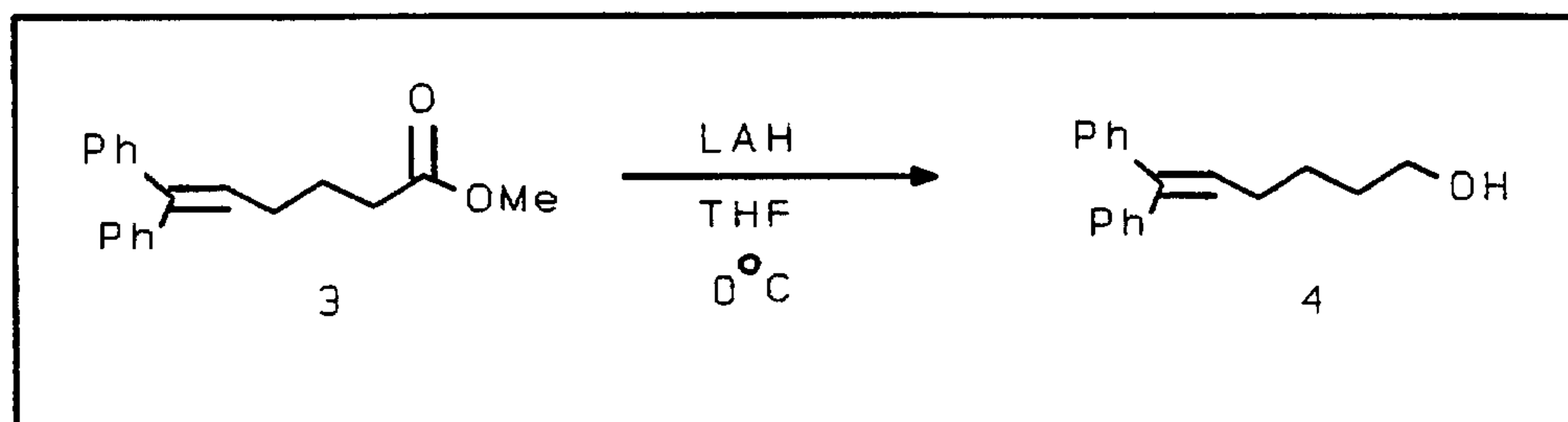
This synthesis produced products K12-6 and K12-17:



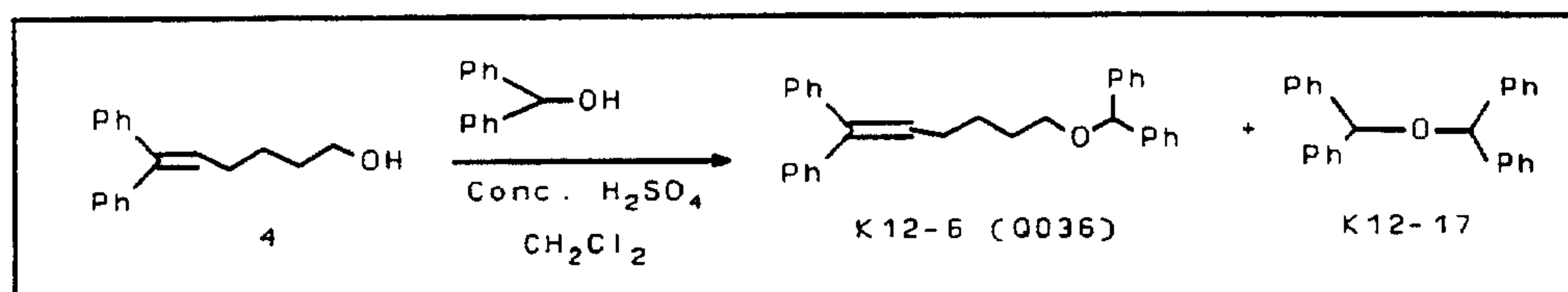
The step above was conducted as follows. The starting phosphonium salt 1 (3 g, 6.76 mmole) in THF (50 ml) was stirred to make a suspension. The mixture was cooled to 0°C, 1.6 M BuLi in hexane (8.45 ml, 13.5 mmole) was added dropwise and stirred for 1 hr. To this mixture, benzophenone (6.7 mmole) was added and stirred for 2 hrs. One more equivalent of benzophenone was added and stirred for 1 hr. To this mixture, water (100 ml) and ether (100 ml) were added and layers were separated. The aqueous layer was acidified by 1N HCl (100 ml), extracted with EtOAc (2x100 ml). The combined EtOAc layer was washed with brine (100 ml), dried over Na₂SO₄, and concentrated. The crude was purified with silica gel column using MeOH/ (0-5%)/CH₂Cl₂ to afford 570 mg of the pure product 2.



The step above was conducted as follows. To a solution of the acid 2 (520 mg, 1.95 mmole) in benzene (20 ml) at RT under Ar, was added 2M TMSCH-N₂ solution in hexane (1 ml, 2 mmole). After 1 hr the reaction was not complete, and therefore TMSCH-N₂ (600 μl) and 3 drops of MeOH were added. The mixture was stirred for 4 hrs. Glacial acetic acid (10 drops) was added and stirred for 25 min. The solvent was evaporated to give 0.66 g of the crude ester 3.



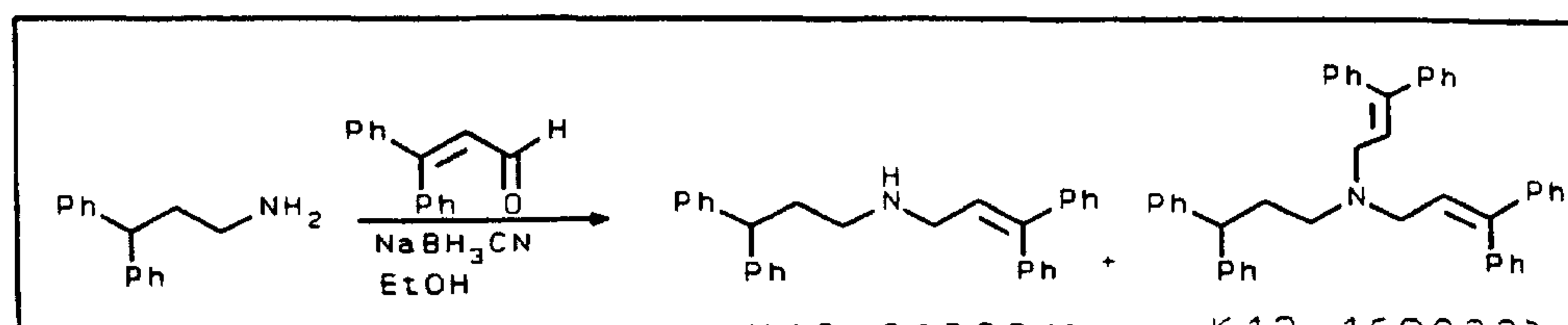
The step above was conducted as follows. To the crude solution of the ester **3** (600 mg, 1.70 mmole) in THF (7 ml) at 0°C was added 1M LAH in THF (1.7 ml, 1.7 mmole) dropwise and stirred at 0°C for 1 hr., when the TLC analysis showed the end of the reaction. To this mixture was added EtOAc (500 μ l) and stirred for 10 min. Water (20 ml) and EtOAc (20 ml) were added slowly. To this mixture was added 1N HCl (2 ml), shaken well and layers were separated. The water layer was extracted with EtOAc (2x20 ml), and then the organic layer was washed with brine (20 ml), dried over Na₂SO₄ and concentrated to give 0.48 g of the compound represented on TLC by one spot with R_f=0.2 in EtOAc/Hexane (1/4).



The step above was conducted as follows. To a solution of alcohol **4** (300 mg, 1.2 mmole) in CH₂Cl₂ (10 ml), benzhydrol (328 mg, 1.8 mmole) and concentrated H₂SO₄ (50 μ l) were added at 0°C. A red orange color appeared then disappeared with stirring after 1 hr. More benzhydrol (150 mg) and also concentrated H₂SO₄ (50 μ l) were added and stirred for 15 min. A saturated solution of NaHCO₃ (20 ml) was added and extracted with CH₂Cl₂ (2x20 ml). The organic layer was dried over Na₂SO₄ and concentrated to give 0.93 g of crude product. Column chromatography on silica gel using EtOAc/Hexane (0-3%) gave 0.81 g of product which is not pure. A second purification on silica gel using CH₂Cl₂/Hexane (0 to 20%) as eluant gave K12-7(303 mg) and K12-6(381 mg).

Synthesis Example 21

This synthesis produced products K12-1 and K12-2:



To The starting amine (500 mg, 2.36 mmole) dissolved in EtOH (7 mL) cooled to 0°C, NaBH₃CN (226 mg, 3.6 mmole) was added under Ar atmosphere. Aldehyde (250 mg, 1.2 mmole) in 1 mL EtOH was added by syringe. EtOH was evaporated and the crude was left under vacuum for 30 min.

5 Water (5 mL) and EtOAc (15 mL) were added and separated. The water layer was extracted with EtOAc again and the combined organic layer was washed with brine and dried over Na₂SO₄. The crude was filtered and concentrated. Silica gel chromatography was performed by EtOAc (10% to 70%)/Hexane to yield K12-1 (90 mg, 13%) and K12-2 (140 mg, 29%).

10 K12-2 from this synthesis was converted to salt quantitatively with 1N HCl and extracted with CH₂Cl₂. The organic layer was dried over Na₂SO₄ and concentrated.

The present invention has been described by reference to the particularly preferred embodiments and examples above. Those skilled in the art will appreciate that various modifications can be made to the invention without

15 departing significantly from the spirit and scope thereof.

WHAT IS CLAIMED IS:

1. A composition for treating cerebral amyloidosis, comprising a pharmaceutically effective amount of:

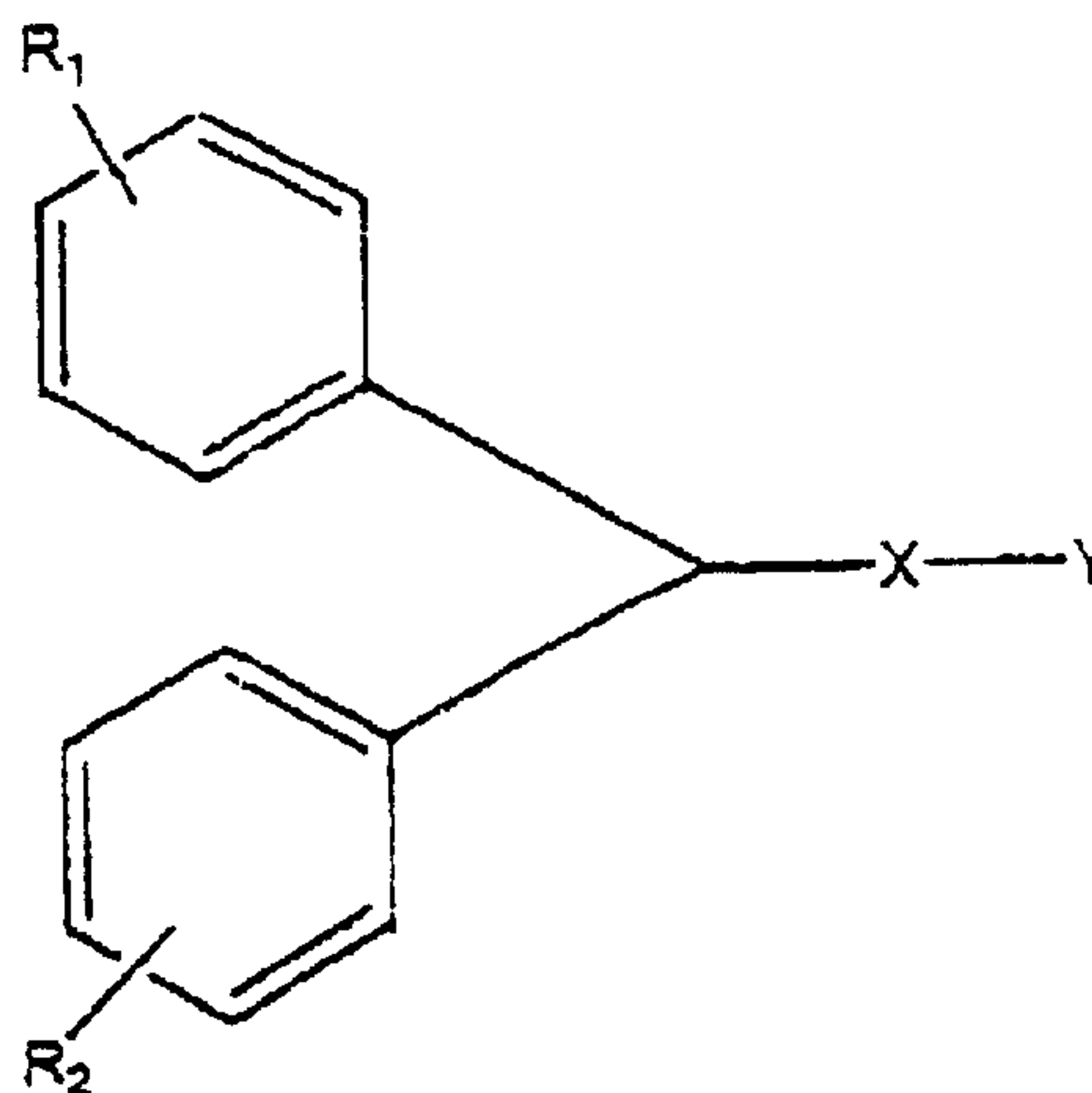
(i) a compound that impedes disruption of intact dense microspheres (DMS) by acting on DMS either to (a) prevent disruption, or (b) if disrupted, act on pre-disrupted DMS in such a way that, when compared to controls, they:

- (1) reduce the mean volume of tissue occupied by disrupted DMS;
- (2) reduce the ratio of the number of inflammatory cells per DMS; or
- (3) increase the ratio of the number of macrophages containing disrupted DMS per DMS;

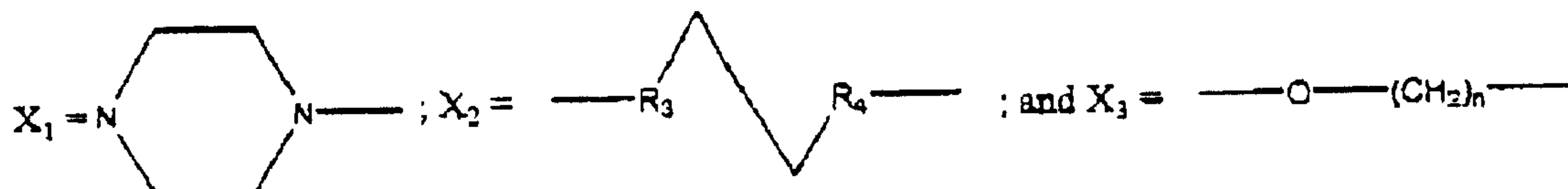
when the compound is administered to a test animal that has received an injection of DMS, and

(ii) a pharmaceutically acceptable vehicle,

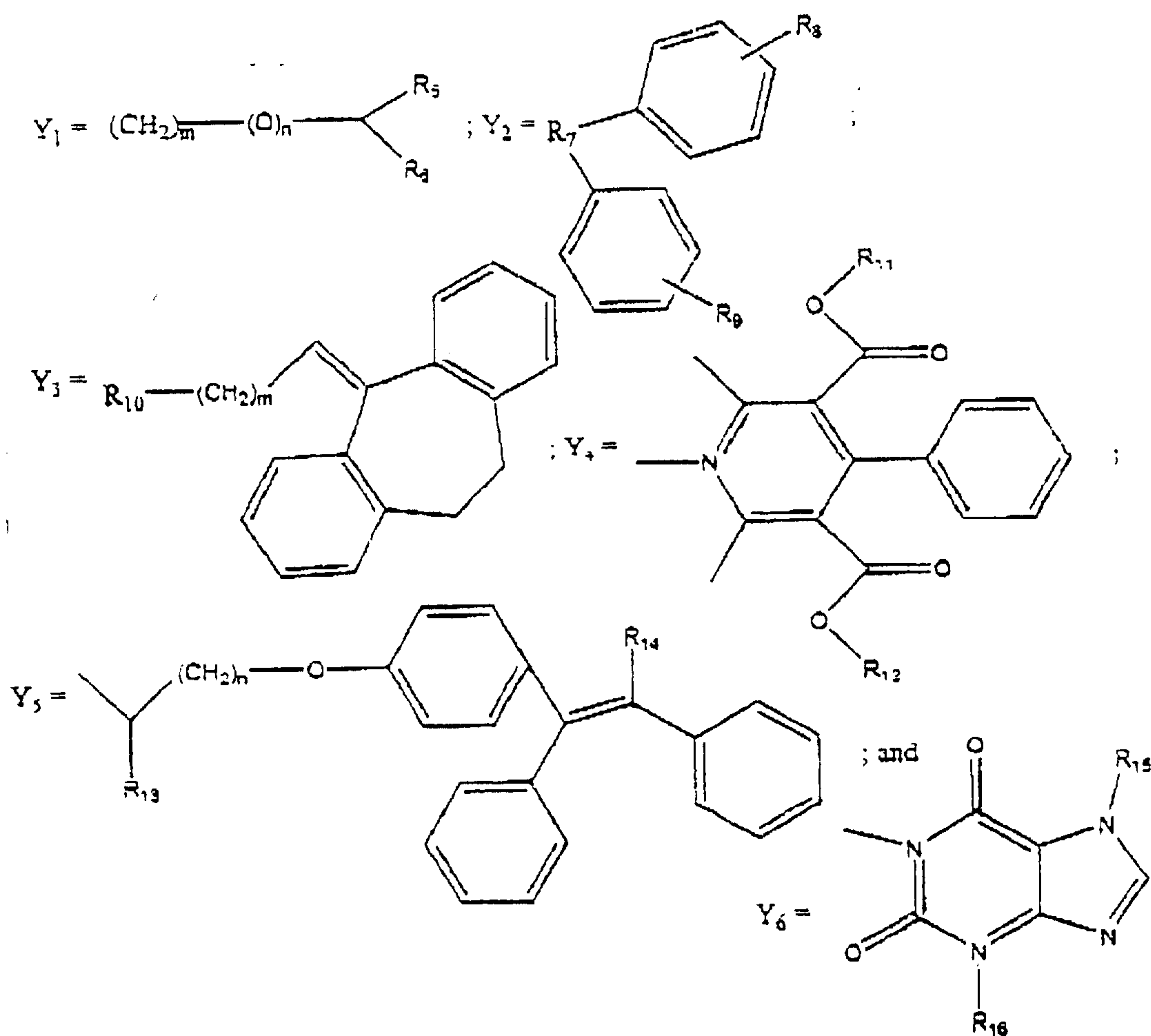
wherein the compound is represented by the following general Formula (A):



where X is selected from



and Y is selected from



wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 alkoxy, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems;

R_8 and R_9 each are one or more independent substituents selected from the group consisting of halogen, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, C1-C5 haloalkyl, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein

(i) if R_1 , R_2 , R_8 , or R_9 is piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from the group consisting of C1-C5

alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, and substituted or unsubstituted C1-C5 alkylamino;

(ii) if R_1 , R_2 , R_4 , or R_9 is alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl; and

(iii) if R_1 , R_2 , R_4 , or R_9 is a fused ring system, each individual R_1 , R_2 , R_4 , or R_9 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from the group consisting of naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; wherein each ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_3 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur, and oxygen;

R_4 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, and oxygen;

R_5 or R_6 are independently selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl, and anthracenyl;

R_7 is nitrogen or carbon;

R_{10} is selected from the group consisting of amino, C1-C5 substituted amino, and CH_2 ;

R_{11} and R_{12} each are independent and selected from the group consisting of hydrogen and C1-C5 alkyl;

R_{13} is selected from hydrogen and C1-C5 alkyl;

R_{14} is selected from hydrogen and C1-C5 alkyl;

R_{15} and R_{16} each are independent and selected from hydrogen and C1-C5 alkyl;

n is an integer of from 1 to 5; and

m is an integer of from 0 to 5 for Y_2 , Y_3 , Y_4 , and Y_5 , and m is an integer selected from the group consisting of 0, 1, 3, 4, and 5 for Y_1 ;

or a pharmaceutically acceptable salt of such compound.

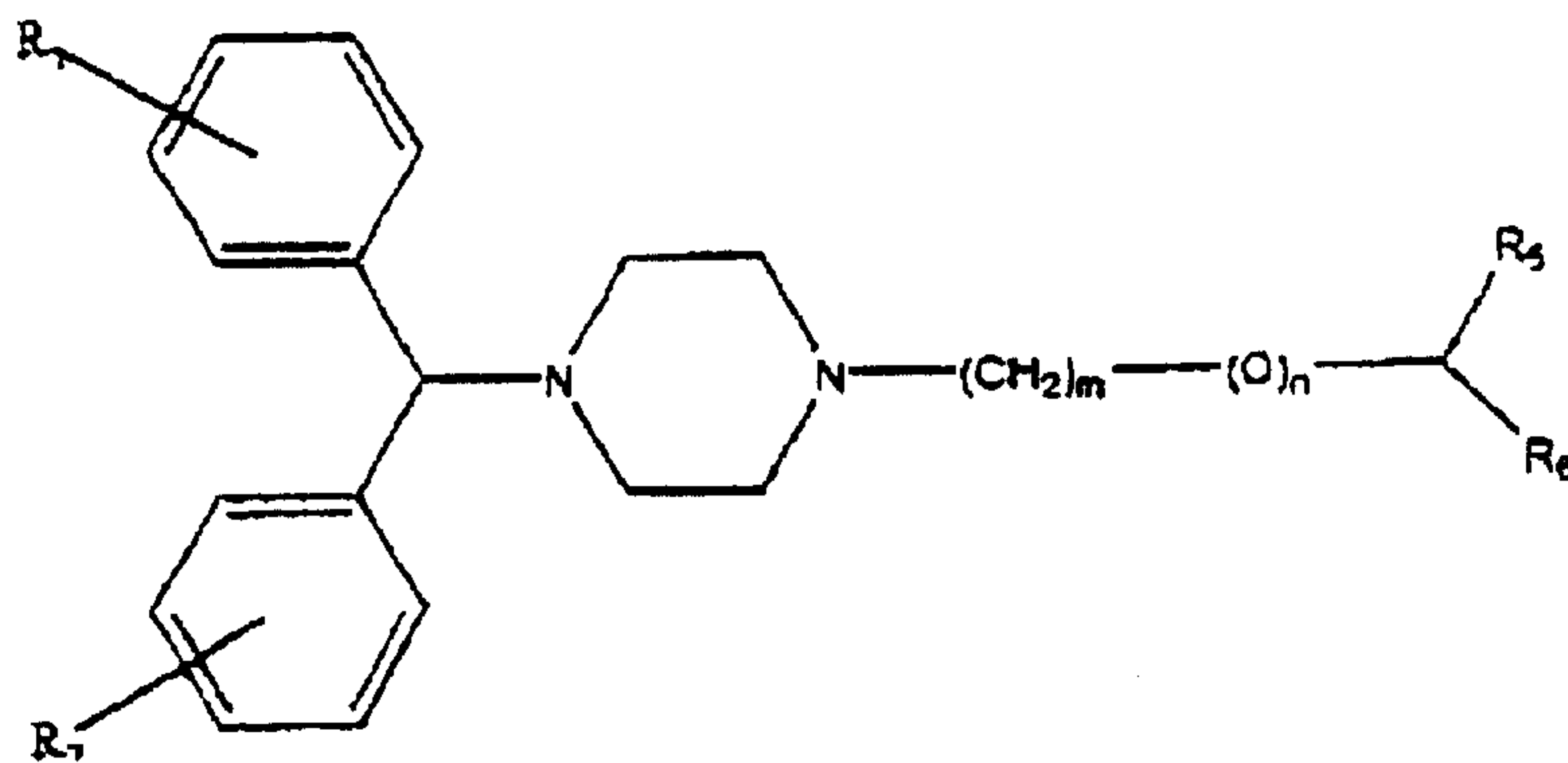
2. A composition as claimed in claim 1, wherein the composition displays an adequate ability to traverse the blood-brain barrier.

3. A composition as claimed in claim 1, whereby the compound prevents the DMS from disrupting.

4. A composition as claimed in claim 1, whereby the compound impedes disruption of intact DMS by acting on DMS prior to disruption in such a way that when disrupted, the compounds, when compared to controls:

- (1) reduce the mean volume of tissue occupied by disrupted DMS;
- (2) reduce the ratio of the number of inflammatory cells per DMS; or
- (3) increase the ratio of the number of macrophages containing disrupted DMS per DMS.

5. The composition as claimed in claim 1, wherein the compound is represented by the following general Formula (I):



-107-

wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, nitro, and halogen;

R_5 and R_6 are independently selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl, and anthracenyl;

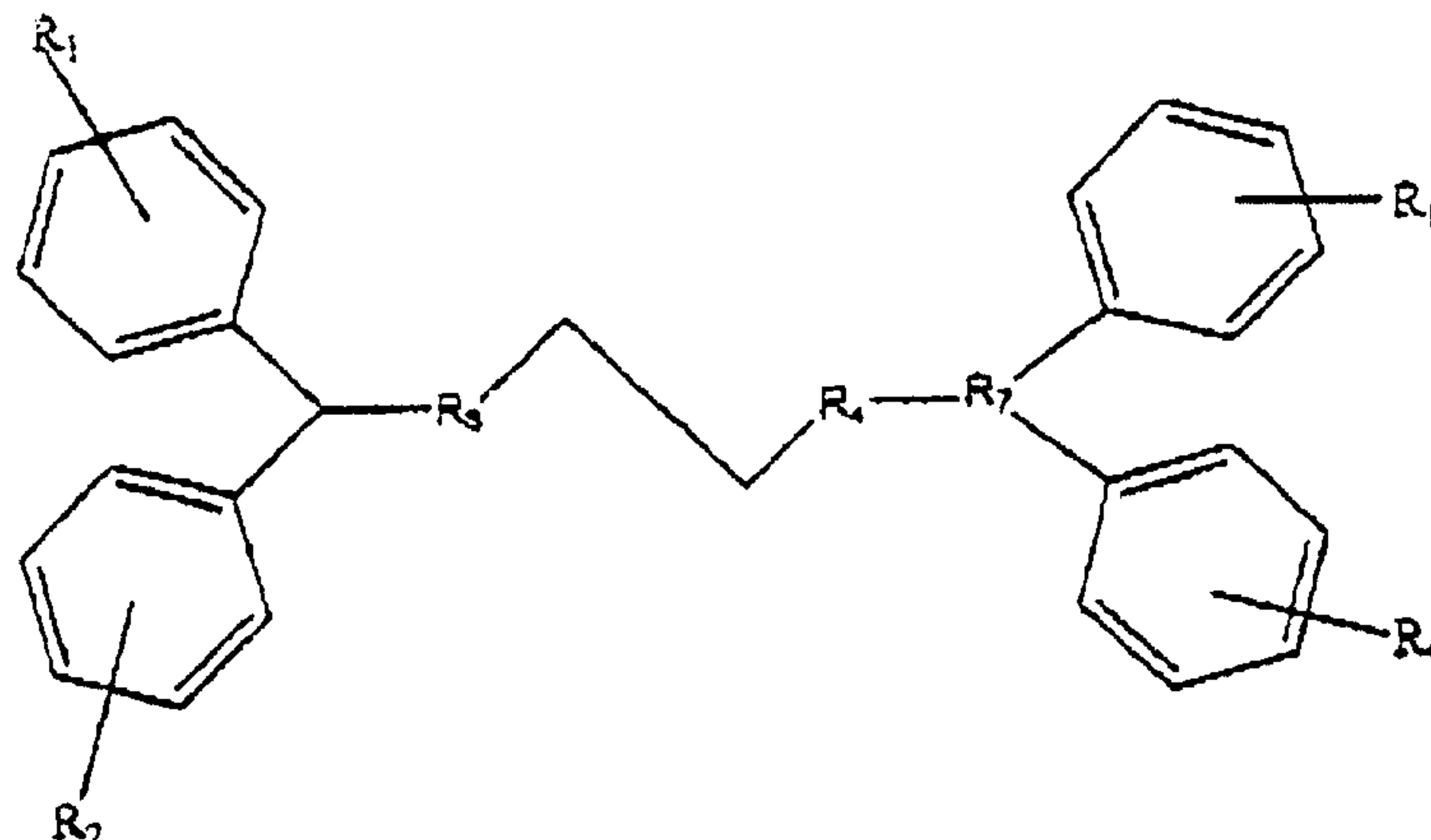
wherein each aromatic ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

m is an integer selected from the group consisting of 0, 1, 3, 4, and 5;

n is 1; or

a pharmaceutically acceptable salt of such compound.

6. The composition as claimed in claim 1, wherein the compound is represented by the following general formula (II):



wherein:

R_1 and R_2 are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems;

AMENDED SHEET

R_3 and R_9 each are one or more independent substituents selected from the group consisting of halogen, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, C1-C5 haloalkyl, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein

(i) if any of R_1 , R_2 , R_8 , and R_9 are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from the group consisting of C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, and substituted or unsubstituted C1-C5 alkylamino, or

(ii) if any of R_1 , R_2 , R_8 , and R_9 are alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with one or more compounds selected from the group consisting of C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl, or C1-C5 alkyl, and

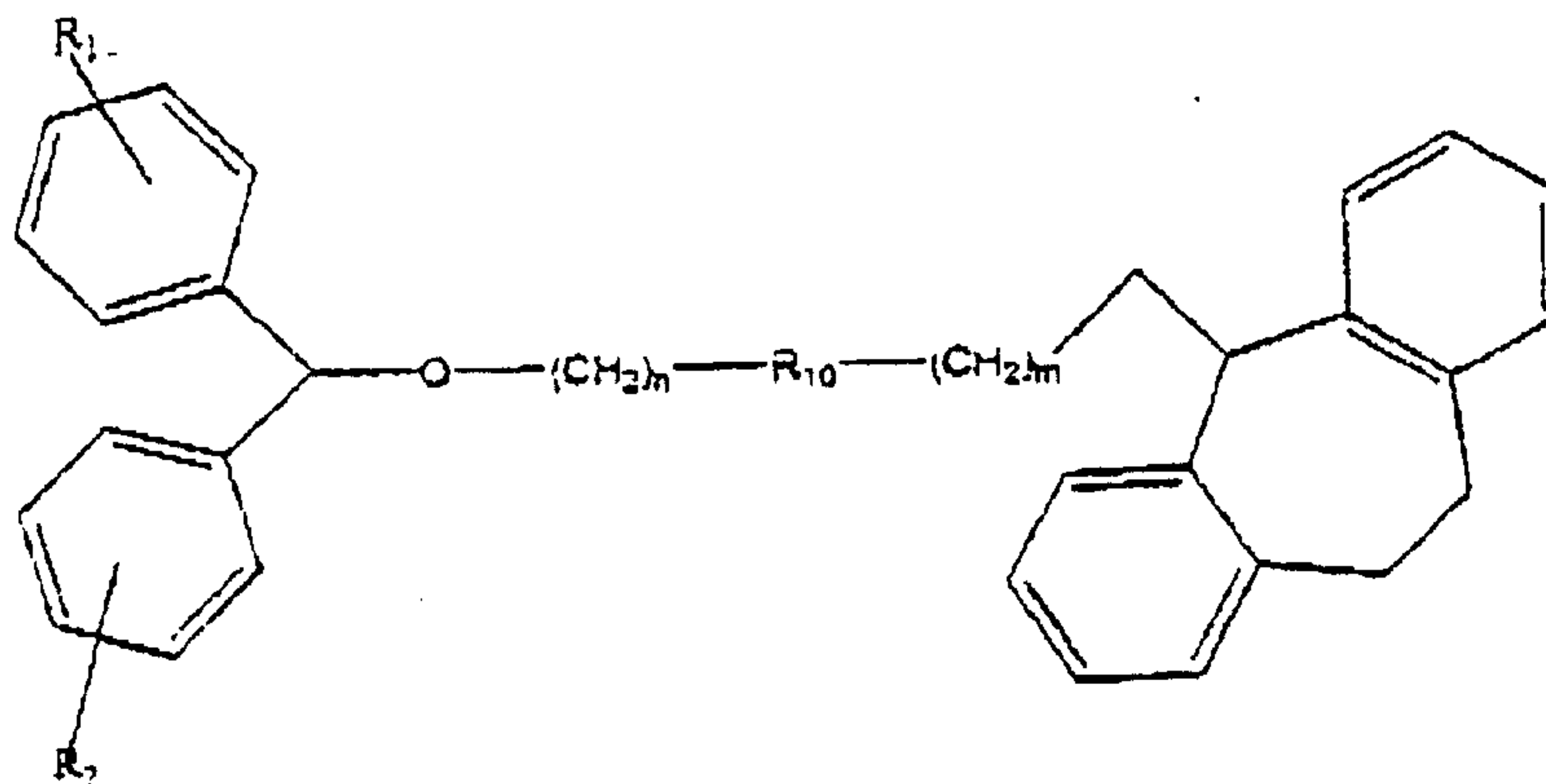
(iii) if any of R_1 , R_2 , R_8 , and R_9 is a fused ring system, each individual R_1 , R_2 , R_8 and R_9 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from the group consisting of naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part, and wherein each ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_3 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur, and oxygen;

R_4 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, and oxygen; and

R_7 is nitrogen or carbon; or
a pharmaceutically acceptable salt of such compound.

7. The composition as claimed in claim 1, wherein the compound is represented by the following general formula (III):



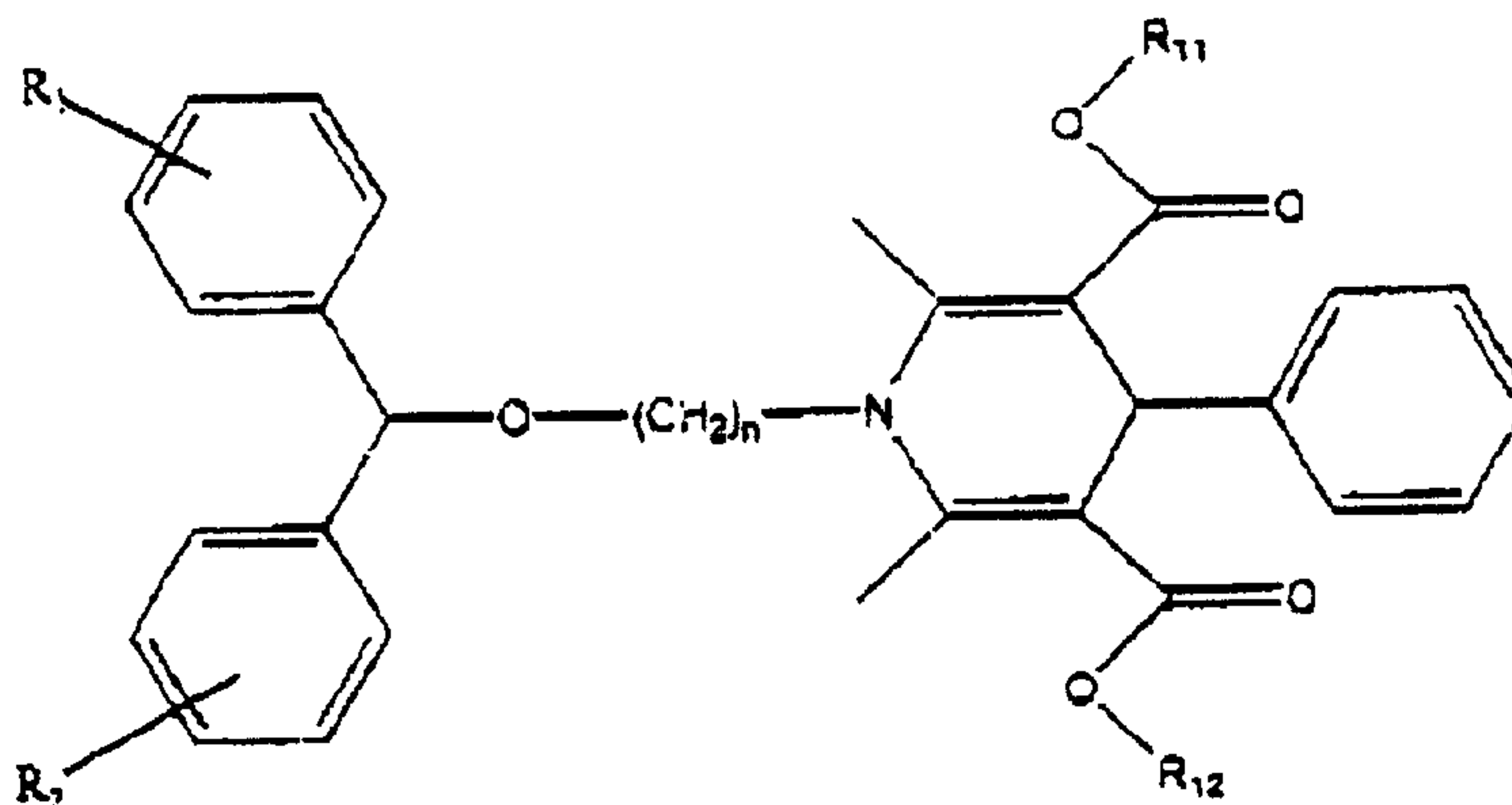
wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{10} is selected from the group consisting of amino, C1-C5 substituted amino, and CH_2 ; and

n and m are independently an integer of from 0-5; or
a pharmaceutically acceptable salt of such compound.

8. The composition as claimed in claim 1, wherein the compound is represented by the following general formula (IV):



-110-

wherein:

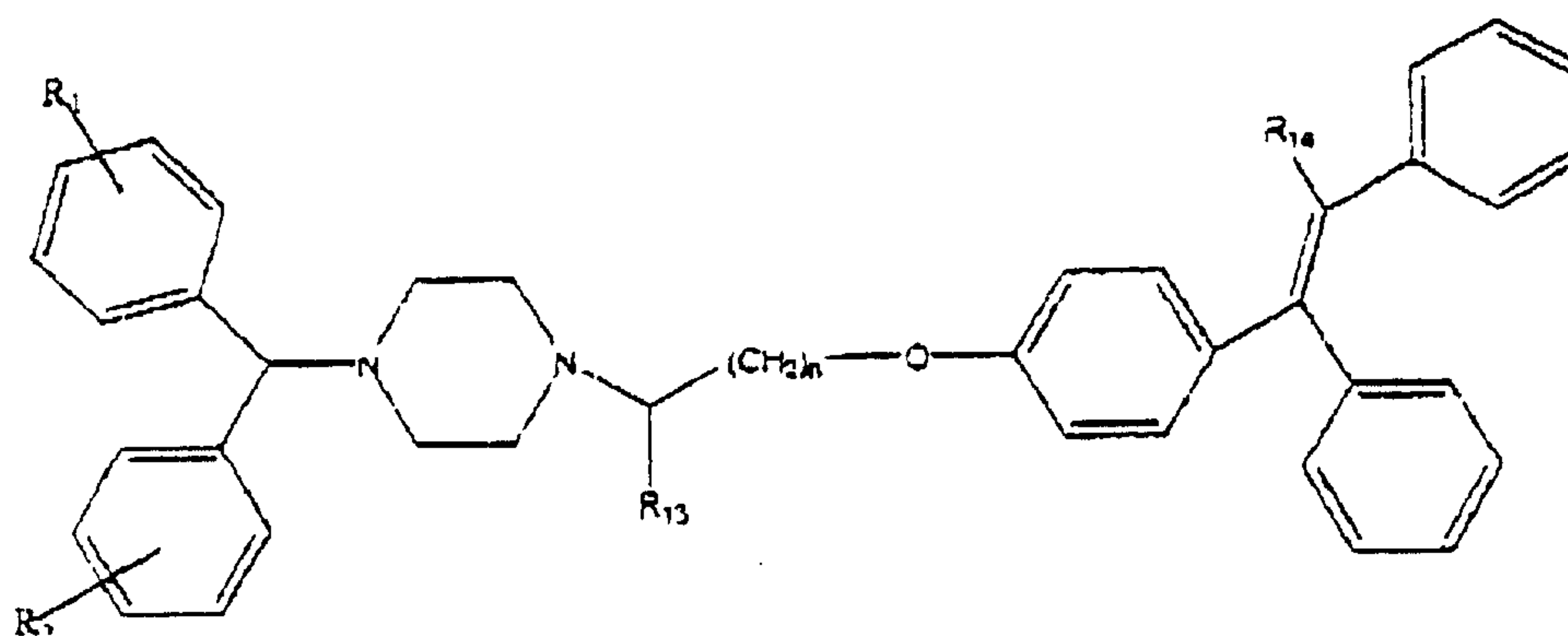
R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{11} and R_{12} each are independent and selected from the group consisting of hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5; or

a pharmaceutically acceptable salt of such compound.

9. The composition as claimed in claim 1, wherein the compound is represented by the following general formula (V):



wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{13} is selected from hydrogen and C1-C5 alkyl;

R_{14} is selected from hydrogen and C1-C5 alkyl; and

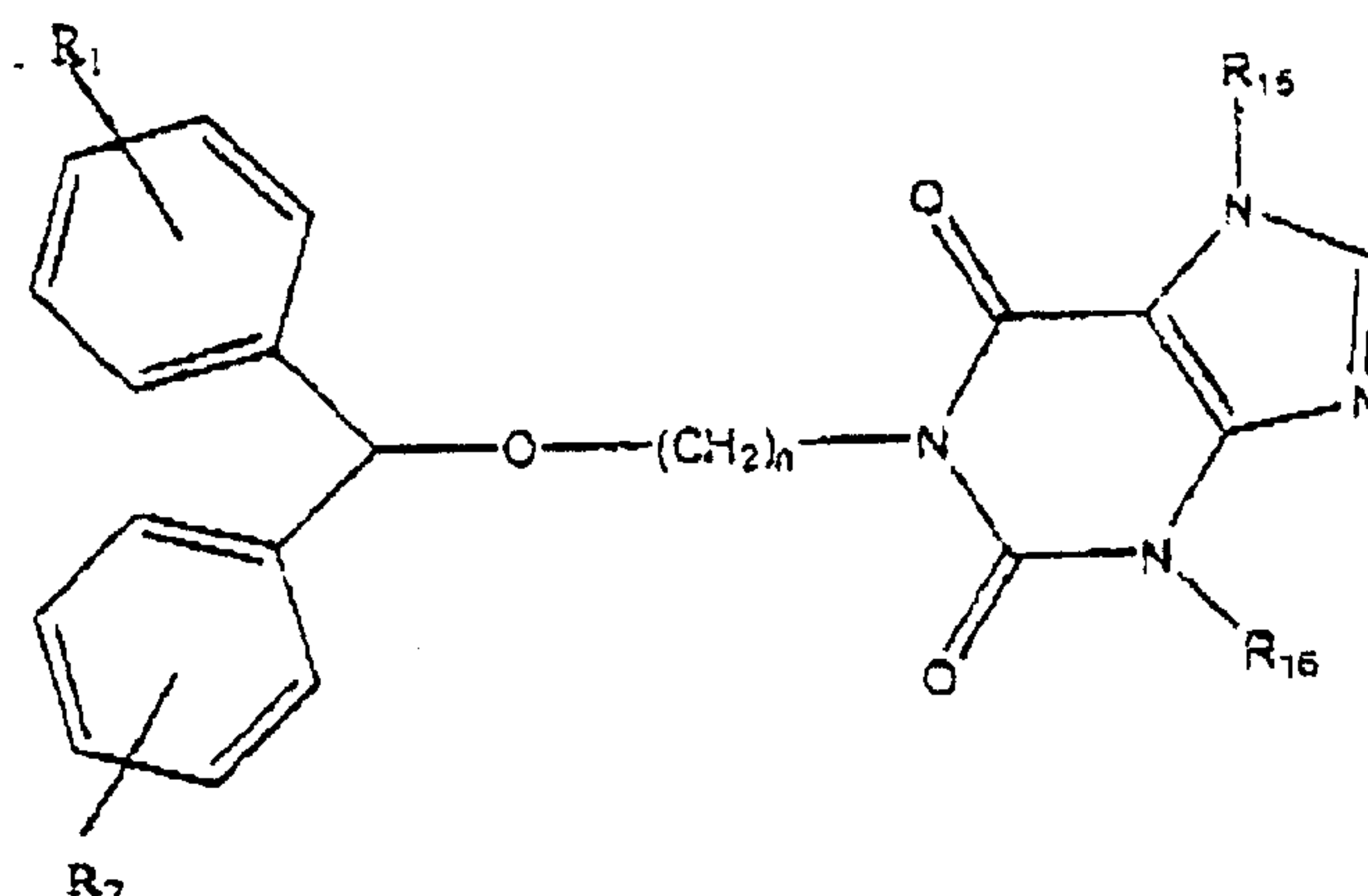
n is an integer of from 1 to 5 inclusive; or

a pharmaceutically acceptable salt of such compound.

AMENDED SHEET

-111-

10. The composition as claimed in claim 1, wherein the compound is represented by the following general formula (VI):



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R₁₅ and R₁₆ each are independent and selected from hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5 inclusive; or
a pharmaceutically acceptable salt of such compound.

11. A method for treating cerebral amyloidosis, comprising the step of administering to a subject, in whom disruption of intact DMS is anticipated, a pharmaceutically effective amount of a composition according to claim 1.

12. A method as claimed in claim 11, wherein the composition displays an adequate ability to traverse the blood-brain barrier.

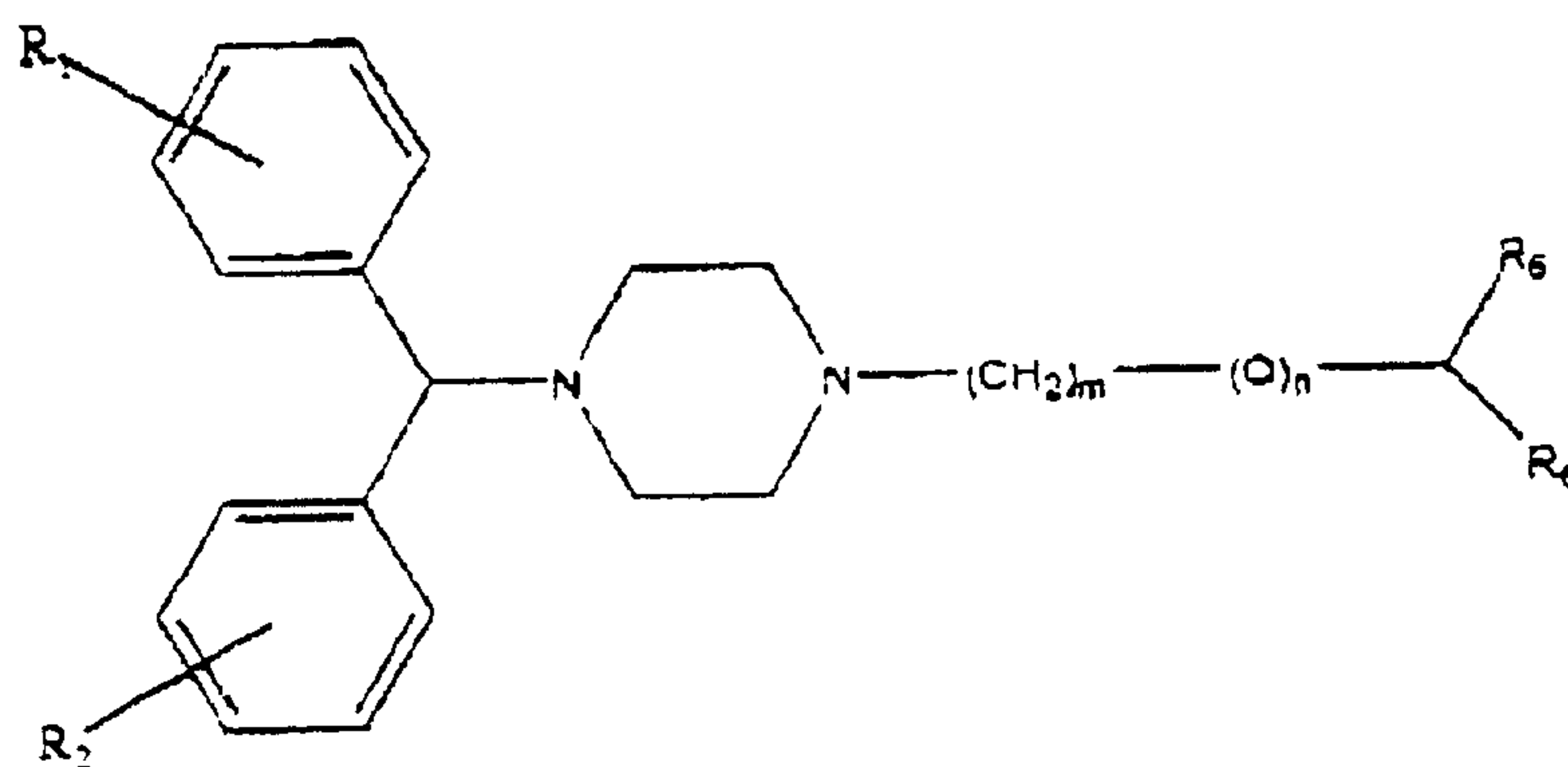
13. A method as claimed in claim 11, whereby said method prevents the DMS from disrupting.

-112-

14. A method as claimed in claim 11, whereby said method impedes disruption of intact DMS by acting on DMS prior to disruption in such a way that when disrupted, the compounds, when compared to controls:

- (1) reduce the mean volume of tissue occupied by disrupted DMS;
- (2) reduce the ratio of the number of inflammatory cells per DMS; or
- (3) increase the ratio of the number of macrophages containing disrupted DMS per DMS.

15. The method as claimed in claim 11, wherein the compound is represented by the following general Formula (1):



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, nitro, and halogen;

R₅ and R₆ are independently selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl, and anthracenyl;

wherein each aromatic ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

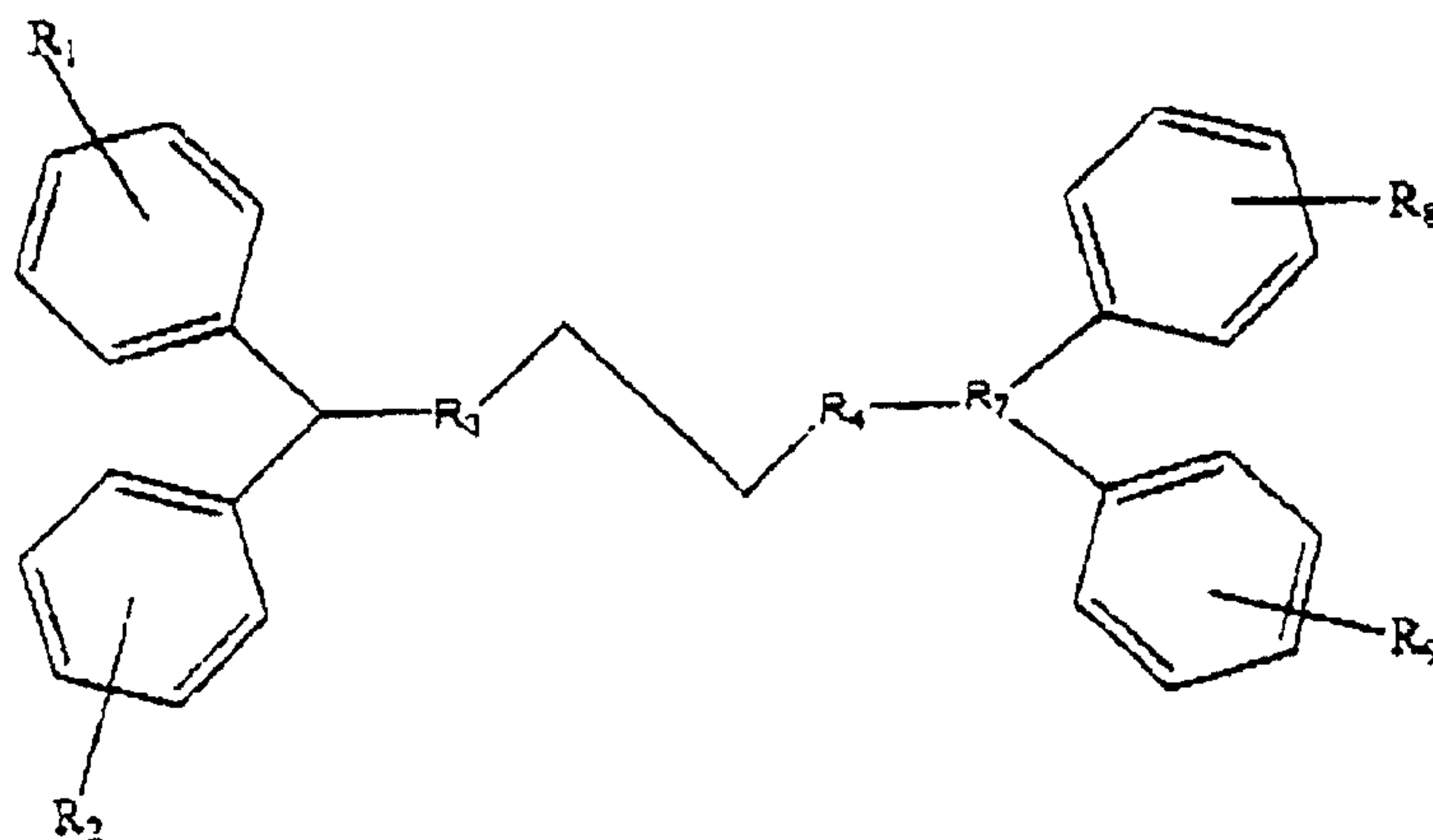
m is an integer selected from the group consisting of 0, 1, 3, 4, and 5; and

n is 1; or

-113-

a pharmaceutically acceptable salt of such compound.

16. The method as claimed in claim 11, represented by the following general formula (II):



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems;

R₈ and R₉ each are one or more independent substituents selected from the group consisting of halogen, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein

(i) if any of R₁, R₂, R₈, and R₉ is piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from the group consisting of C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, and substituted or unsubstituted C1-C5 alkylamino;

(ii) if any of R₁, R₂, R₈, and R₉ is alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with one or more compounds selected from the group consisting of C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl, or C1-C5 alkyl, and

-114-

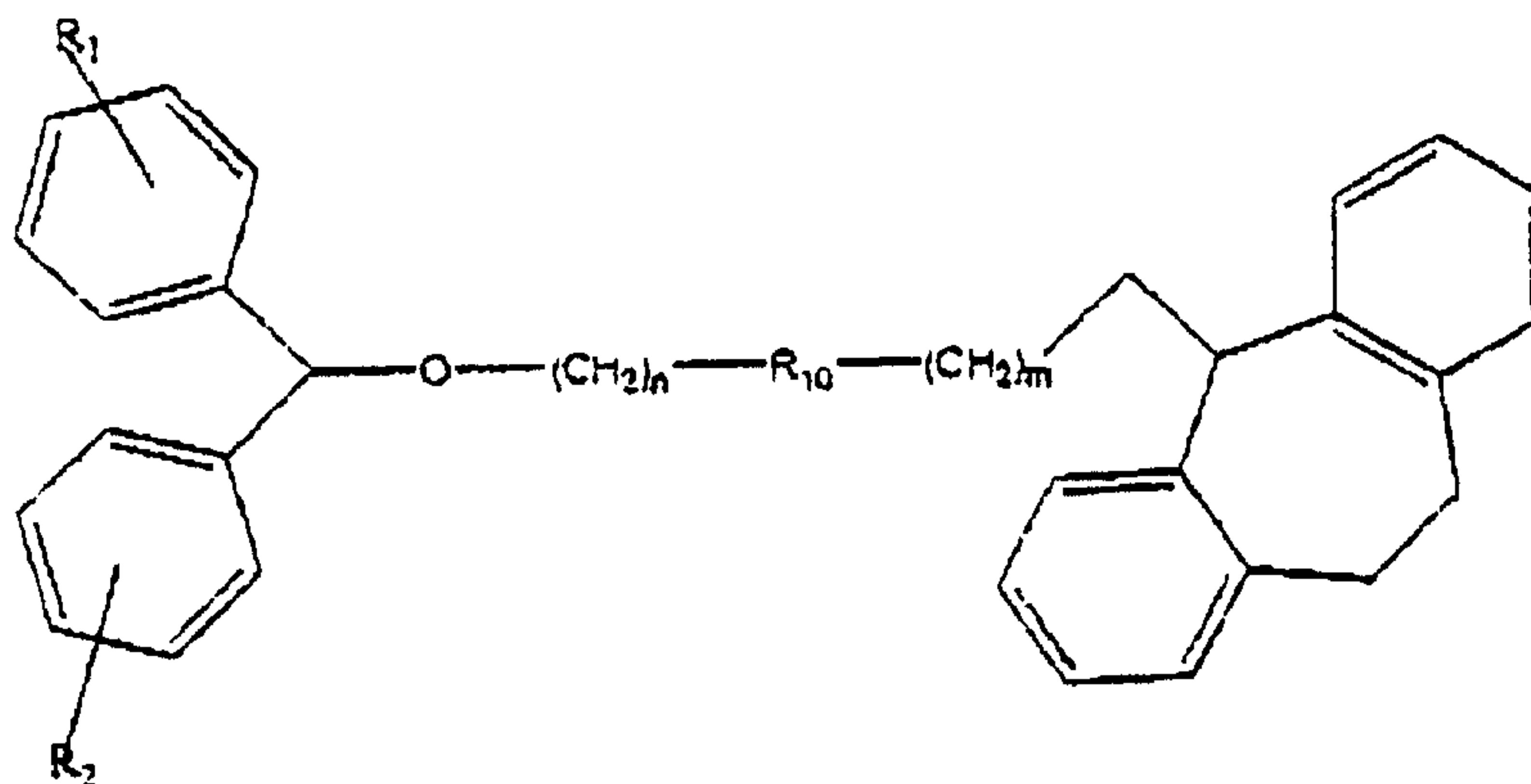
(iii) if any of R_1 , R_2 , R_3 , and R_9 is a fused ring system, each individual R_1 , R_2 , R_3 , and R_9 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from the group consisting of naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part, and wherein each ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkynyl, C2-C5 alkenyl, and C1-C5 haloalkyl;

R_3 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur, and oxygen;

R_4 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, and oxygen; and

R_7 is nitrogen or carbon; or
a pharmaceutically acceptable salt of such compound.

17. The method as claimed in claim 11, wherein the compound is represented by the following general formula (III):



AMENDED SHEET

-115-

wherein:

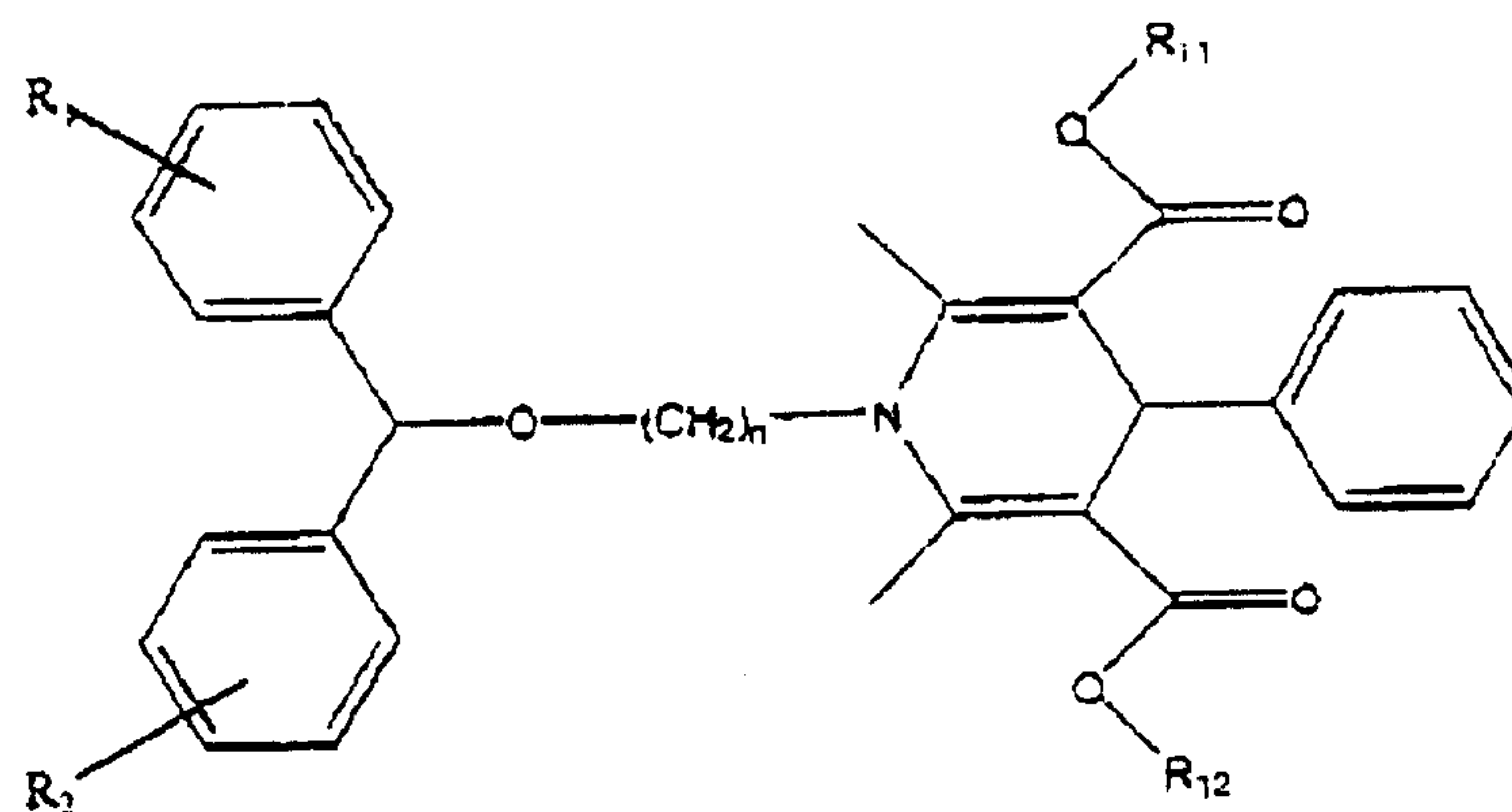
R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{10} is selected from the group consisting of amino, C1-C5 substituted amino, and CH_2 ; and

n and m are independently an integer of from 0-5; or

a pharmaceutically acceptable salt of such compound.

18. The method as claimed in claim 11, wherein the compound is represented by the following general formula (IV):



wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

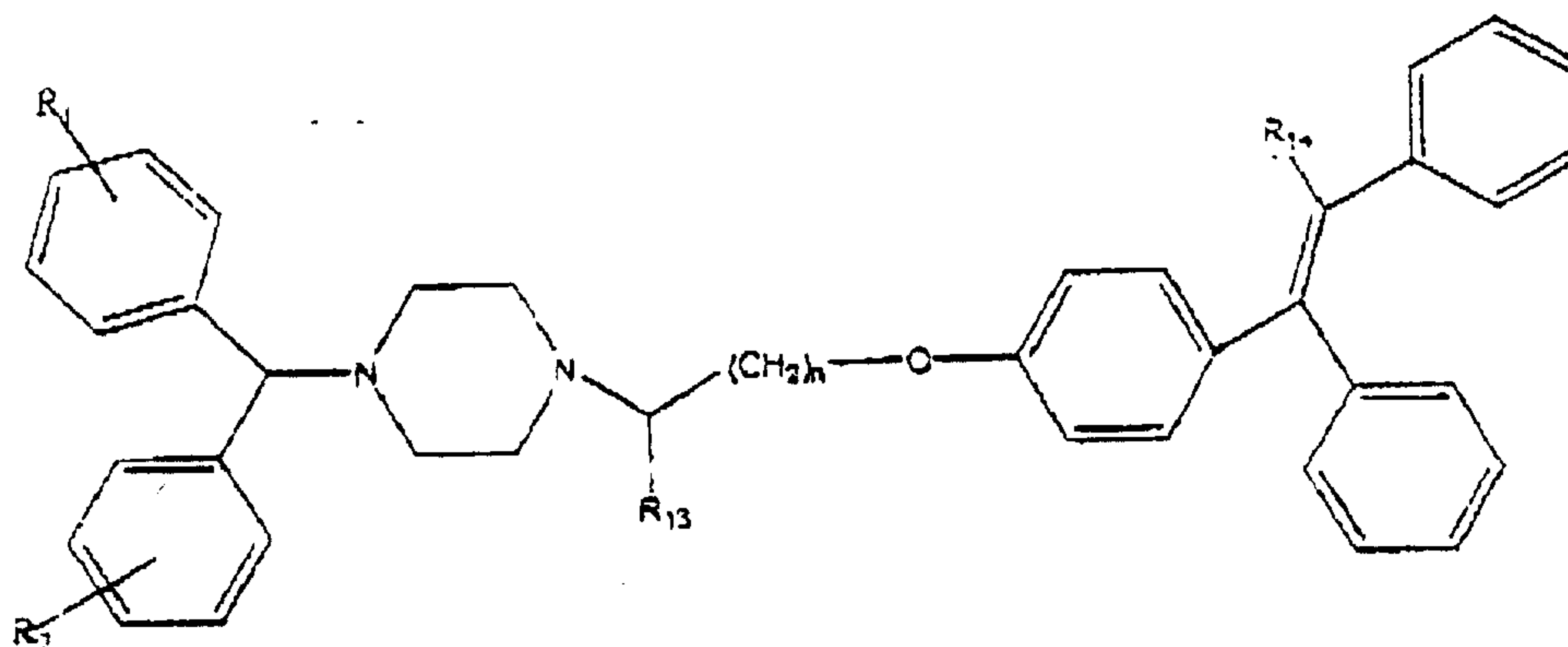
R_{11} and R_{12} each are independent and selected from the group consisting of hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5; or

a pharmaceutically acceptable salt of such compound.

AMENDED SHEET

19. The method as claimed in claim 11, wherein the compound is represented by the following general formula (V):



wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

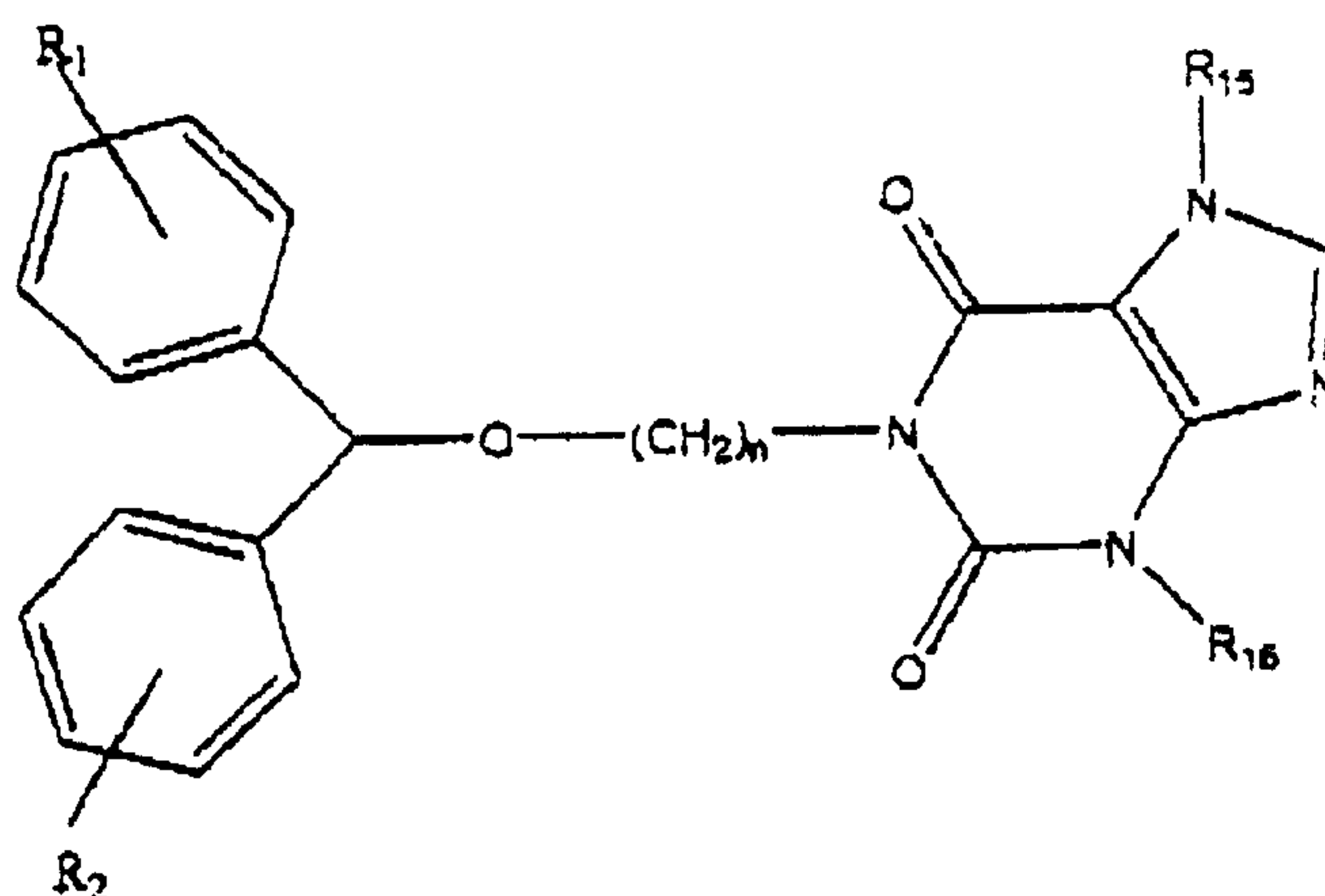
R_{13} is selected from hydrogen and C1-C5 alkyl;

R_{14} is selected from hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5 inclusive; or

a pharmaceutically acceptable salt of such compound.

20. The method as claimed in claim 11, wherein the compound is represented by the following general formula (VI):



-117-

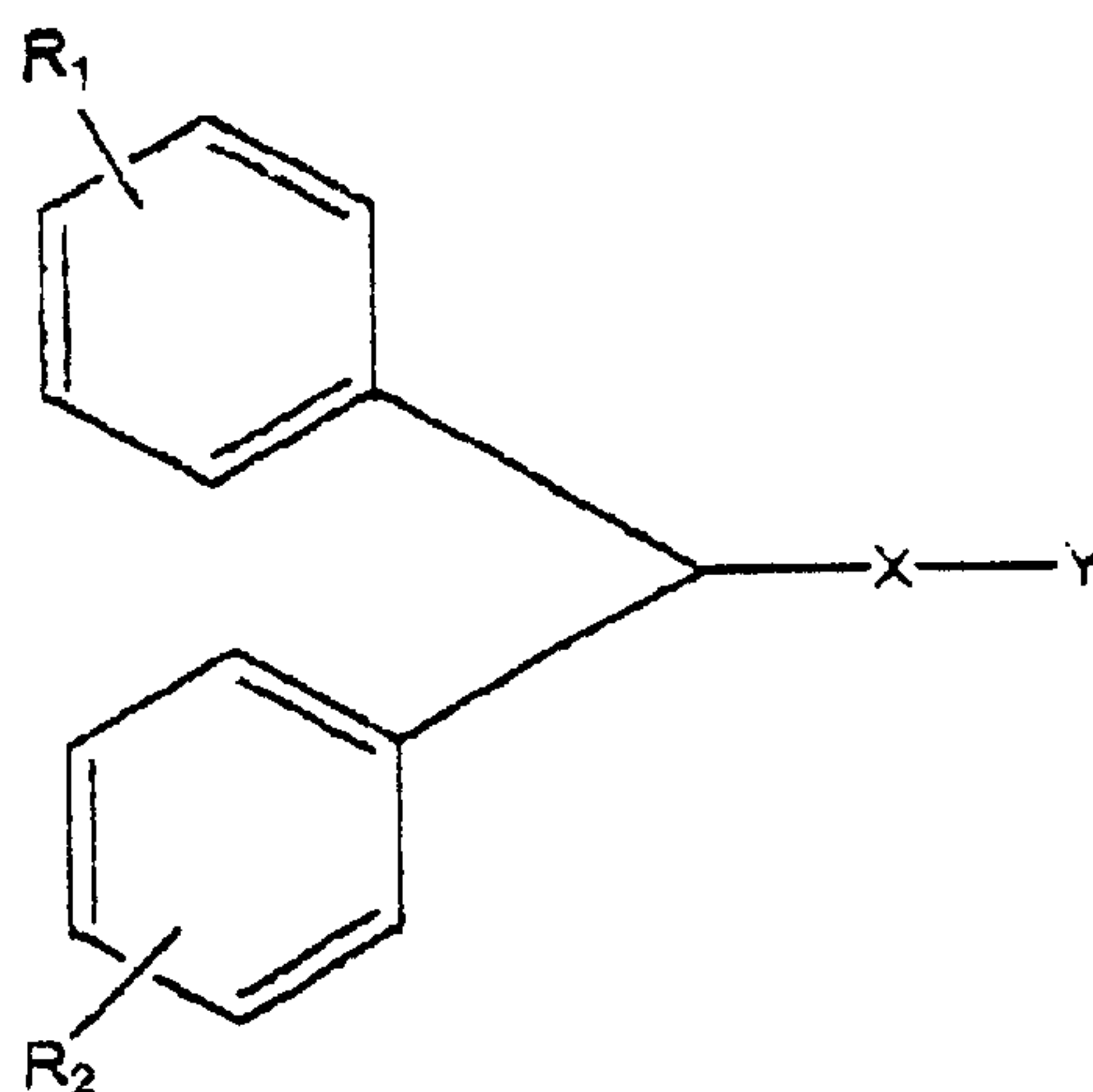
wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

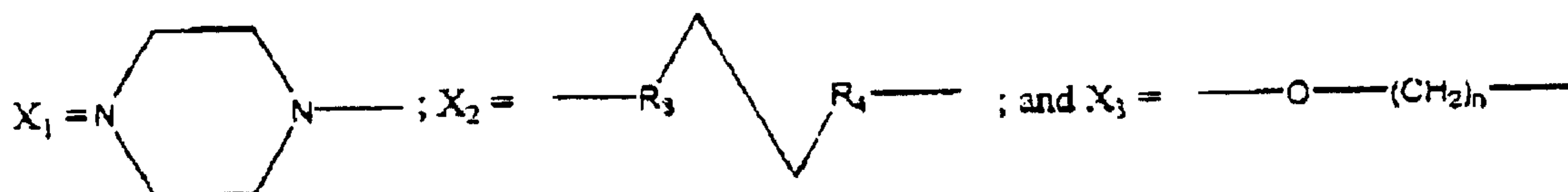
R_{15} and R_{16} each are independent and selected from hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5 inclusive; or
a pharmaceutically acceptable salt of such compound.

21. A compound represented by the general formula (A):

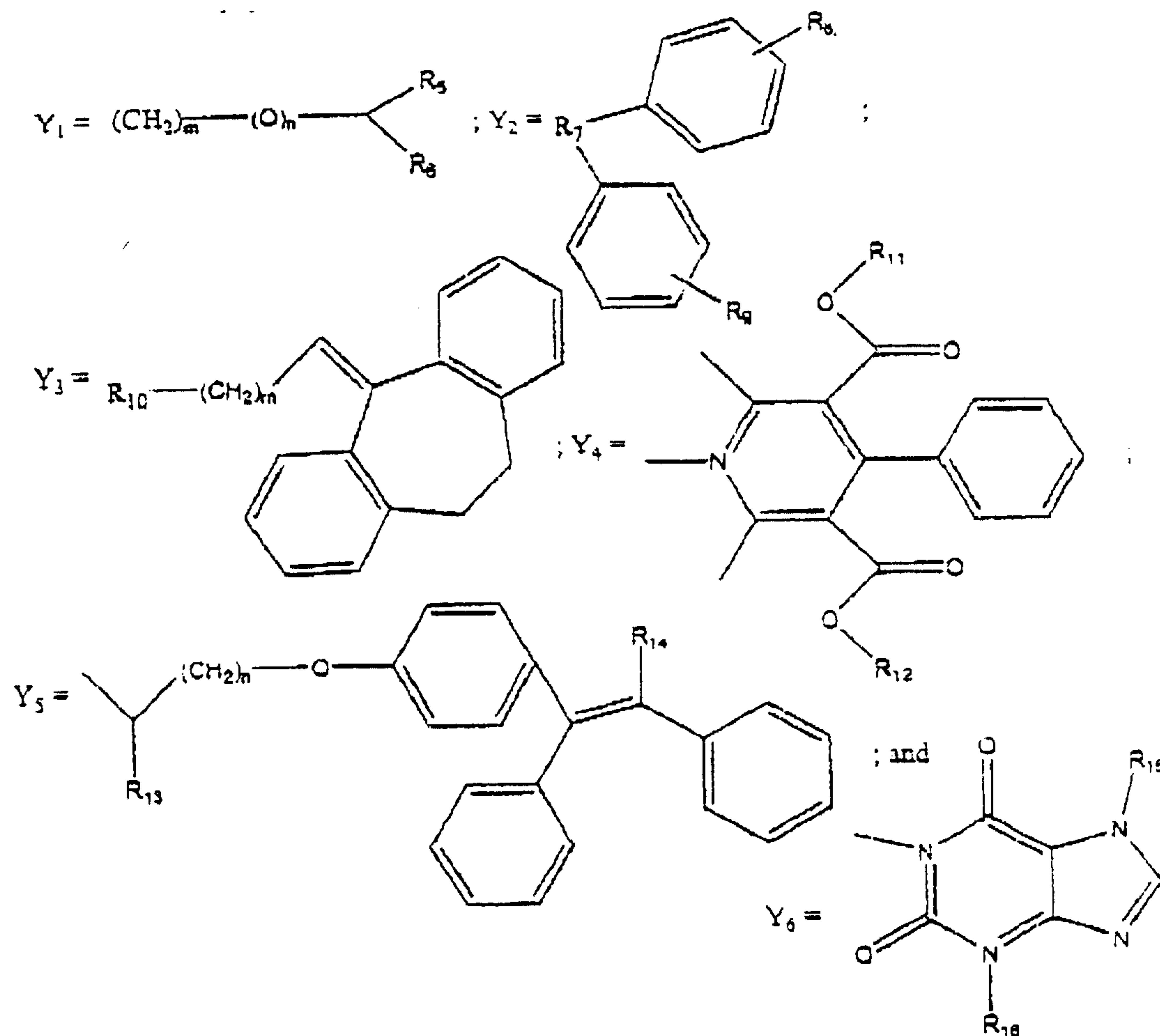


where X is selected from



-118-

and Y is selected from



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 alkoxy, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems;

R₈ and R₉ each are one or more independent substituents selected from the group consisting of halogen, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, C1-C5 haloalkyl, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein

(i) if R_1 , R_2 , R_3 , or R_9 is piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from the group consisting of C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, and substituted or unsubstituted C1-C5 alkylamino;

(ii) if R_1 , R_2 , R_3 , or R_9 is alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or di-substituted with C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl or C1-C5 alkyl; and

(iii) if R_1 , R_2 , R_3 , or R_9 is a fused ring system, each individual R_1 , R_2 , R_3 , or R_9 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from the group consisting of naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, aceanthrylene, isoindole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quiazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part; wherein each ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_2 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur, and oxygen;

R_4 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, and oxygen;

R_5 or R_6 are independently selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl, and anthracenyl;

R_7 is nitrogen or carbon;

R_{10} is selected from the group consisting of amino, C1-C5 substituted amino and CH_2 ;

R_{11} and R_{12} each are independent and selected from the group consisting of hydrogen and C1-C5 alkyl;

R_{13} is selected from hydrogen and C1-C5 alkyl;

R_{14} is selected from hydrogen and C1-C5 alkyl;

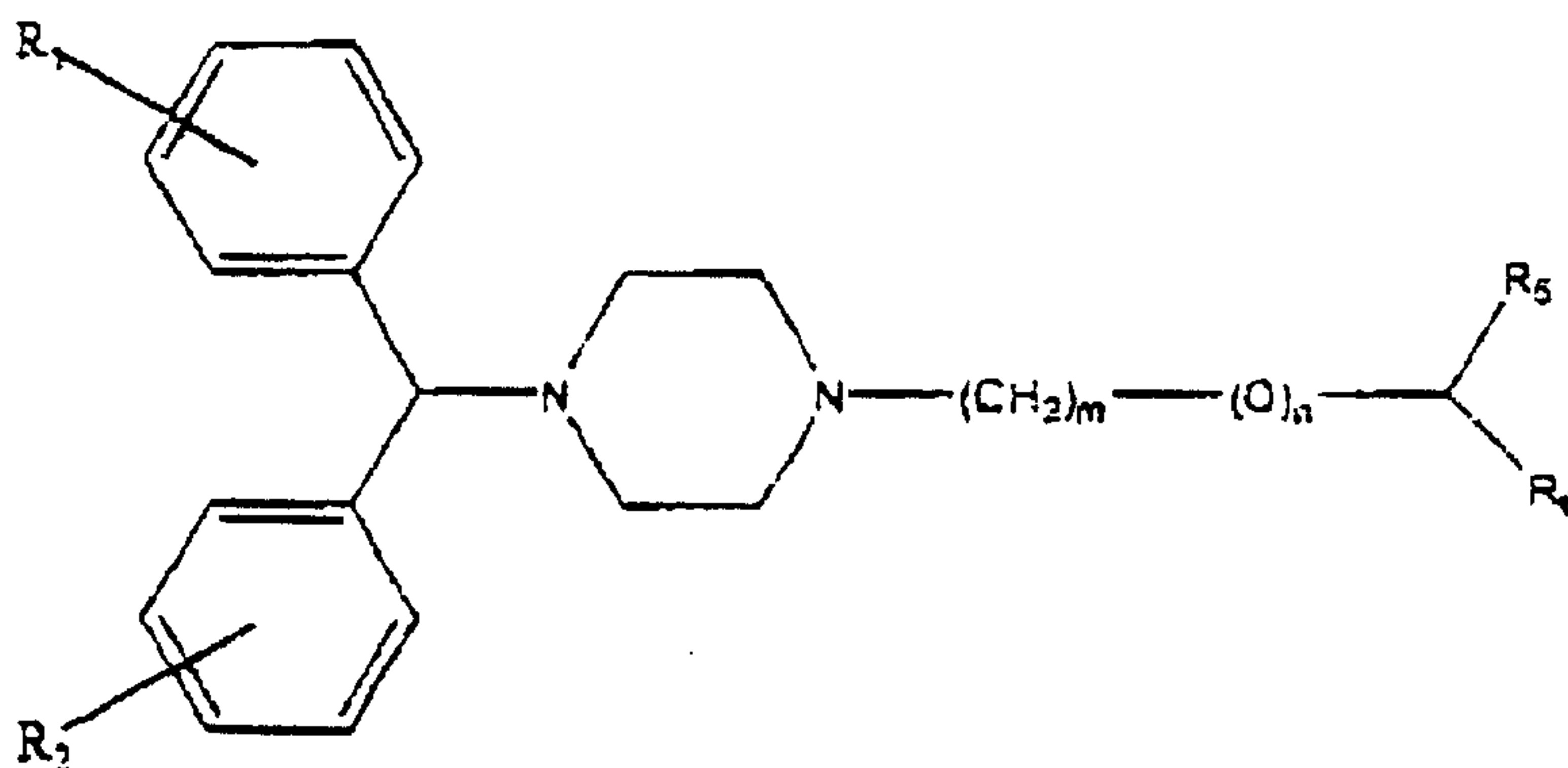
R_{15} and R_{16} each are independent and selected from hydrogen and C1-C5 alkyl;

n is an integer of from 0 to 5 for Y_2 , Y_3 , Y_4 , Y_5 , and Y_6 , and n is an integer from 1 to 5 for Y_1 ; and

m is an integer of from 0 to 5 for Y_2 , Y_3 , Y_4 , and Y_5 , and m is an integer selected from the group consisting of 0, 1, 3, 4, and 5 for Y_1 ;

or a pharmaceutically acceptable salt of such compound.

22. The compound as claimed in claim 21, wherein the compound is represented by the following general Formula (I):



wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, nitro, and halogen;

R_5 or R_6 are independently selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C2-C5 alkynyl, phenyl, benzyl, naphthyl, and anthracenyl;

wherein each aromatic ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

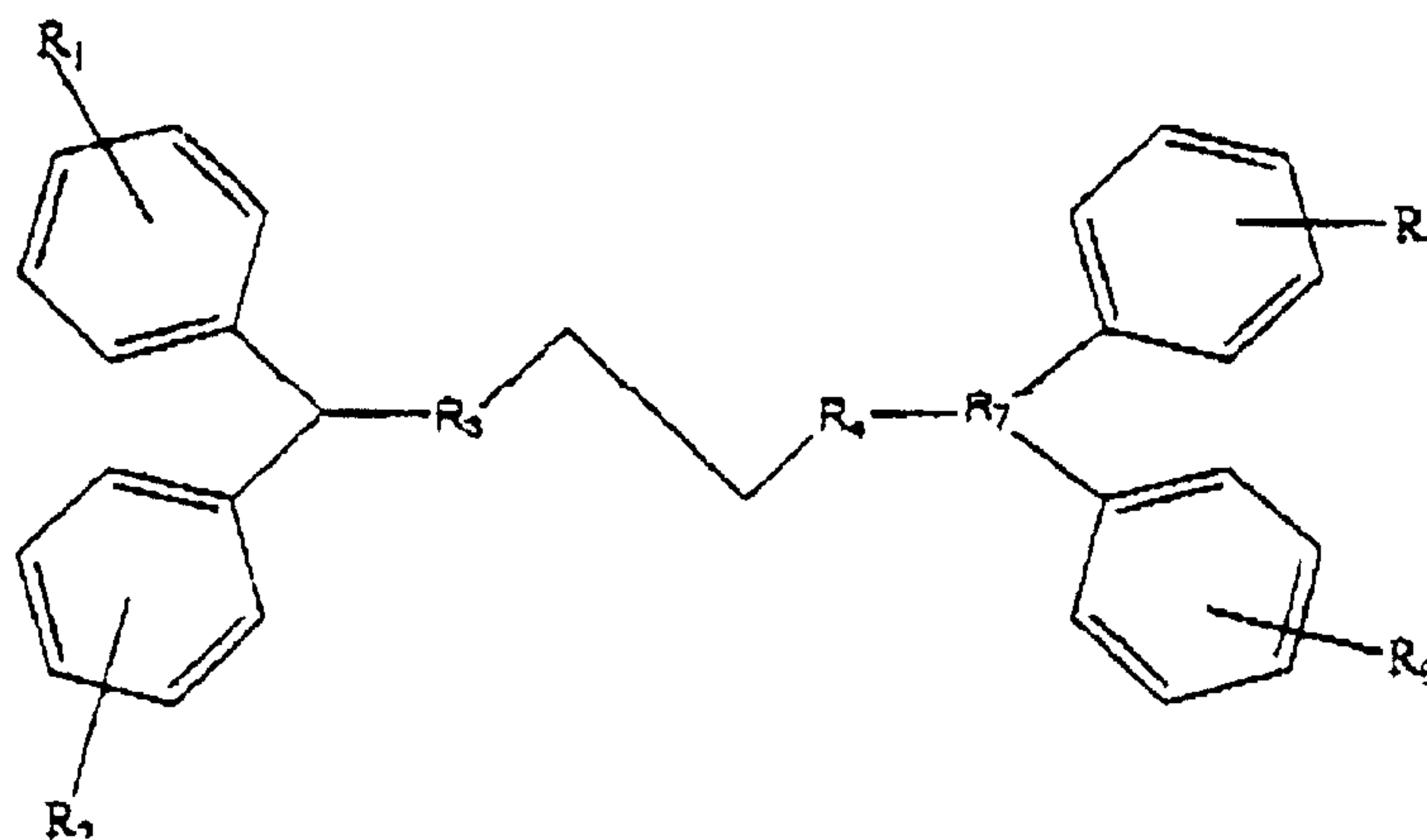
m is an integer selected from the group consisting of 0, 1, 3, 4, and 5; and

-121-

n is 1; or

a pharmaceutically acceptable salt of such compound.

23. The compound as claimed in claim 21, wherein the compound is represented by the following general formula (II):



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems;

R₈ and R₉ each are one or more independent substituents selected from the group consisting of halogen, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, C1-C5 haloalkyl, phenyl, nitro, carboxyl, piperazinyl, pyridyl, and fused ring systems; wherein

(i) if any of R₁, R₂, R₈ and R₉ are piperazinyl, a nitrogen atom on each piperazinyl may be substituted with a moiety selected from the group consisting of C1-C5 alkyl, C3-C5 cycloalkyl, C1-C5 haloalkyl, and substituted or unsubstituted C1-C5 alkylamino,

(ii) if any of R₁, R₂, R₈ and R₉ are alkylamino, each alkylamino consists of 1 to 5 carbon atoms and the amino group is unsubstituted or mono- or

di-substituted with one or more compounds selected from the group consisting of C3-C5 cycloalkyl, C2-C5 alkenyl, C2-C5 alkynyl, or C1-C5 alkyl, and

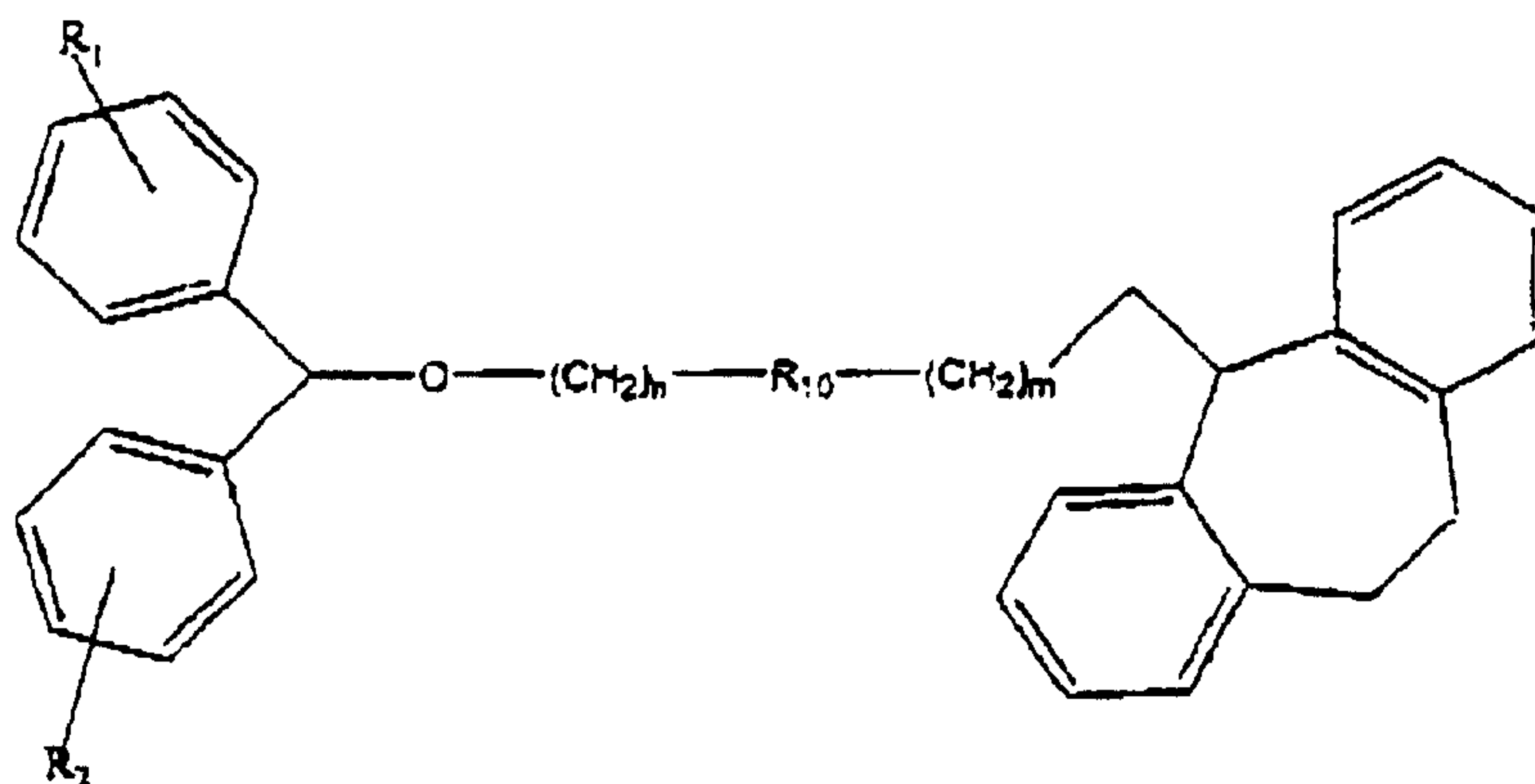
(iii) if any of R_1 , R_2 , R_8 and R_9 is a fused ring system, each individual R_1 , R_2 , R_8 and R_9 involved in the fused ring system, together with the phenyl to which it is attached, forms a fused ring system selected from the group consisting of naphthalene, anthracene, acenaphthylene, fluorene, phenalene, phenanthrene, fluoranthene, acephenanthrylene, aceanthrylene, isoincrole, indole, quinolizine, isoquinoline, phthalazine, quinoxaline, quinoline, phthalazine, quinazoline, and cinnoline, wherein one or more carbon atoms in the fused ring may be replaced with a nitrogen atom and each ring may be saturated or unsaturated, in whole or in part, and wherein each ring may be substituted by one or more substituents selected from the group consisting of halogen, C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, and C1-C5 haloalkyl;

R_3 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, sulfur, and oxygen;

R_4 is selected from the group consisting of C1-C5 alkyl, C2-C5 alkenyl, C2-C5 alkynyl, amino, C1-C5 alkyl substituted amino, C1-C5 alkylamino, C2-C5 alkenylamino, C1-C5 alkyl substituted C1-C5 alkylamino, C1-C5 alkyl substituted C2-C5 alkenylamino, sulfur, and oxygen; and

R_7 is nitrogen or carbon; or
a pharmaceutically acceptable salt of such compound.

24. The compound as claimed in claim 21, wherein the compound is represented by the following general formula (II):



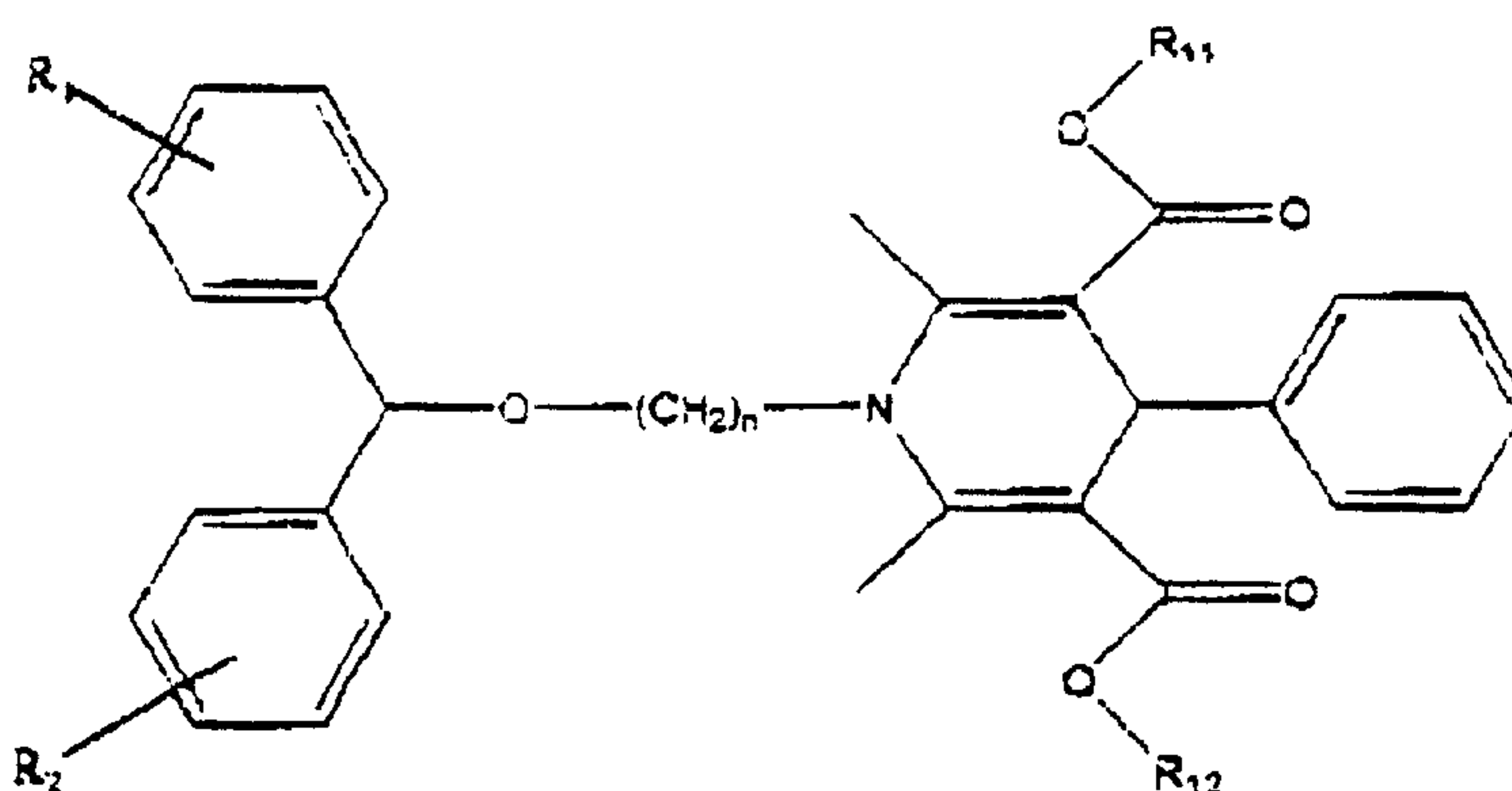
wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{10} is selected from the group consisting of amino, C1-C5 substituted amino, and CH_2 ; and

n and m are independently an integer of from 0-5; or
a pharmaceutically acceptable salt of such compound.

25. The compound as claimed in claim 21, wherein the compound is represented by the following general formula (IV):



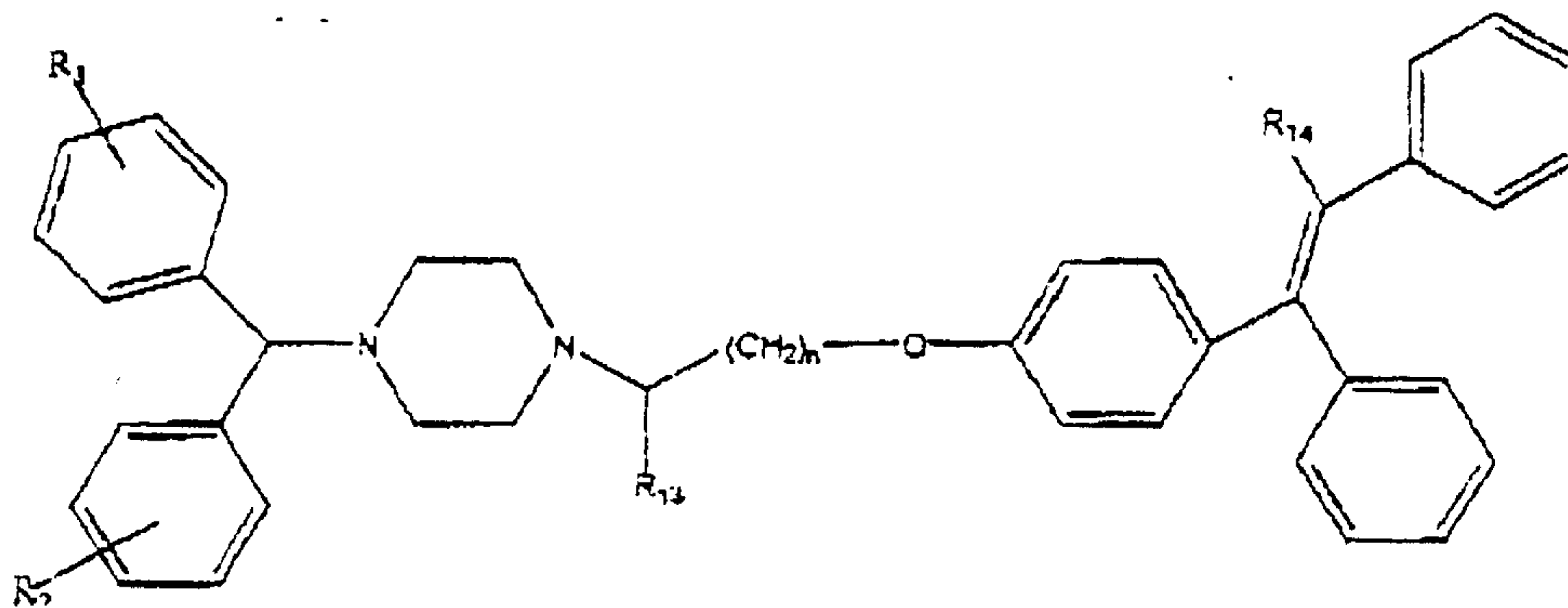
wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{11} and R_{12} each are independent and selected from the group consisting of hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5; or
a pharmaceutically acceptable salt of such compound.

26. The compound as claimed in claim 21, wherein the compound is represented by the following general formula (V):



wherein:

R₁ and R₂ each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

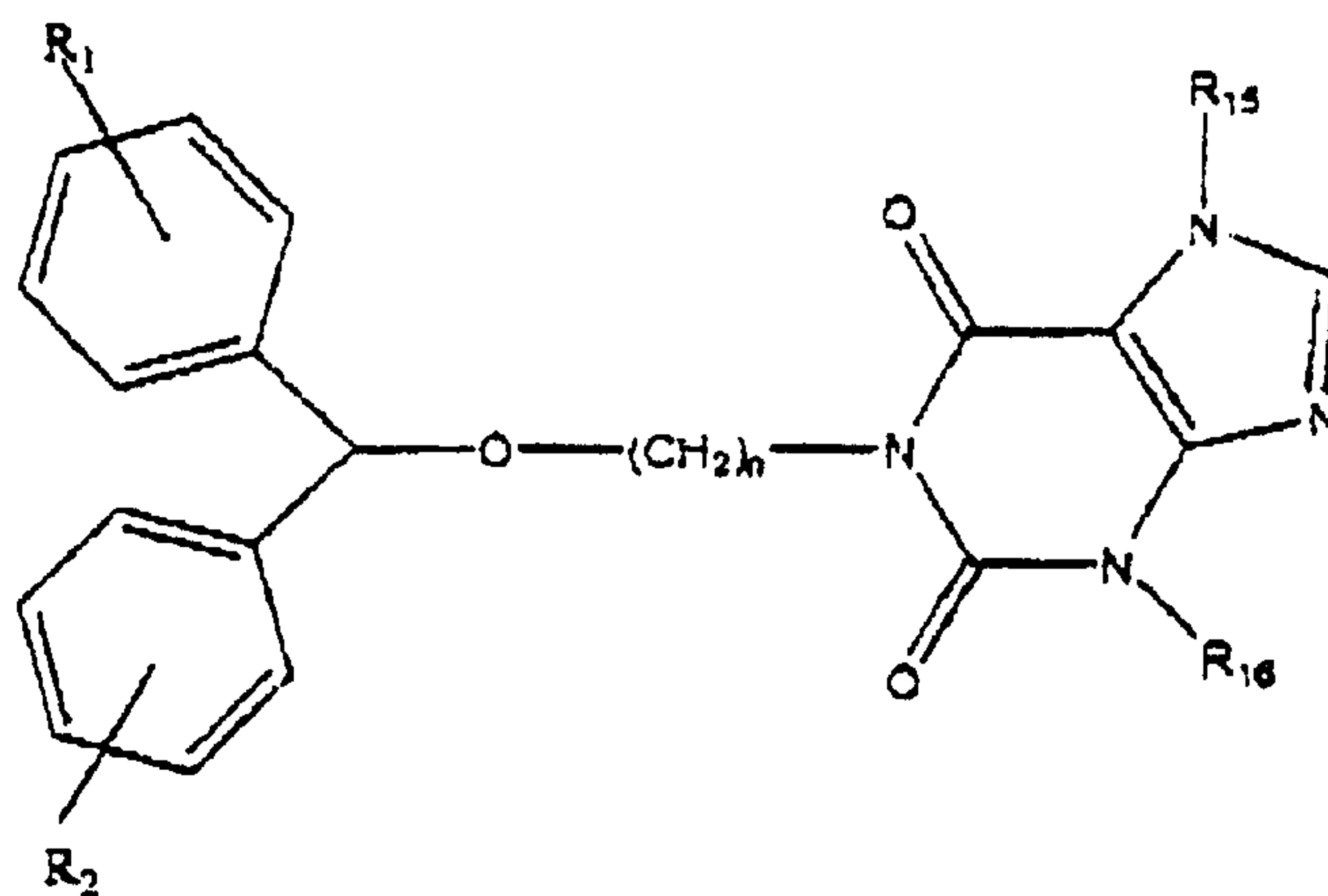
R₁₃ is selected from hydrogen and C1-C5 alkyl;

R₁₄ is selected from hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5 inclusive; or

a pharmaceutically acceptable salt of such compound.

27. The compound as claimed in claim 21, wherein the compound is represented by the following general formula (VI):



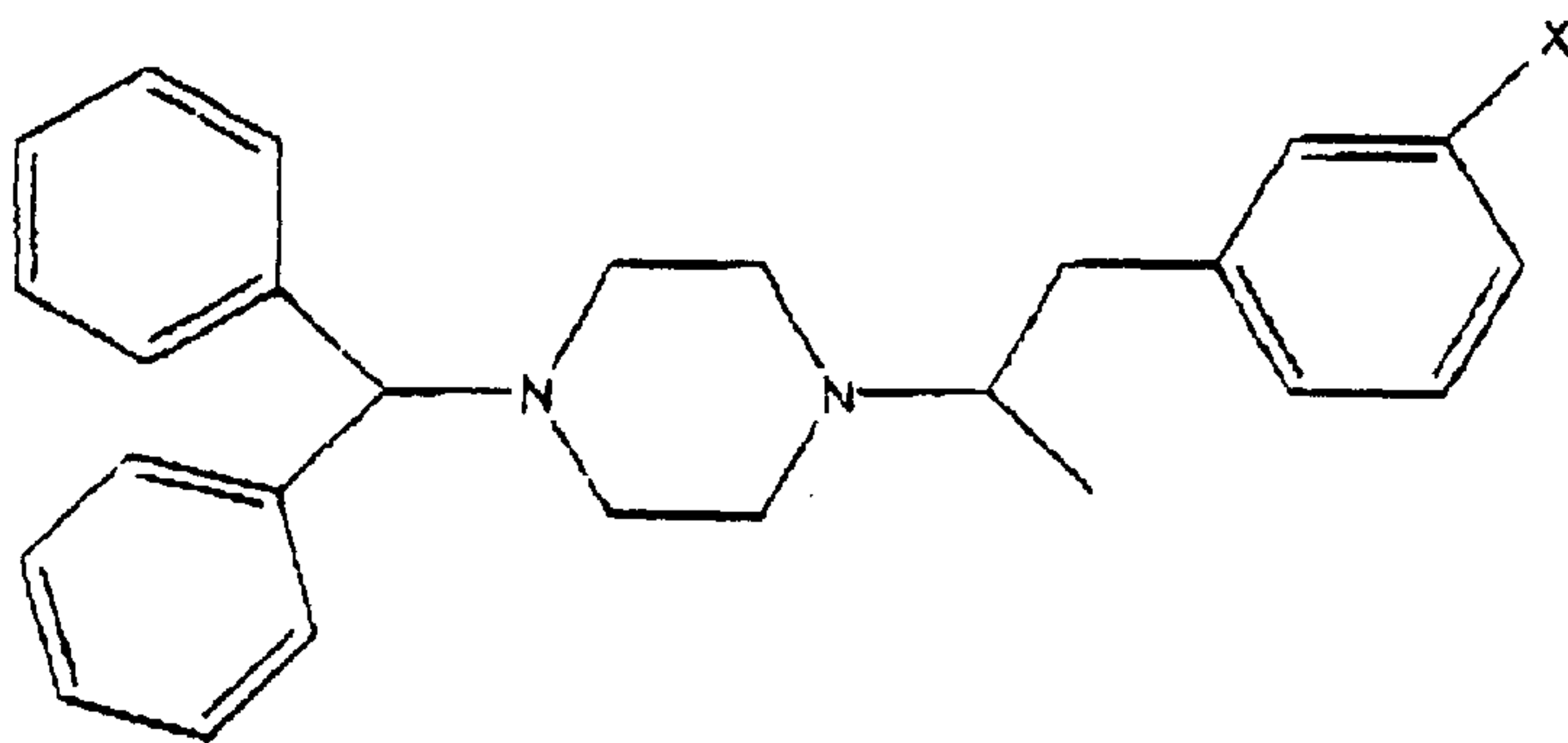
wherein:

R_1 and R_2 each are one or more independent substituents selected from the group consisting of hydrogen, C1-C5 alkyl, C2-C5 alkenyl, C3-C5 cycloalkyl, C1-C5 alkoxy, C2-C5 alkynyl, halogen, C1-C5 haloalkyl, alkylamino, phenyl, nitro, and carboxyl;

R_{15} and R_{16} each are independent and selected from hydrogen and C1-C5 alkyl; and

n is an integer of from 1 to 5 inclusive; or
a pharmaceutically acceptable salt of such compound.

28. The compound as claimed in claim 21, wherein the compound is represented by the following general formula:



wherein X is selected from the group consisting of hydrogen, chlorine, CF_3 , and C1-C5 alkyl.

1/13

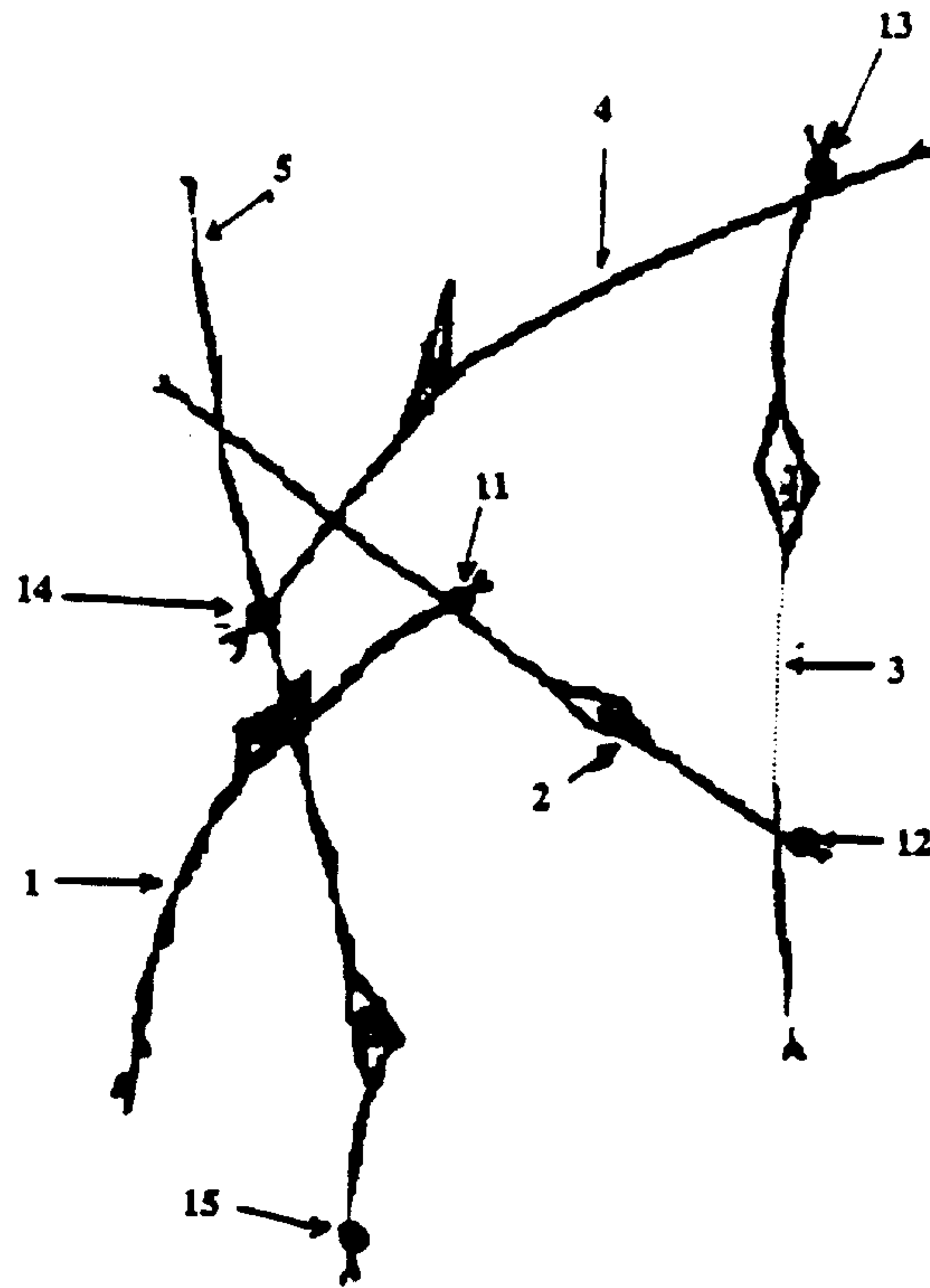


FIGURE 1

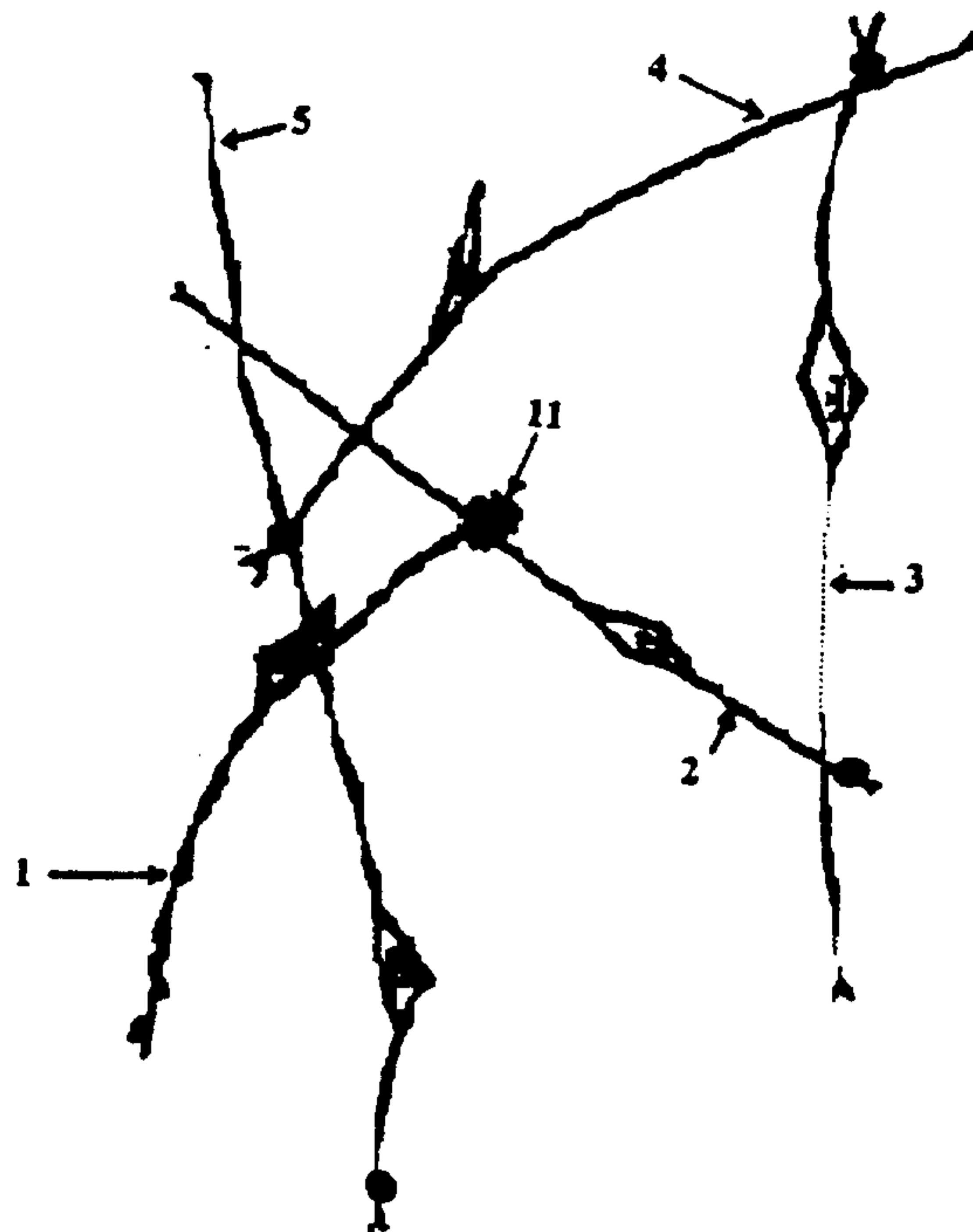


FIGURE 2

WO 98/52898

PCT/US98/10218

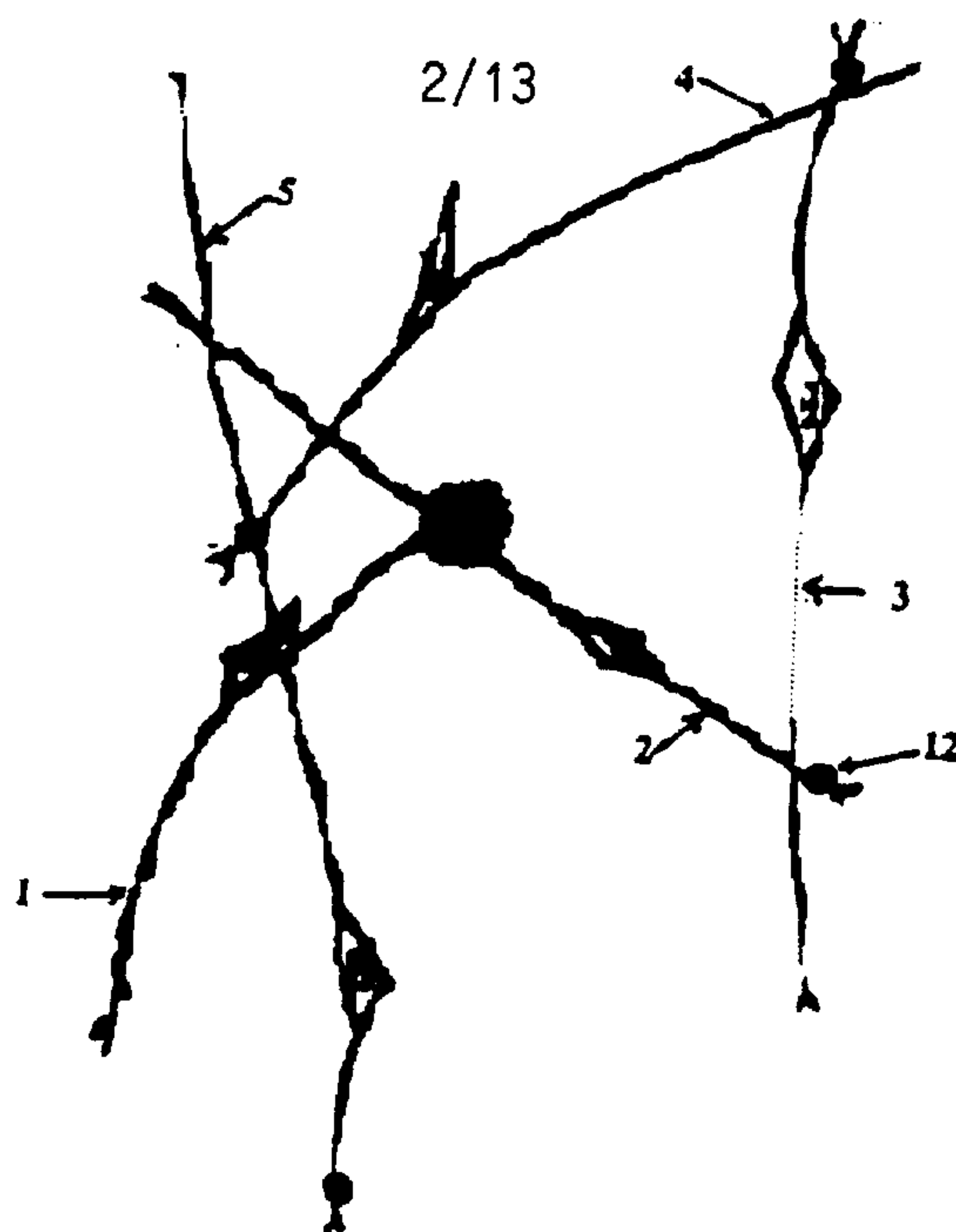


FIGURE 3

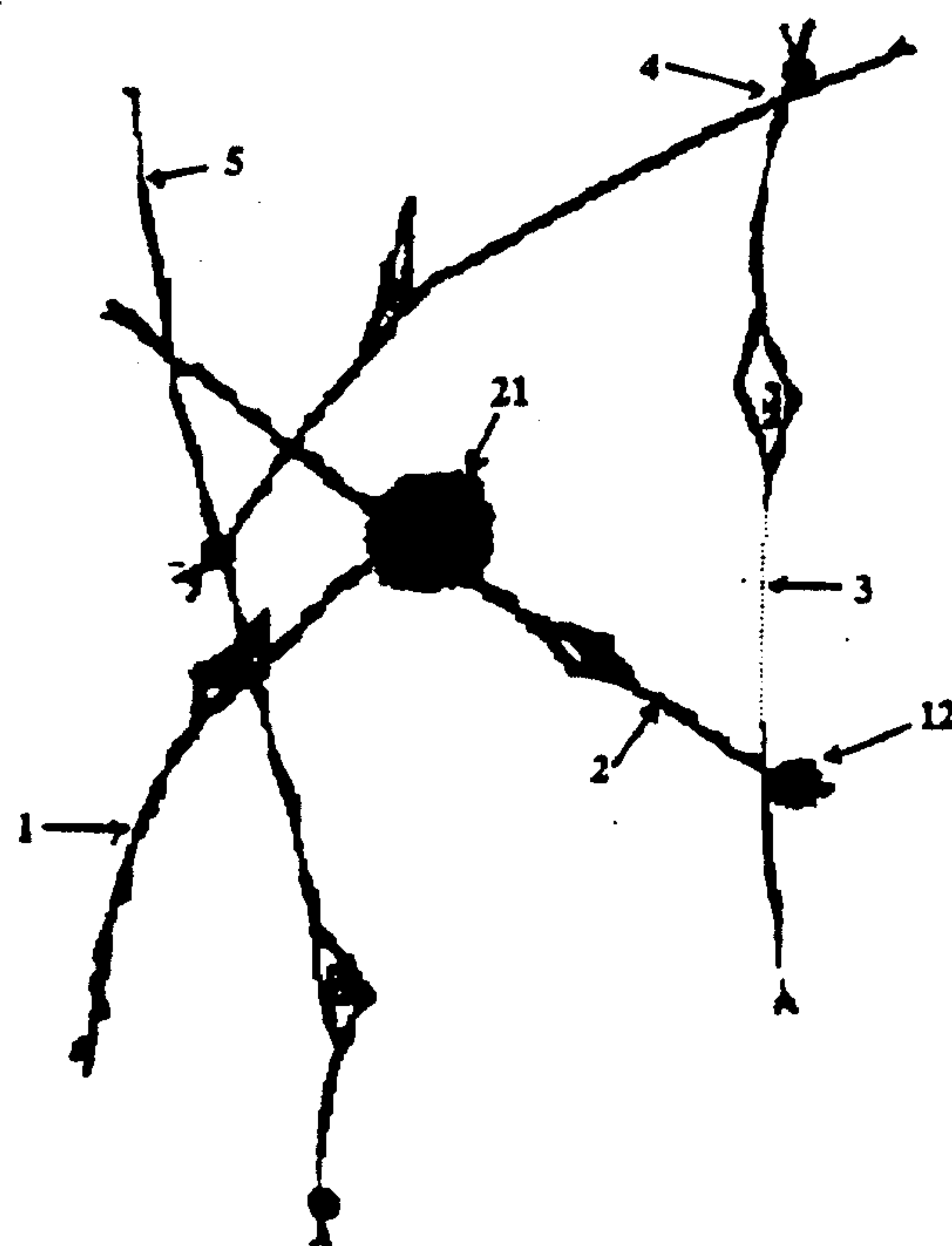


FIGURE 4

WO 98/52898

PCT/US98/10218

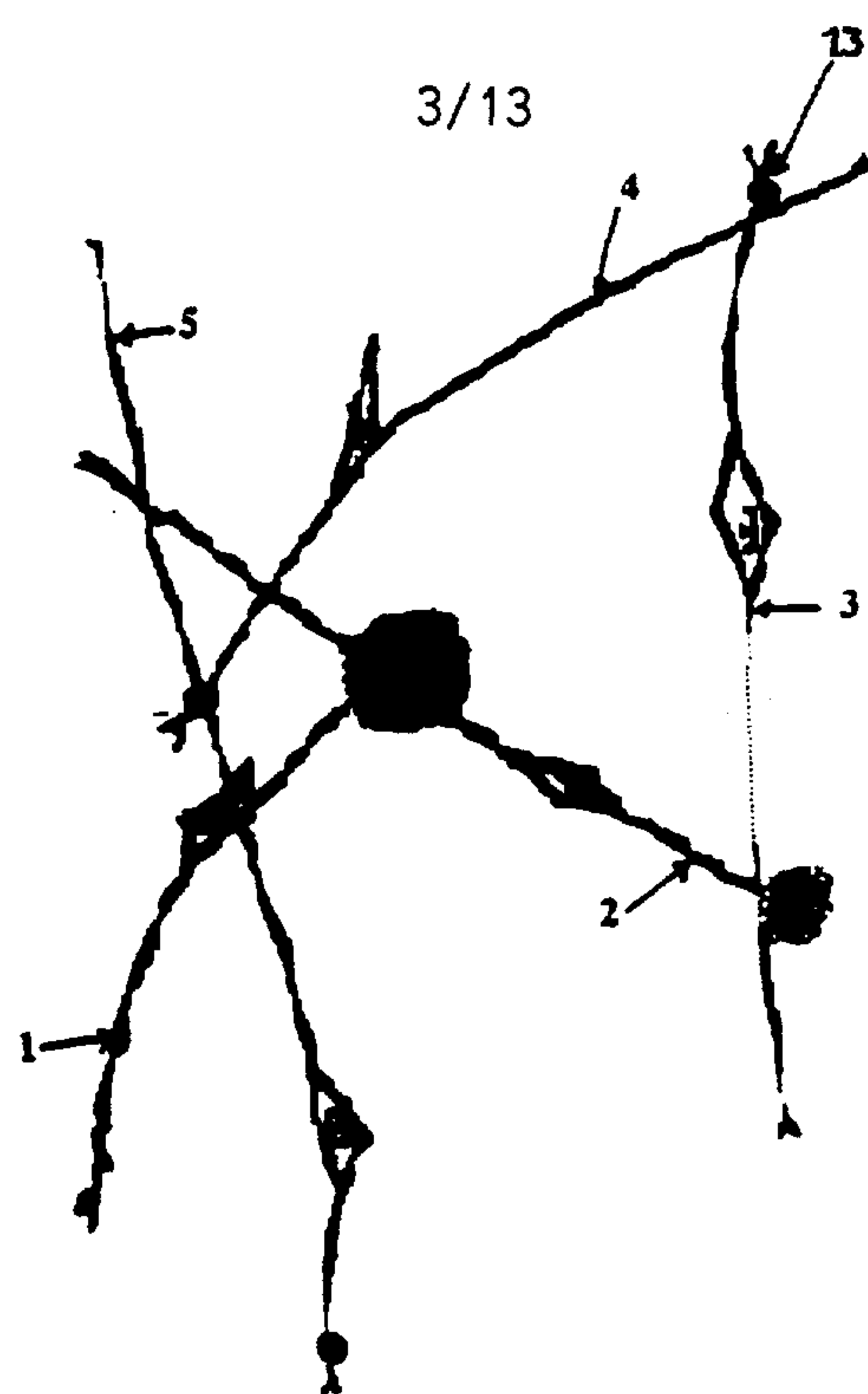


FIGURE 5

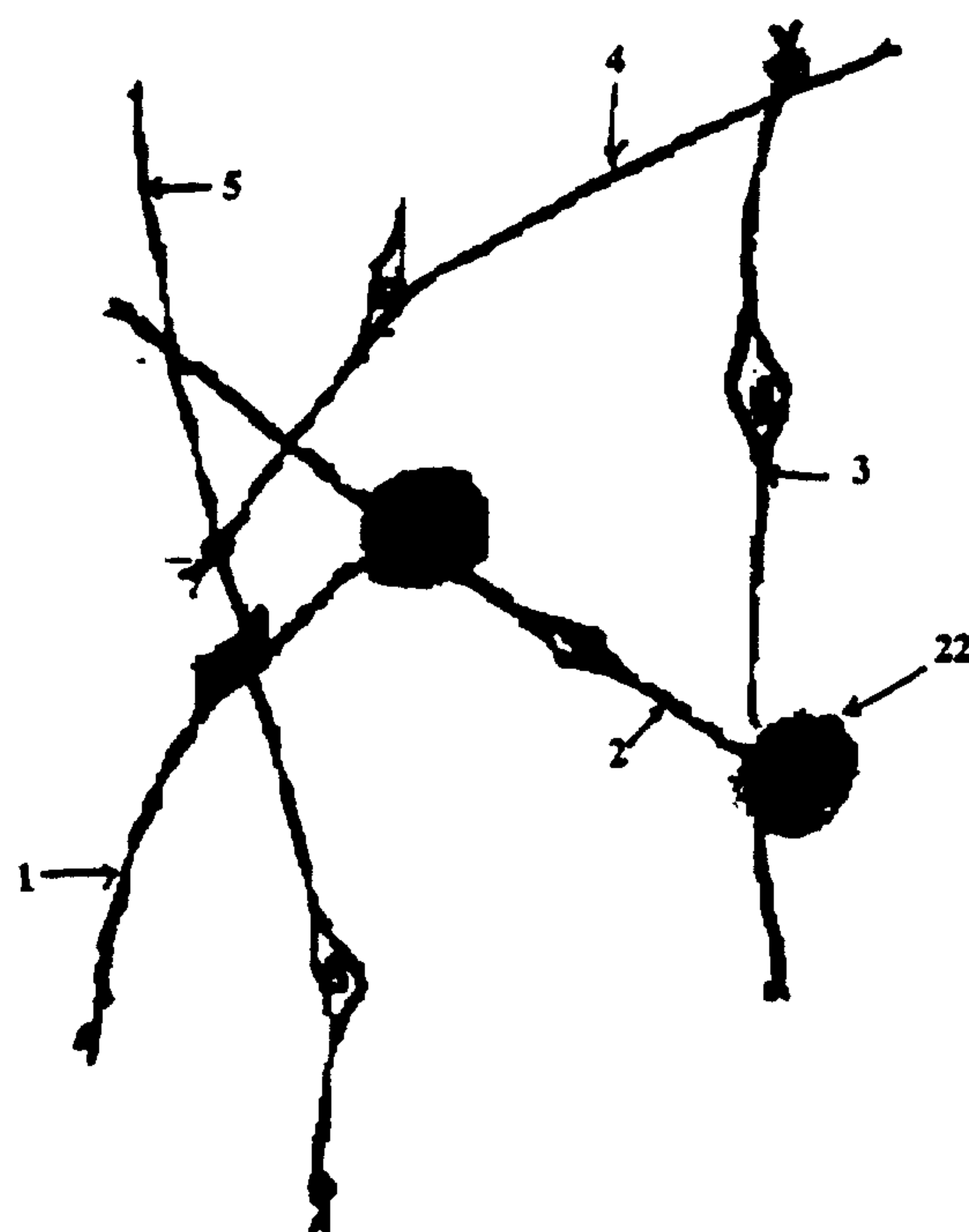


FIGURE 6

WO 98/52898

PCT/US98/10218

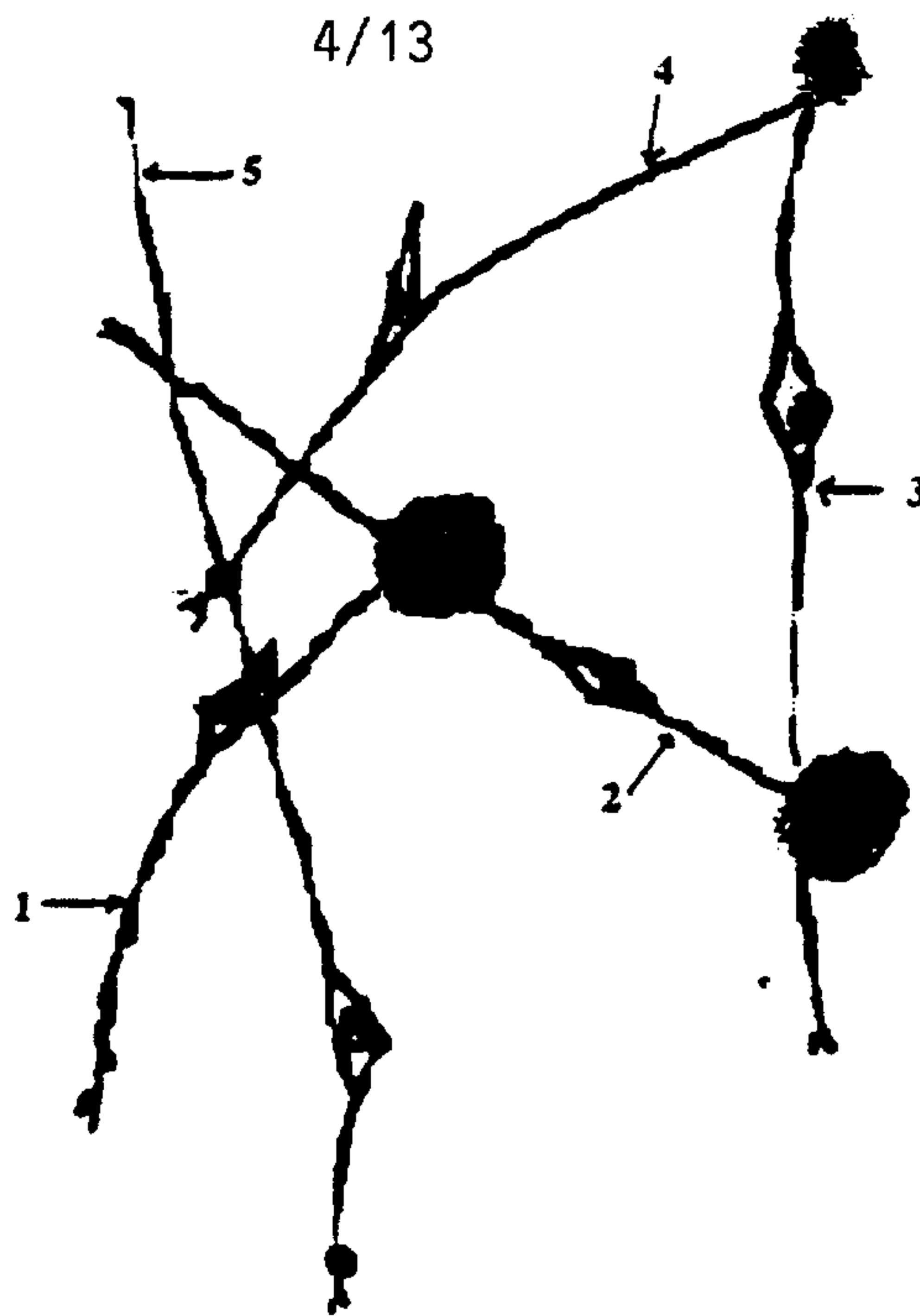


FIGURE 7

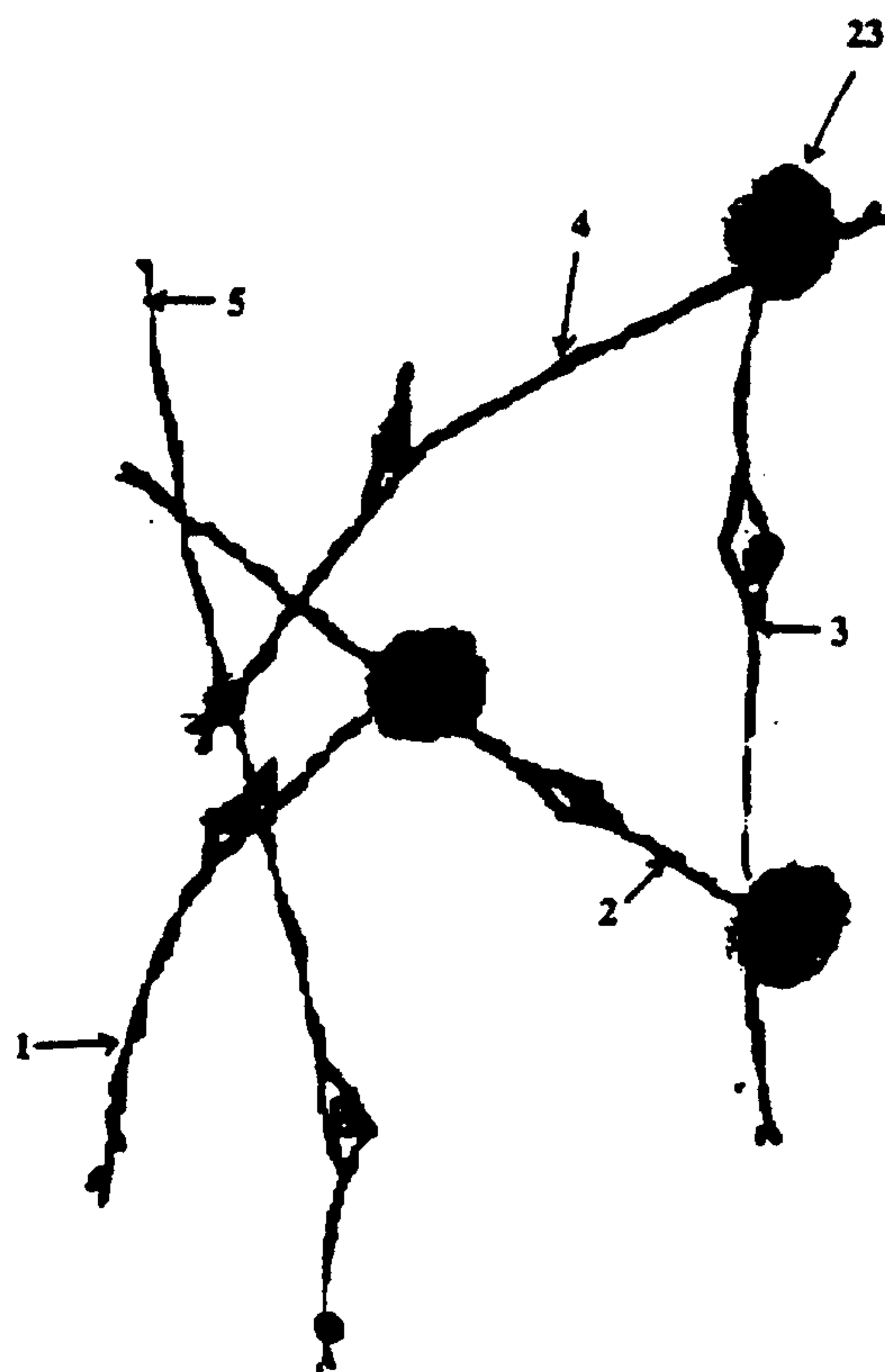


FIGURE 8

WO 98/52898

PCT/US98/10218

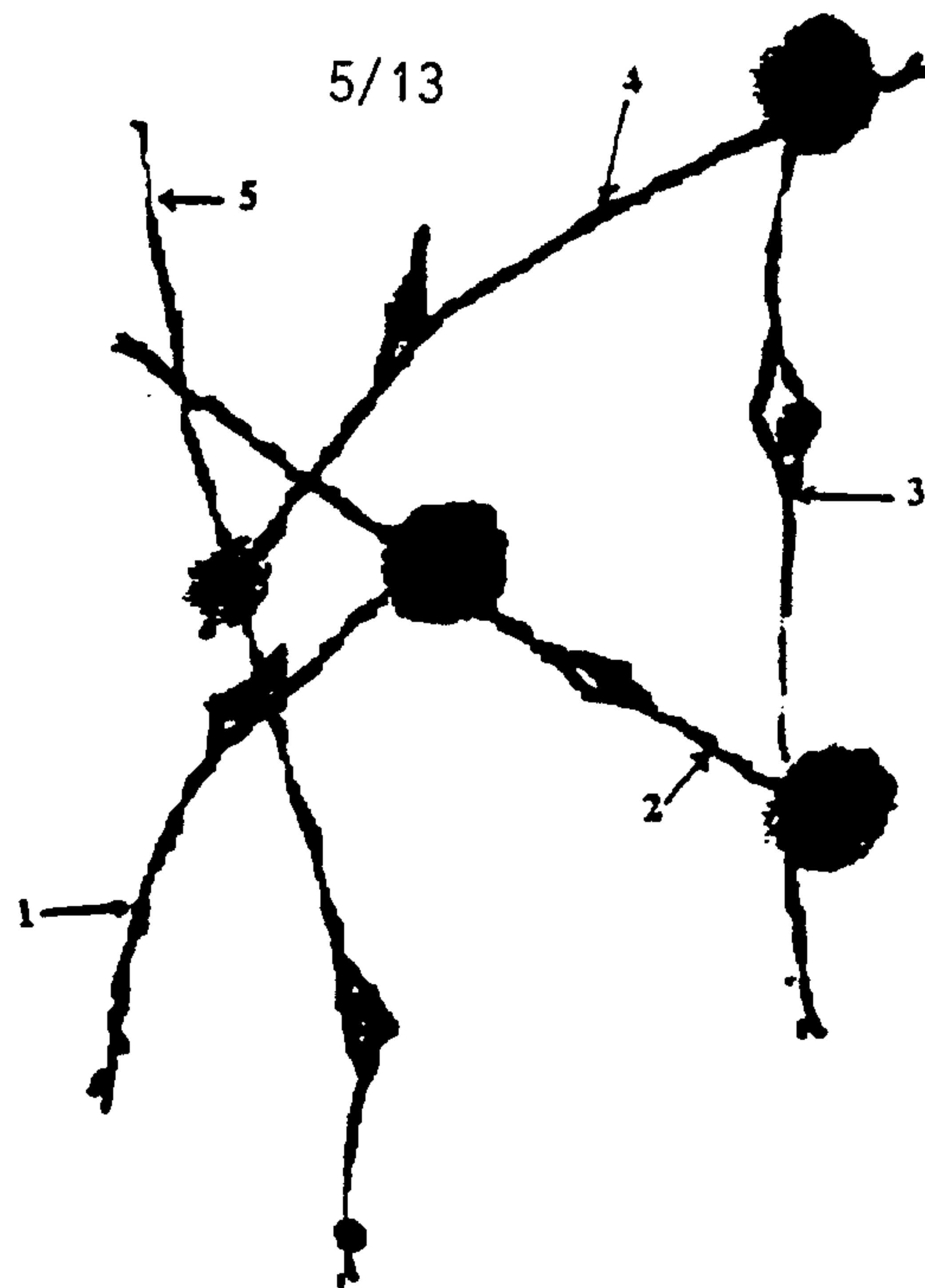


FIGURE 9

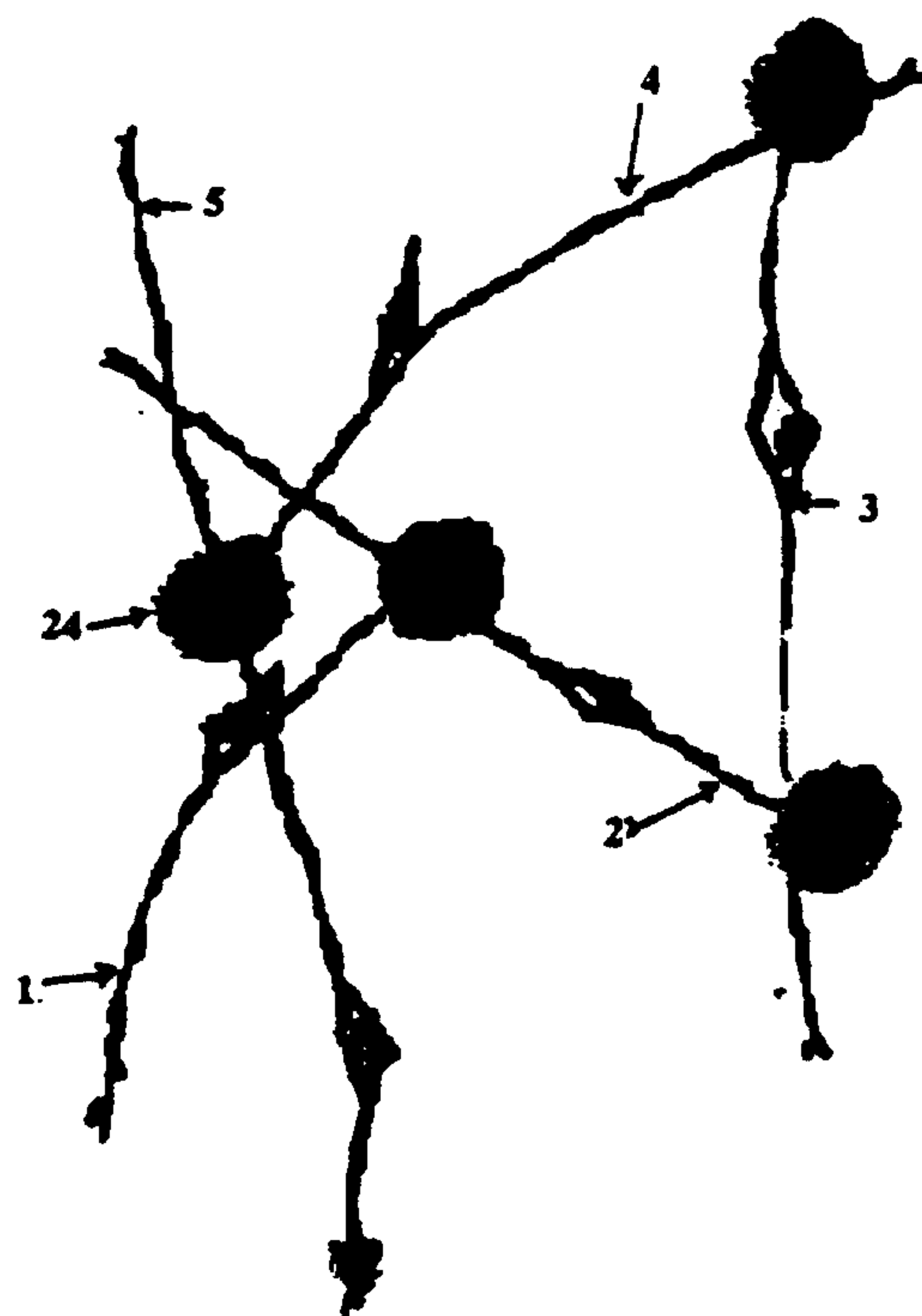


FIGURE 10

WO 98/52898

PCT/US98/10218

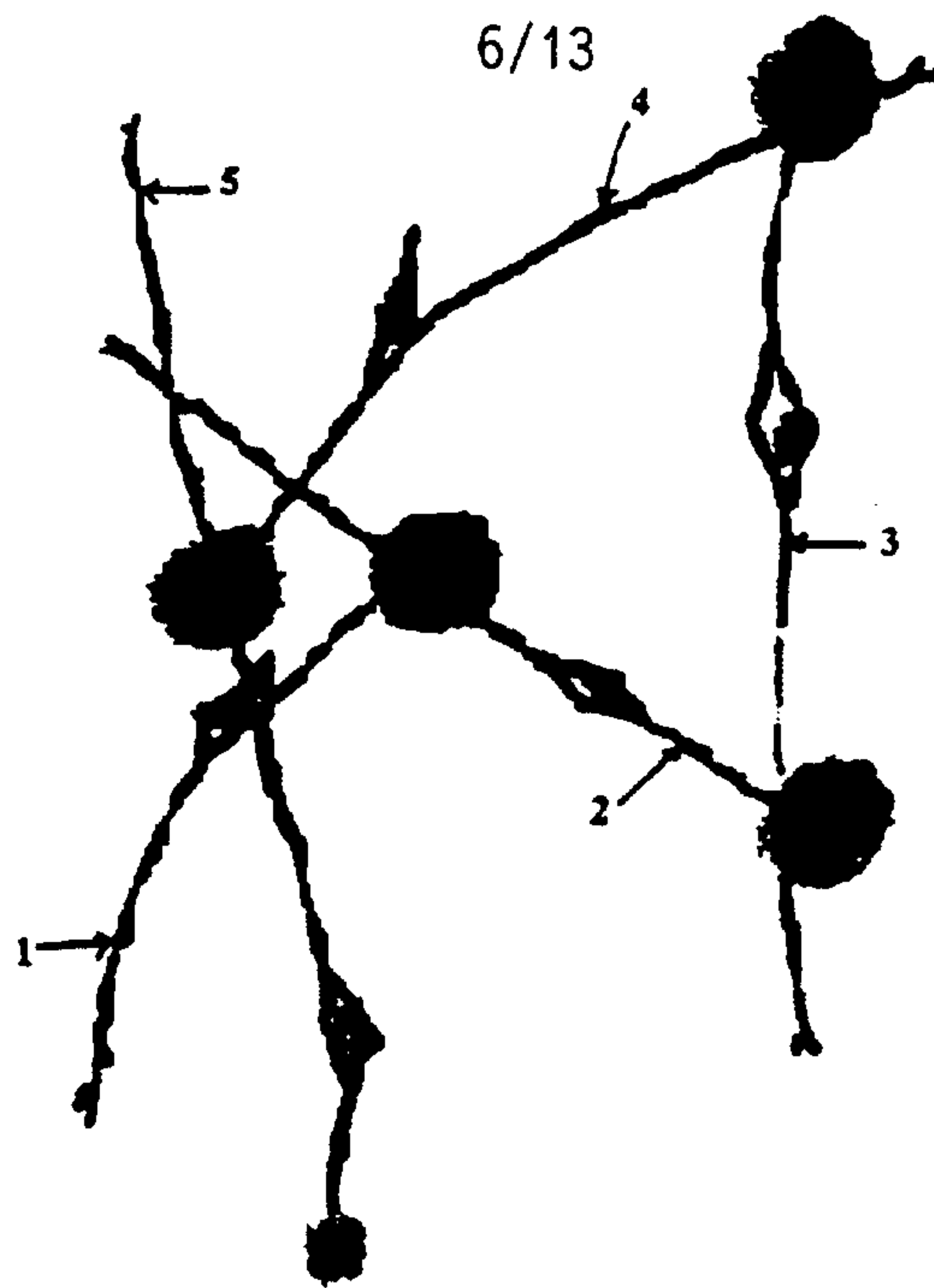


FIGURE 11

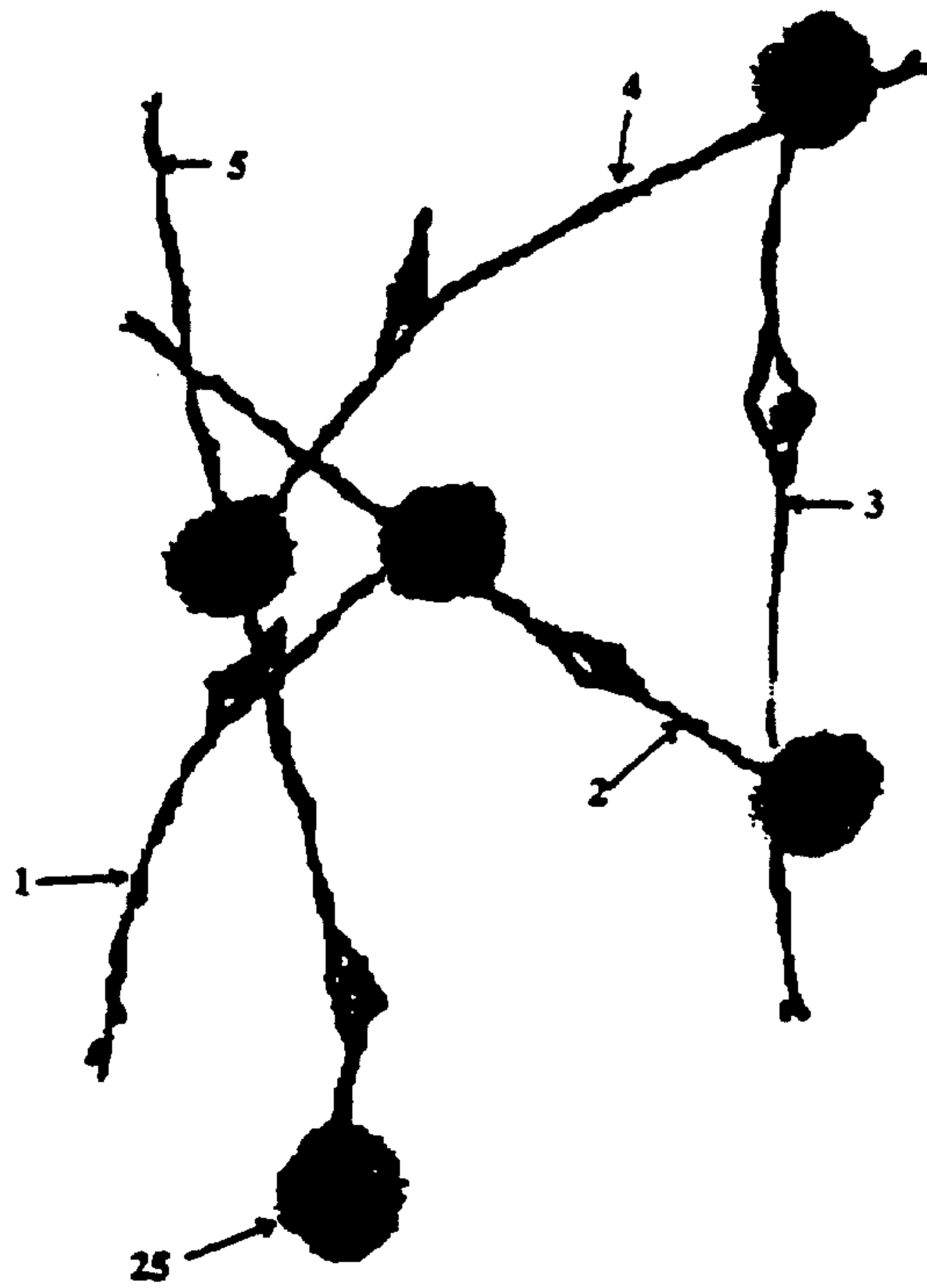


FIGURE 12

7/13

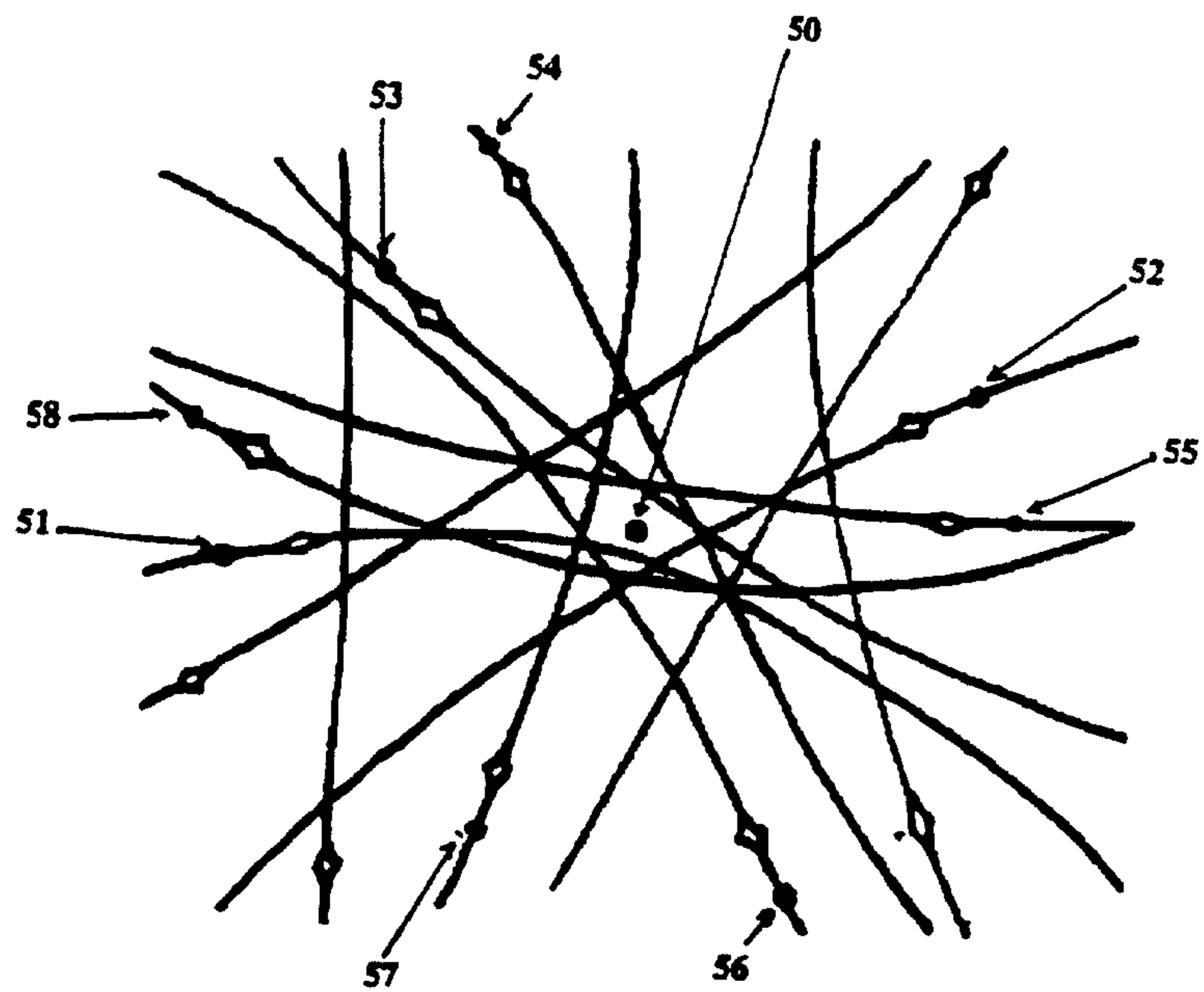


FIGURE 13

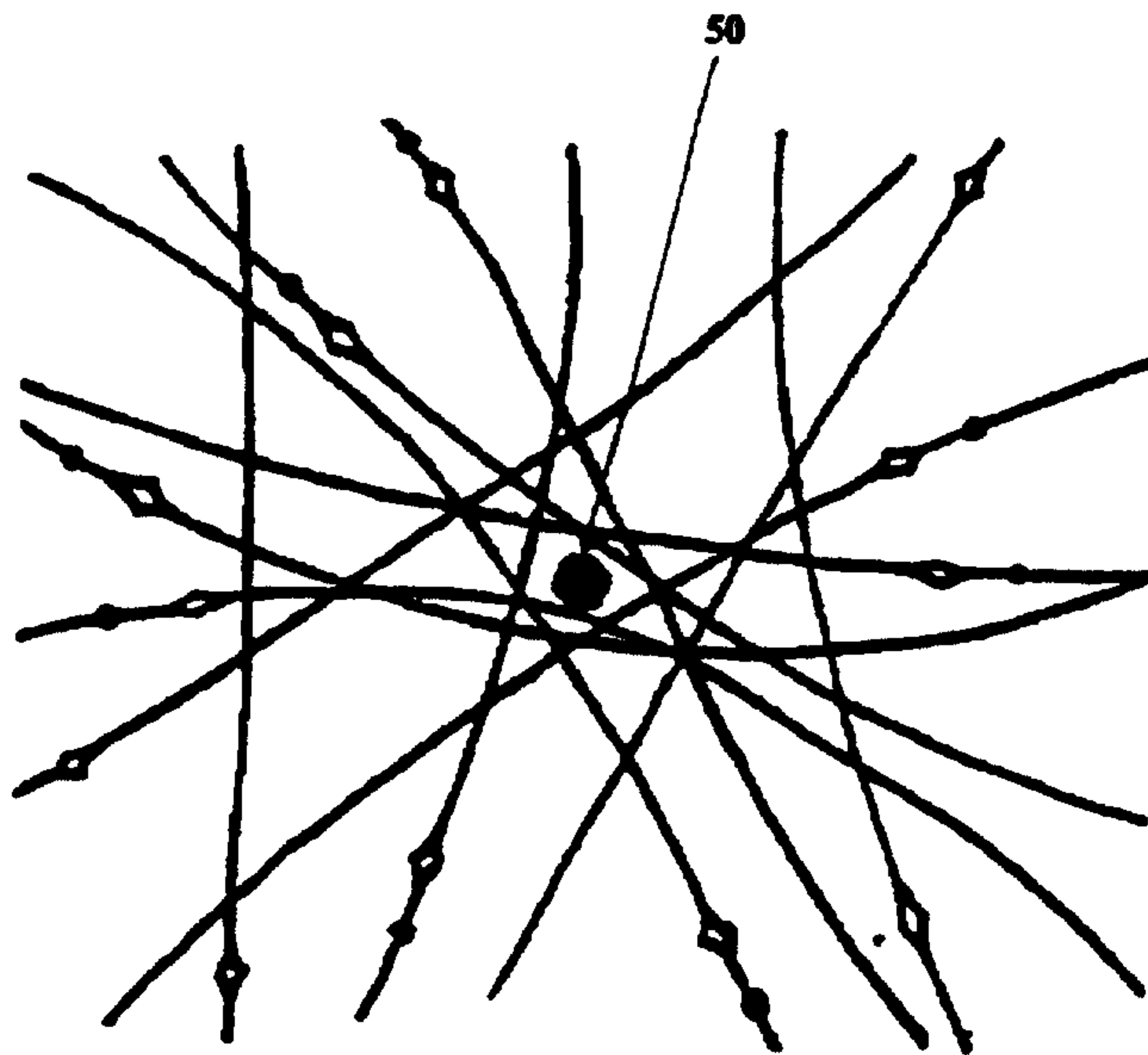


FIGURE 14

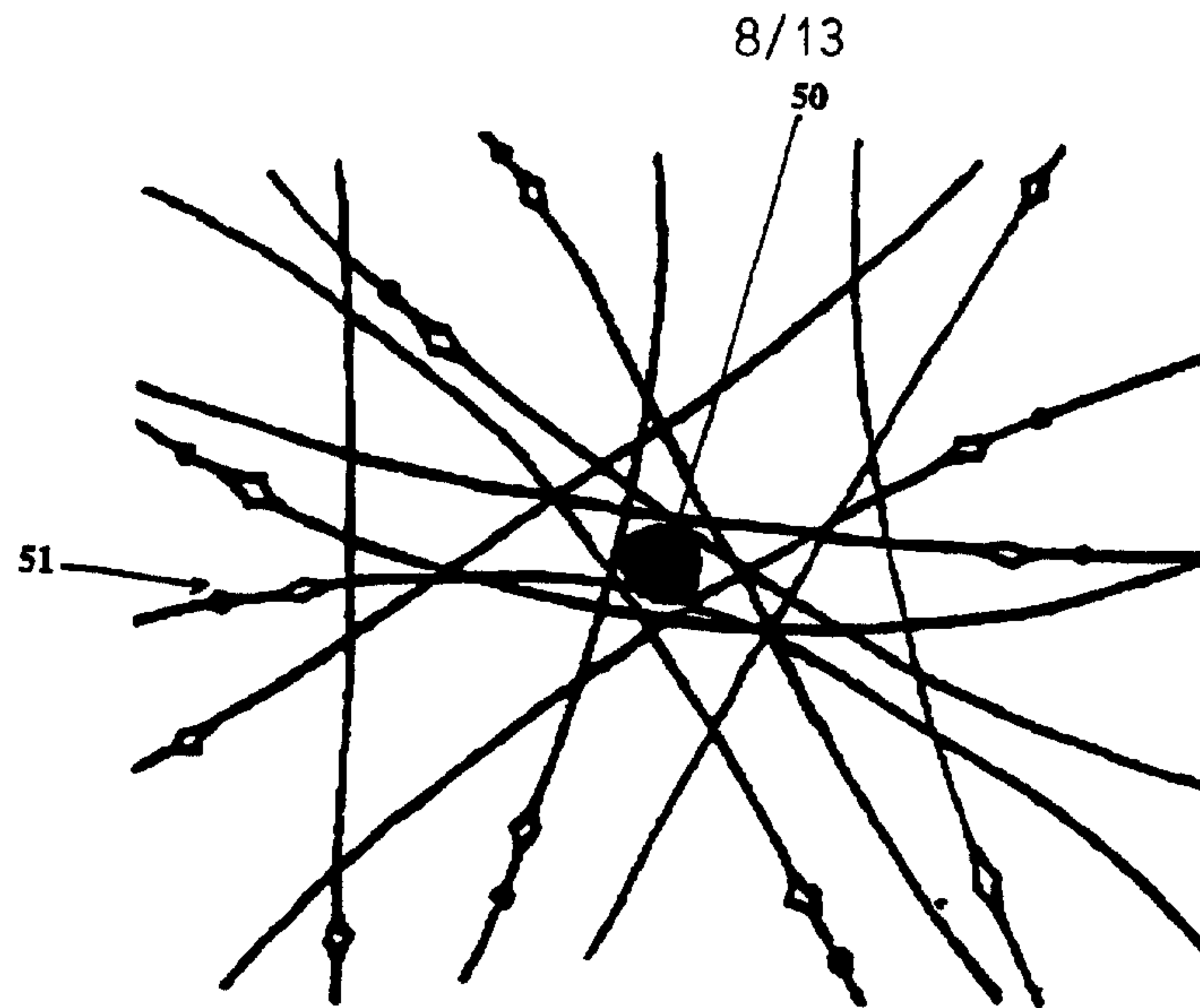


FIGURE 15

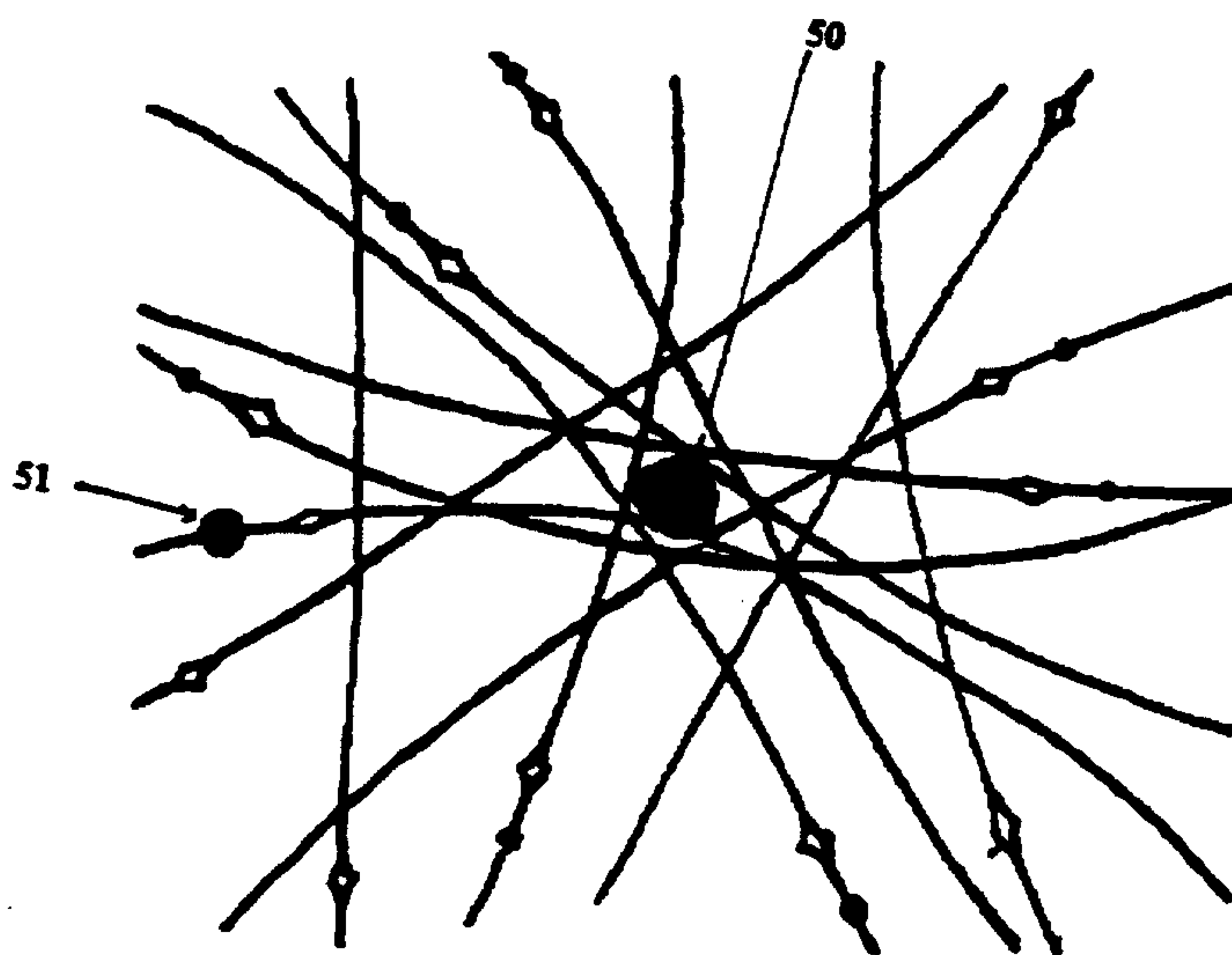


FIGURE 16

9/13

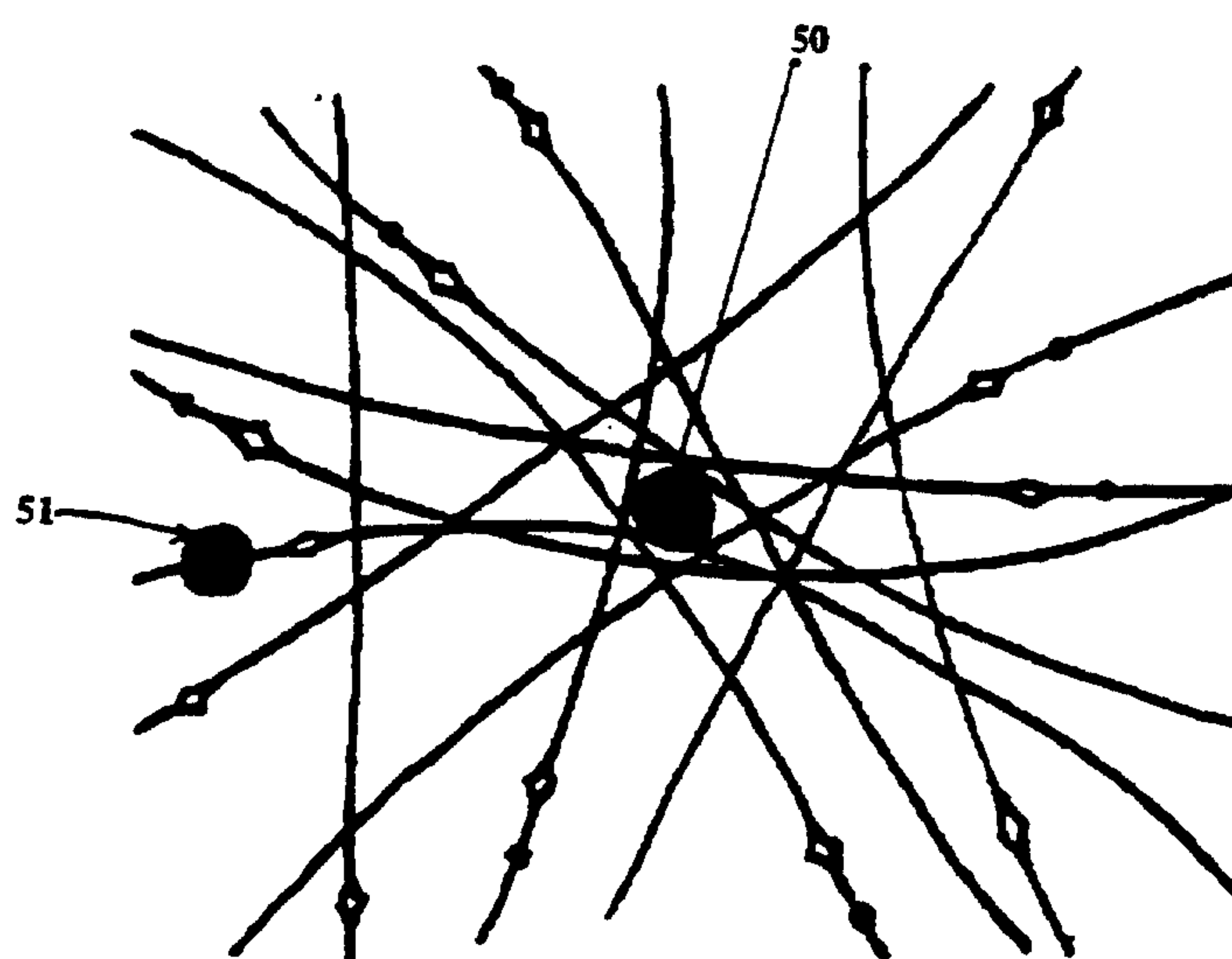


FIGURE 17

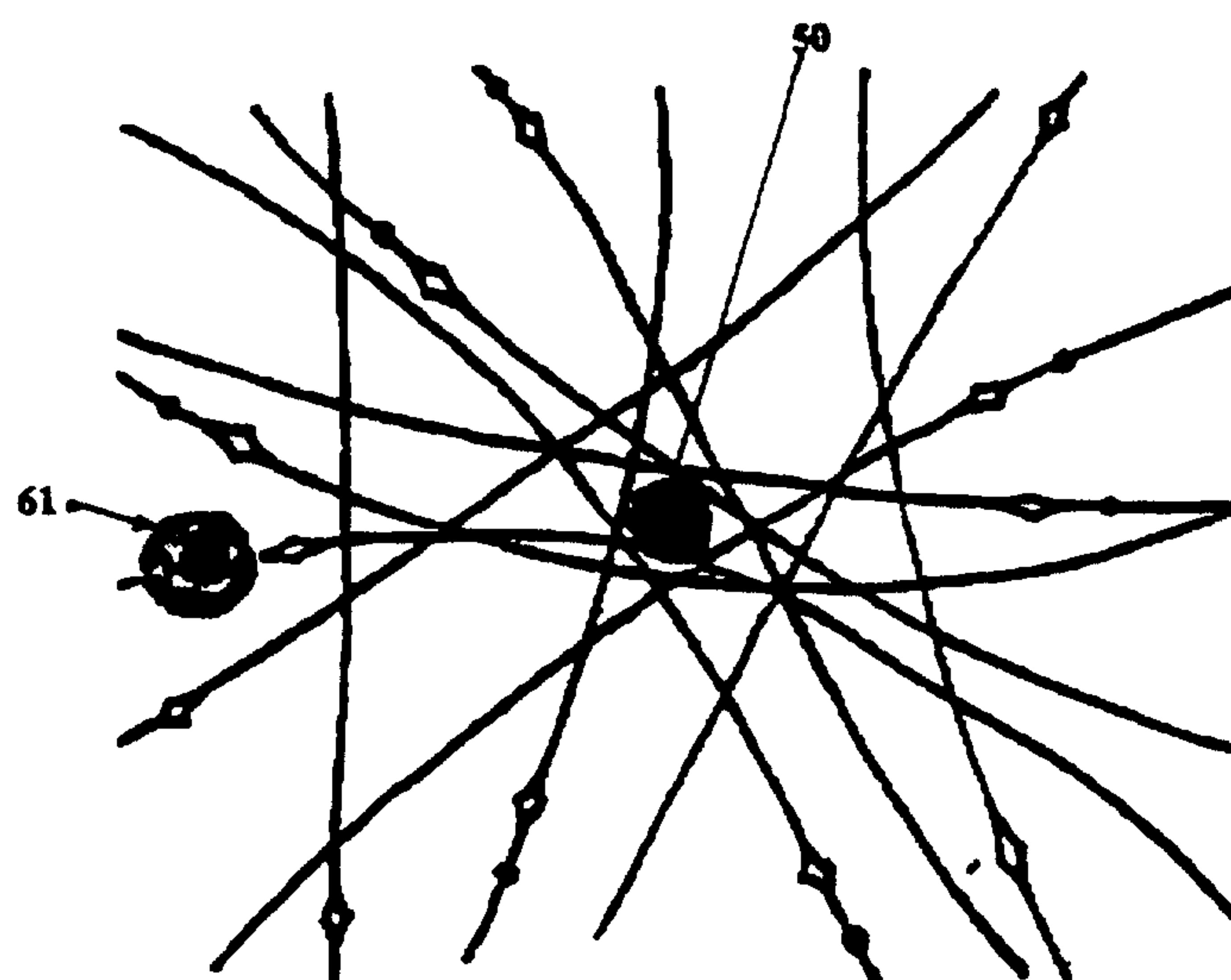


FIGURE 18

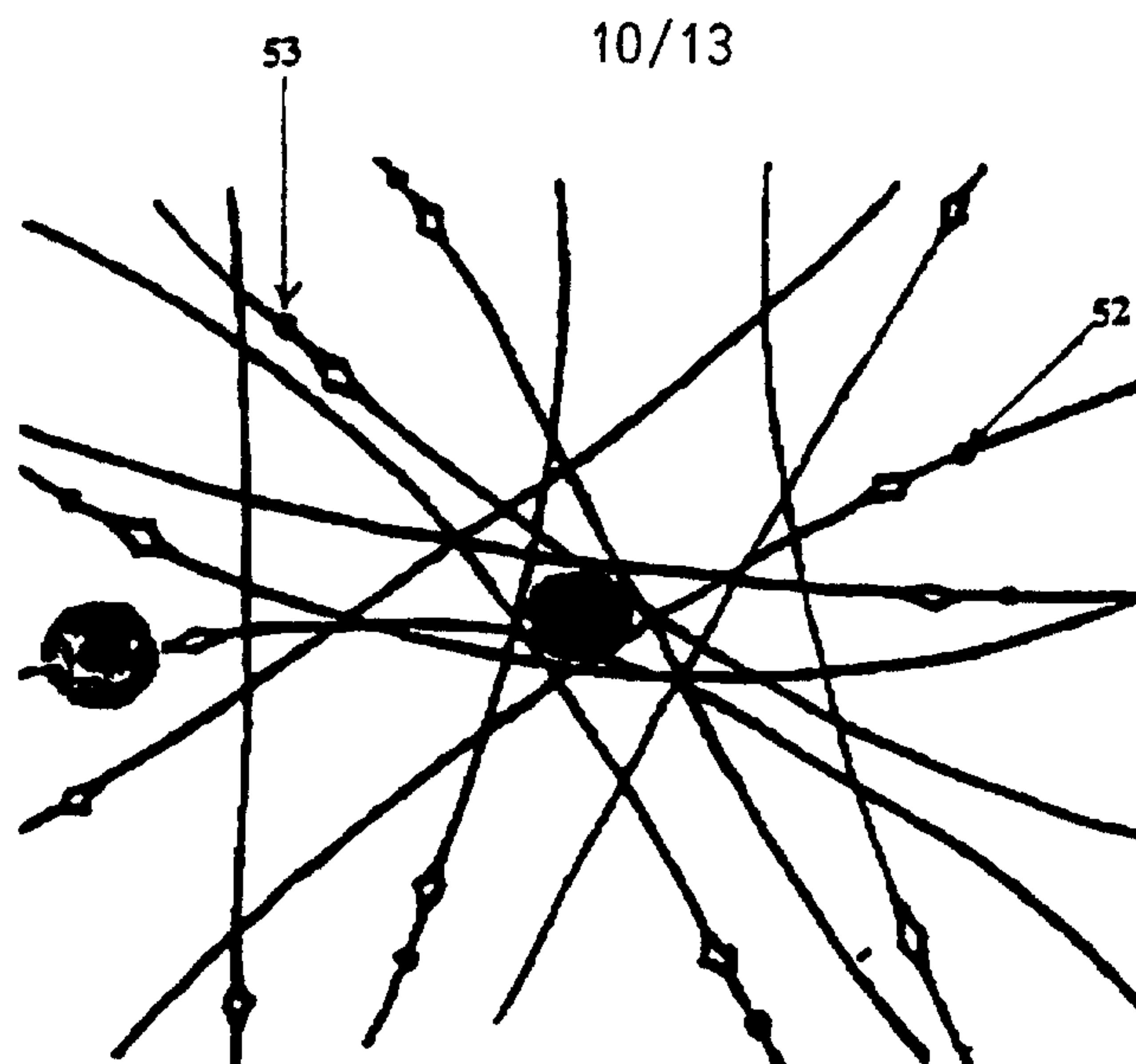


FIGURE 19

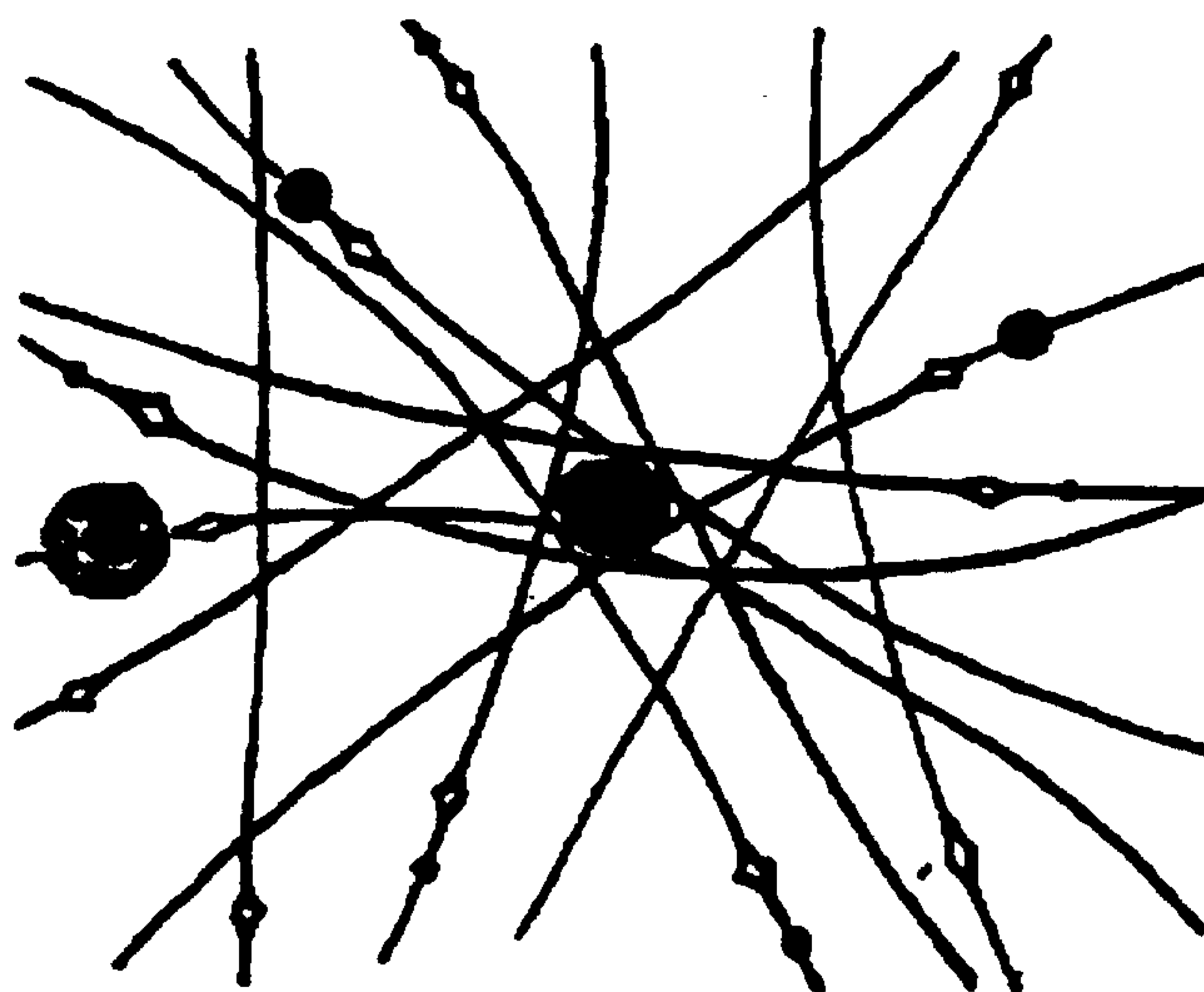


FIGURE 20

11/13

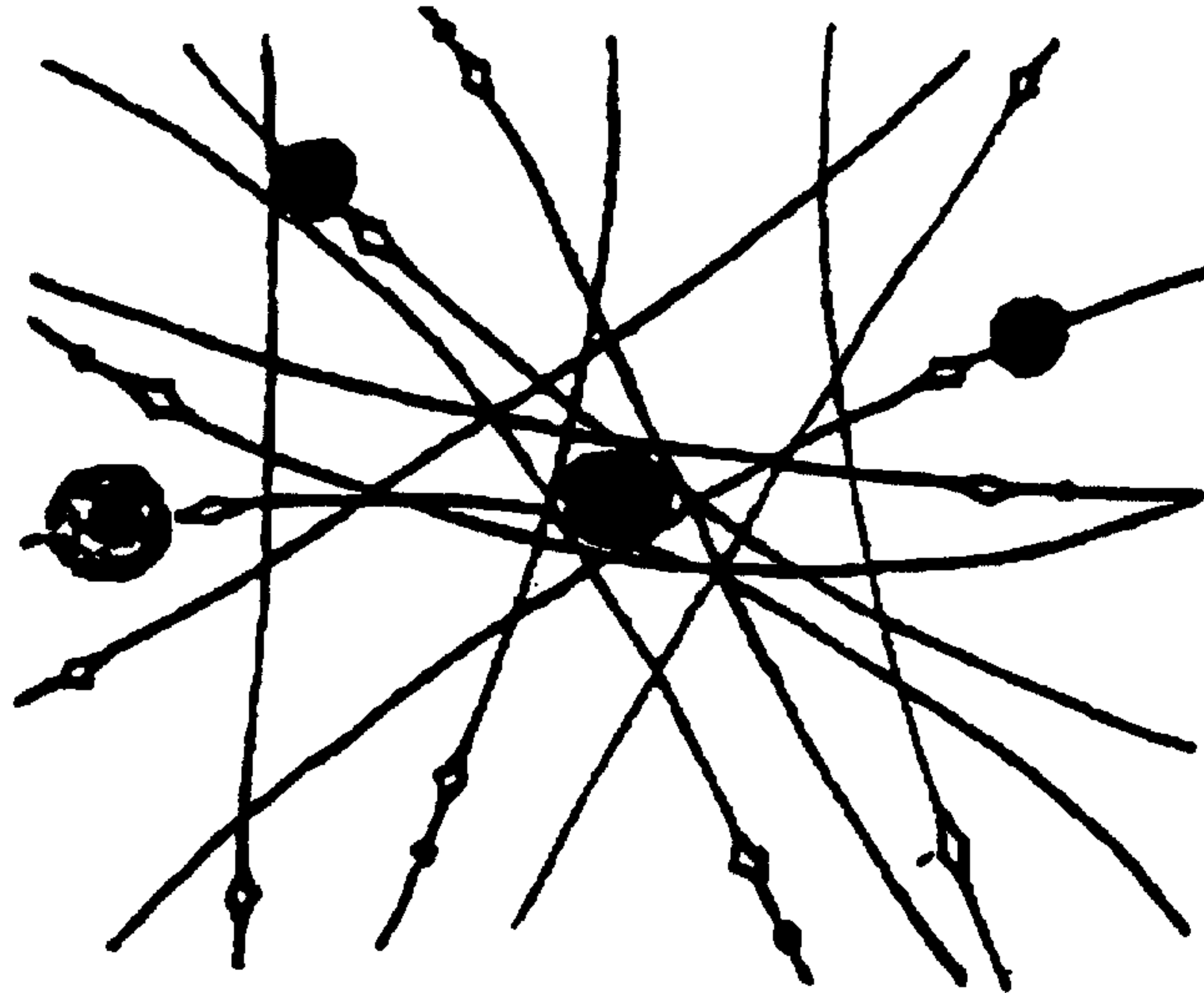


FIGURE 21

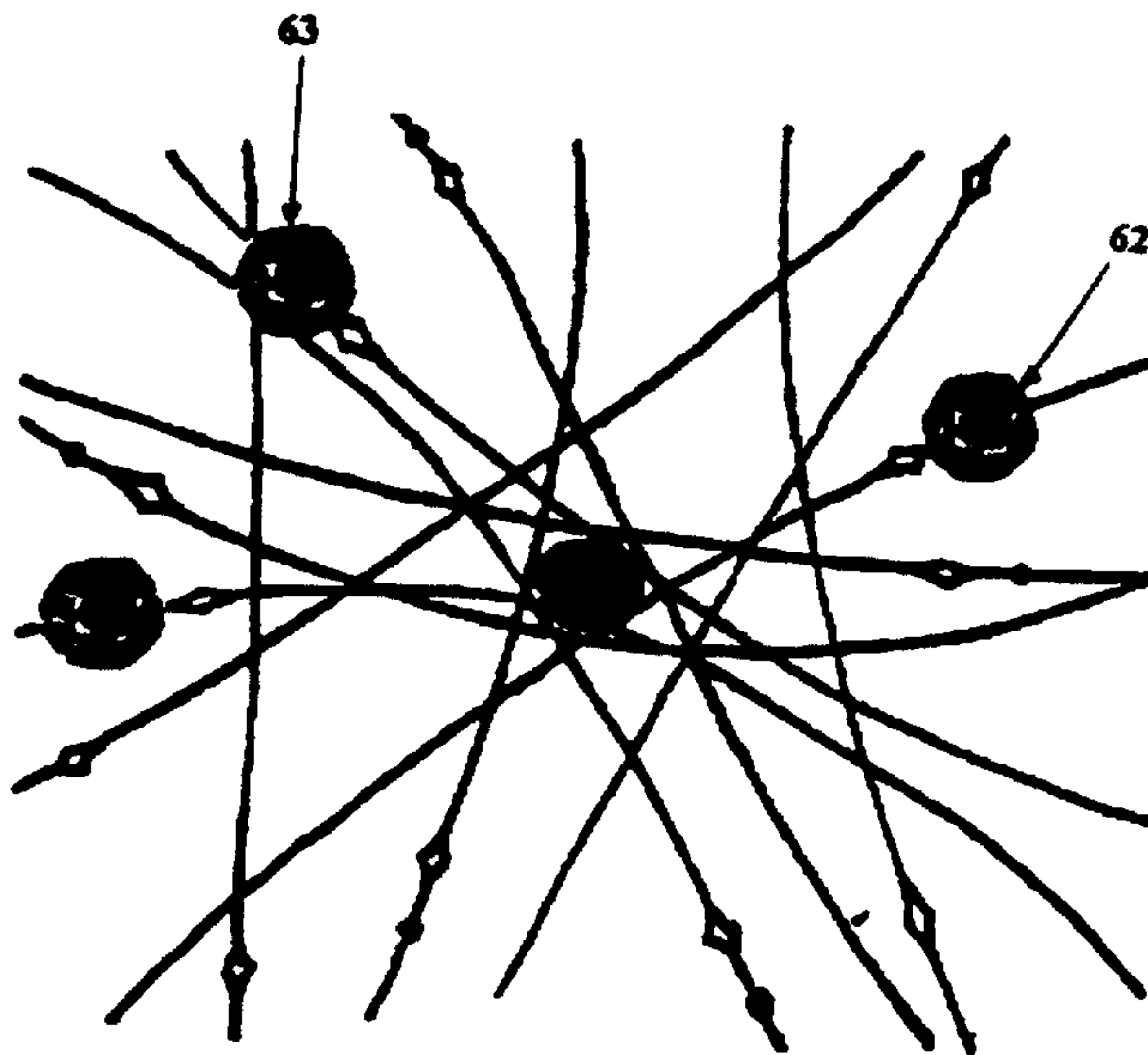


FIGURE 22

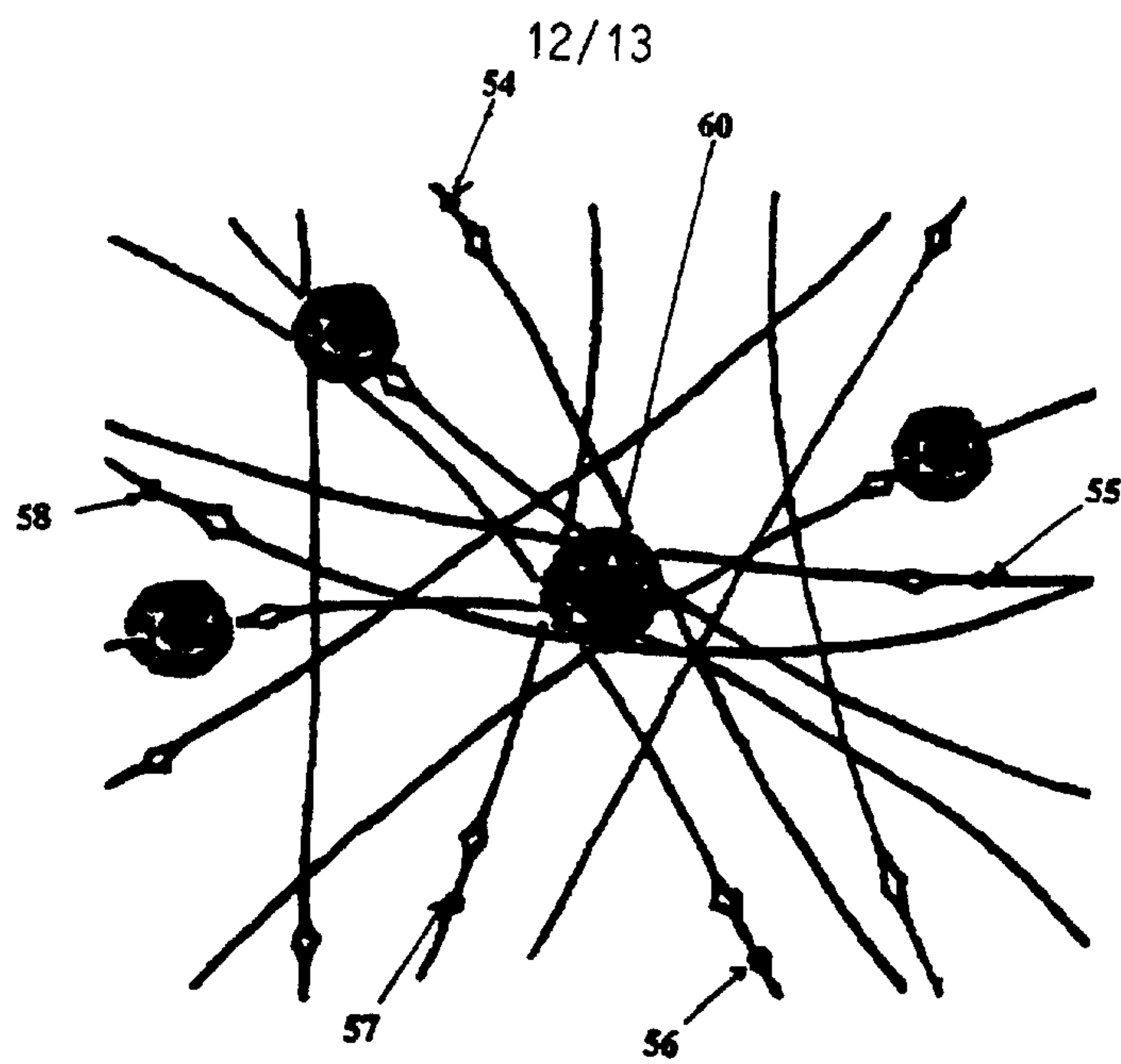


FIGURE 23

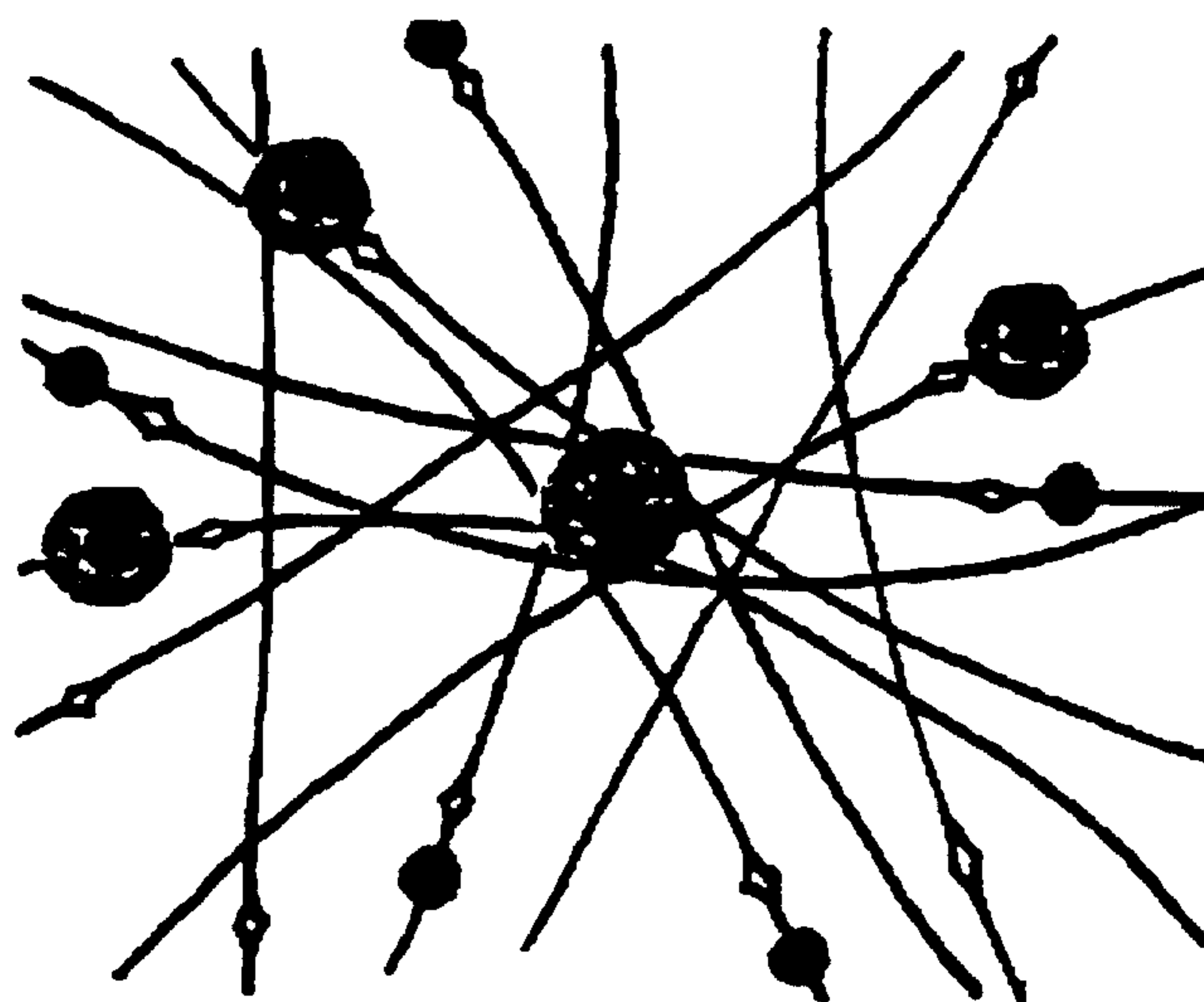


FIGURE 24

13/13

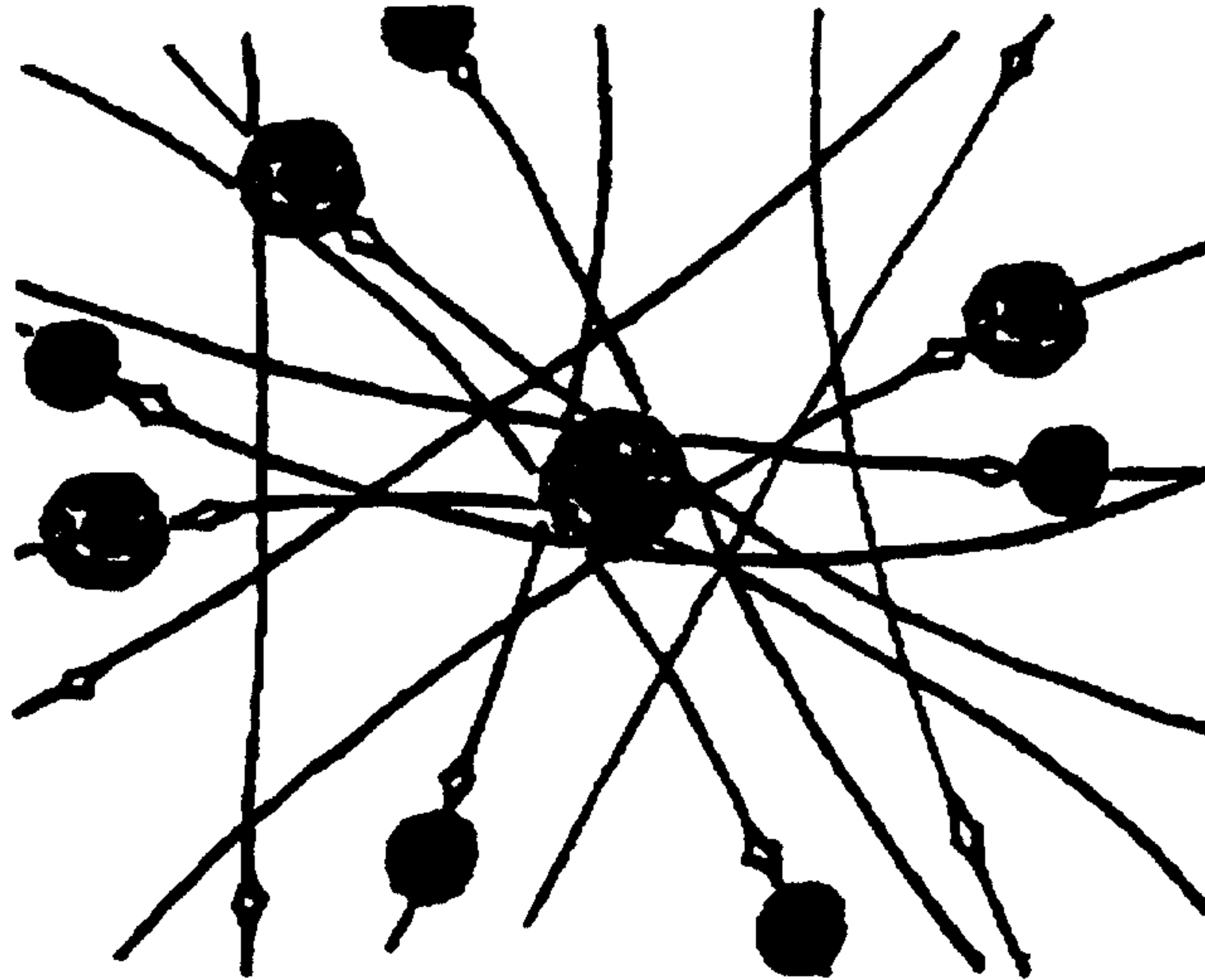


FIGURE 25

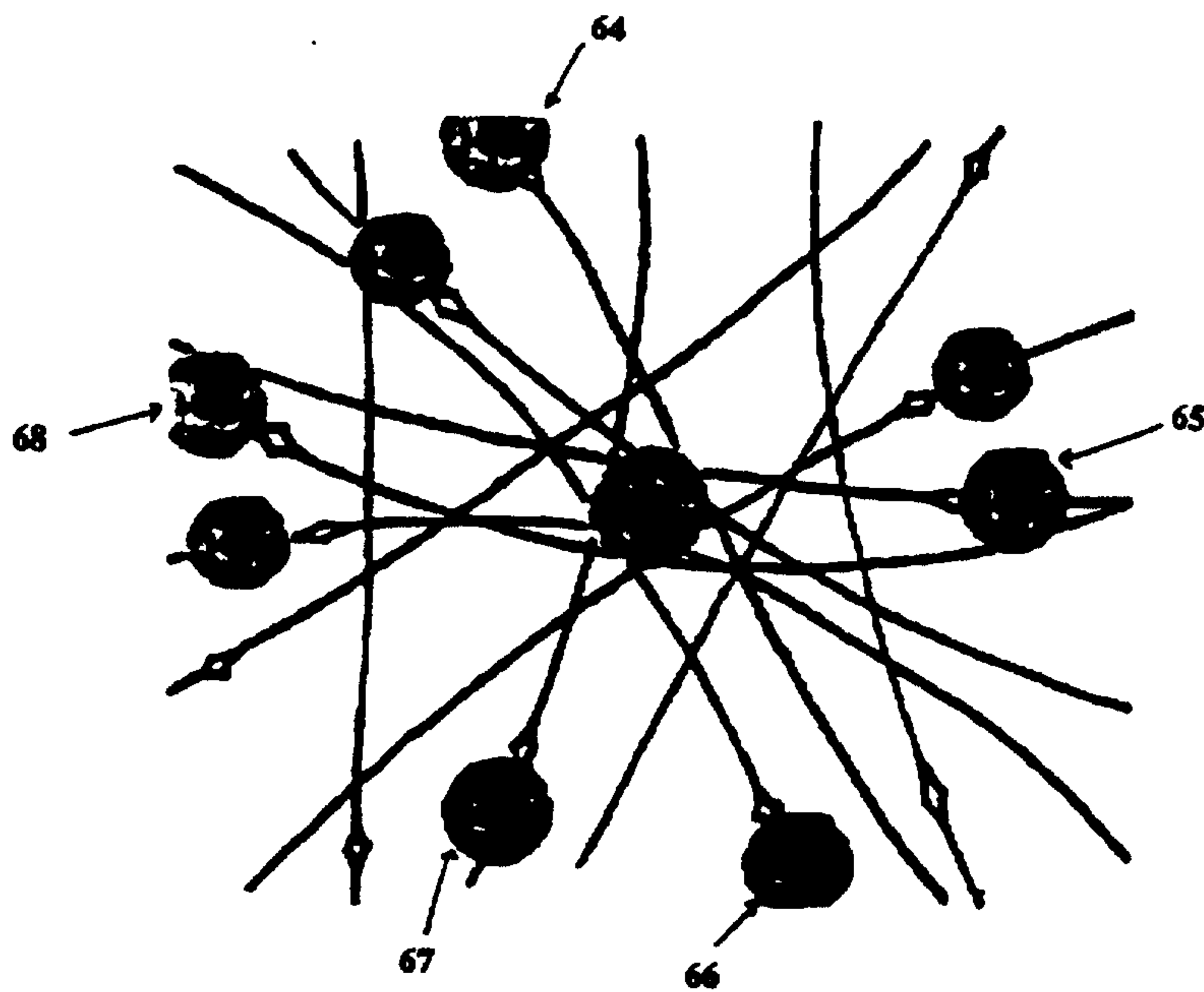


FIGURE 26