Title: TREATMENT OF DYSKINESIA

A Spontaneous activity

Control

Evoked activity

+ D5 stimulation

B

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.


(21) International Application Number: PCT/GB01/03265

(22) International Filing Date: 20 July 2001 (20.07.2001)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data: 0017952.3 22 July 2000 (22.07.2000) GB

(74) Agent: BANFORD, Paul, Clifford; Marks & Clerk, 43 Park Place, Leeds LS1 2RY (GB).


Published: without international search report and to be republished upon receipt of that report.

(54) Title: TREATMENT OF DYSKINESIA

Abstract: The present invention relates to compounds, which enhance D5-dopamine receptor activity, or activation (e.g. selective D5-dopamine receptor agonists), that may be used in the treatment of dyskinesia.
TREATMENT OF DYSKINESIA

The present invention relates to the treatment of dyskinesia.

Dyskinesias are characterised by the development in a subject of abnormal involuntary movements and may manifest as chorea (irregular, involuntary movements of the body, especially the face and extremities) or dystonia (disorder of muscle tonicity and/or posture).

One way in which dyskinesias may arise is as a side effect of dopamine replacement therapy for parkinsonism or other basal ganglia-related movement disorders. Parkinsonism is a syndrome of symptoms characterised by slowness of movement (bradykinesia), rigidity and/or tremor. Parkinsonian symptoms are seen in a variety of conditions, most commonly in idiopathic parkinsonism (i.e. Parkinson’s disease) but also following treatment of schizophrenia, exposure to toxins/drugs and, head injury.

The use of dopamine-replacing agents (e.g. L-DOPA and apomorphine) as symptomatic treatments for conditions such as Parkinson’s disease have undoubtedly been successful in increasing the quality of life of patients suffering from the conditions. However, dopamine-replacement therapy does have limitations, especially following long-term treatment. Problems can include a wearing-off of the anti-parkinsonian efficacy of the treatment and in particular the appearance of a range of side-effects. These side-effects may manifest as dyskinesias such as chorea and dystonia. Dyskinesia can be seen either when the patient is undergoing dopamine-replacement therapy (in the case of chorea and/or dystonia) or even when off therapy (when dystonia is prevalent). Ultimately, these side-effects severely limit the usefulness of dopaminergic treatments.

Many attempts have been made to develop agents that will prevent the development of, and/or treat, dyskinesias although such attempts have met with limited success. There is, therefore, a need to develop ways by which dyskinesias may be treated.
According to a first aspect of the present invention, there is provided a use of a compound which enhances D₂-dopamine receptor activity, or activation, for the manufacture of a medicament for the treatment of dyskinesia.

According to a second aspect of the present invention, there is provided a composition for use in the treatment of dyskinesia comprising a therapeutically effective amount of a compound which enhances D₂-dopamine receptor activity, or activation, and a pharmaceutically acceptable vehicle.

According to a third aspect of the present invention, there is provided a method for the treatment of dyskinesia comprising administering to a person or animal in need of such treatment a therapeutically effective amount of a compound which enhances D₂-dopamine receptor activity, or activation.

D₂-dopamine receptors are a subclass of dopamine receptors which are found in neural tissues.

By “dyskinesia” we mean the development in a subject of abnormal involuntary movements. These movements may manifest as chorea (irregular, involuntary movements of the body, especially the face and extremities) or dystonia (disorder or lack of muscle tonicuity). Such movements include ballistic movements and athetoid movements of the trunk, limbs and facial musculature.

The invention is based upon our studies relating to the neural mechanisms underlying movement disorders. Although we do not wish to be bound by any hypothesis, we believe that movement disorders involve abnormal activity of basal ganglia output pathways and in many cases this is brought about by abnormal function of subthalamic nucleus efferent pathways. These consist of excitatory pathways to both the medial segment of the globus pallidus and the pars reticulata of the substantia nigra. One of the pathophysiological hallmarks of dyskinesia is underactivity of the subthalamic nucleus output pathways. We believe compounds,
which enhance D_5-dopamine receptor activity, or activation, increase the activity of the subthalamic output pathways and thereby reduce dyskinesia.

We have found that compounds which enhance D_5-dopamine receptor activity, or activation, are highly effective for the treatment of dyskinesias. For instance, we have found that dyskinesias (e.g. chorea and dystonia) do not develop, or are at least reduced, when the compounds are given to subjects on dopamine-replacement therapy for the treatment of a movement disorder.

The present invention is considered to relate to a surprising development for several reasons. For instance, a skilled person would not expect a compound that stimulates dopamine receptor activity to have beneficial effects in treating dyskinesias. This is because dyskinesia is associated with dopamine receptor stimulation and it is counterintuitive that further stimulation of dopamine receptors would be of benefit. Dziewczapolski et al. ((1998) NeuroReports 9, 1-5), have reported that D_1-dopamine receptors and D_5-dopamine receptors may have opposite roles in locomotion. However this paper discloses that D_5 stimulation has no significant effect on behaviour and more specifically there was no disclosure relating to the modulation of dyskinesia. Accordingly the present invention may be considered surprising in the light of this prior art.

Several classes of compound, which may be used according to the invention, are capable of enhancing D_5-dopamine receptor activity. These compounds include:

(i) D_5-Dopamine receptor agonists and partial agonists (e.g. 2-amino-6, 7-
dihydroxytetralin);
(ii) compounds which enhance synthesis of endogenous D_5-dopamine receptor agonists by increasing the synthesis of precursors or the conversion of precursors into D_5-dopamine receptor activating ligands;
(iii) compounds which enhance release of D_5-dopamine receptor agonists;
(iv) compounds which block the rate of inactivation or metabolism of D_5-
dopamine receptor agonists (e.g. MAO-B inhibitors, CONT inhibitors); and
(v) compounds which promote/increase D₂-dopamine receptor expression and/or transcription.

The compound may modulate any type of dopamine receptor provided that D₃-dopamine receptor activity is selectively enhanced. By “selectively” we mean the compound enhances D₃-dopamine receptor activity or activation to a greater extent than other types of dopamine receptor (e.g. D₁- or D₂- receptors). It is preferred that the compound is specific to D₂-dopamine receptors.

D₂-dopamine receptor agonists (i) above) are preferred compounds for use according to the invention. An example of a selective D₂-dopamine receptor agonist which is suitable for treating dyskinesias is 2-amino-6, 7-dihydroxytetralin (this agonist is about 10-fold more potent on D₂-dopamine receptors compared to D₁-dopamine receptors).

Other D₂-dopamine receptor agonists that may be used according to the present invention include:

(a) \(+/-\)6-chloro-7, 8-dihydroxy-3-ally1-phenyl-2,3,4,5-tetrahydro-1H-3benzazepine\(+/-\) + hydrobromide (SKF-82958);

(b) \(+/-\)1-phenyl-2,3,4,5-tetrahydro-(1H)-3-benzazepine-7,8-diol hydrochloride (SKF-38393);

(c) R\(+\)6chloro-7, 8-dihydroxy-1-phenyl-2,3,4,5-tetrahydro-1H-3-benzazepine hydrobromide (SKF-81297);

(d) trans-10,11-dihydroxy-5,6,6a,7,8,12b hexahydrobenzo[a] phenanthridine (Dihydrexidine); and

(e) \(-\)4,6,6a,7,8,12b-hexahydro-7 methyl-indolo[4,3-ab]phenanthridine (CY 208-243).

The compounds (and compositions or medicaments containing them) may be used to treat many types of dyskinesia. For instance the compounds may be used to treat dyskinesia associated with Huntington’s disease, idiopathic torsion dystonia,
tardive dyskinesia or off-dystonia in Parkinson’s disease and most particularly for dyskinesia associated with movement disorders such as parkinsonism (e.g. idiopathic Parkinson’s disease, post-encephalitic parkinsonism or parkinsonism resulting from head injury), treatment of schizophrenia, drug intoxication, the effect of toxins and the like.

The compounds may also be used in the treatment of dyskinesias which manifest as hyperkinetic activity (e.g. Tourette’s syndrome) and particularly basal ganglia-related hyperkinetic activity.

The compounds are also useful for treatment of dyskinesias which arise as a side-effect of other therapeutic agents. For instance, the compounds are useful for the treatment of dyskinesia associated with ropinirole, pramipexole, cabergoline, bromocriptine, lisuride, pergolide, L-DOPA or apomorphine treatment. The compounds are preferably used for the treatment of dyskinesia associated with L-DOPA or apomorphine treatment.

The compounds are particularly useful for treating dyskinesia caused by agents used to treat movement disorders such as parkinsonism. In this respect a preferred use of the compounds is in the treatment of dyskinetic side-effects associated with L-DOPA or apomorphine therapy for parkinsonism.

The compounds may be used to treat existing dyskinesias but may also be used when prophylactic treatment is considered medically necessary, for instance, when it is considered necessary to initiate L-DOPA therapy and it is feared that dyskinesias may develop.

The compounds may be used to treat dyskinesia as a monotherapy (i.e. use of the compound alone); as an adjunct to medicaments to prevent dyskinetic side-effects caused by the medicament (e.g. as an adjunct to L-DOPA or apomorphine given to treat parkinsonian patients) or alternatively the compounds may be given in combination with other compounds which also reduce dyskinesia (e.g. μ-opioid
receptor antagonists, δ-opioid receptor antagonists, α2-adrenoreceptor-antagonists, cannabinoid CB1-antagonists, NMDA receptor-antagonists, cholinergic receptor-antagonists, histamine H3-receptor agonists, globus pallidus/subthalamic nucleus lesion/deep brain stimulation).

The compounds are preferably used as an adjunct or in combination with known therapies. For instance, we have found that the combination of a D1 receptor agonist (e.g. L-DOPA) with a D2-dopamine selective receptor agonist results in movement disorders such as Parkinson’s disease being treated with significantly reduced dyskinetic side-effects.

The compositions of the first and second aspects of the invention may take a number of different forms depending, in particular on the manner in which the composition is to be used. Thus, for example, the composition may be in the form of a powder, tablet, capsule, liquid, ointment, cream, gel, hydrogel, aerosol, spray, micelle, liposome or any other suitable form that may be administered to a person or animal. It will be appreciated that the vehicle of the composition of the invention should be one which is well tolerated by the subject to whom it is given and enables delivery of the compounds to the brain.

The composition of the invention may be used in a number of ways. For instance, systemic administration may be required in which case the compound may be contained within a composition which may, for example, be ingested orally in the form of a tablet, capsule or liquid. Alternatively the composition may be administered by injection into the blood stream. Injections may be intravenous (bolus or infusion) or subcutaneous (bolus or infusion). The compounds may also be administered by inhalation (e.g. intranasally).

Compounds enhancing D2-dopamine receptor activity may also be administered centrally by means of intracerebral, intracerebroventricular, or intrathecal delivery.
The compound may also be incorporated within a slow or delayed release device. Such devices may, for example, be inserted under the skin and the compound may be released over weeks or even months. Such a device may be particularly useful for patients with long term dyskinesia such as patients on continuous L-DOPA therapy for the treatment of parkinsonism. The devices may be particularly advantageous when a compound is used which would normally require frequent administration (e.g. at least daily ingestion of a tablet or daily injection).

It will be appreciated that the amount of a compound required is determined by biological activity and bioavailability which in turn depends on the mode of administration, the physicochemical properties of the compound employed and whether the compound is being used as a monotherapy or in a combined therapy. The frequency of administration will also be influenced by the above mentioned factors and particularly the half-life of the compound within the subject being treated.

Known procedures, such as those conventionally employed by the pharmaceutical industry (e.g. in vivo experimentation, clinical trials etc), may be used to establish specific formulations of compositions and precise therapeutic regimes (such as daily doses of the compounds and the frequency of administration).

Generally, a daily dose of between 0.01µg/kg of body weight and 1.0g/kg of body weight of a compound which enhances D3-dopamine receptor activity may be used for the treatment of dyskinesia depending upon which specific compound is used more preferably the daily dose is between 0.01mg/kg of body weight and 100mg/kg of body weight.

Purely by way of example a suitable dose of 2-amino-6, 7-dihydroxytetralin for treating chloro-APB induced dyskinesia in subjects with Parkinson's disease is between 0.1mgs/kg/day and 100mgs/kg/day (depending upon the health status of the individual). It is preferred that between 0.25mgs/kg/day and 20mgs/kg/day of 2-amino-6, 7-dihydroxytetralin is given to a person daily and most preferred that about
1 – 5 mgs/kg/day are given. When given intravenously 0.1-10 mg/kg is a preferred dose whereas 30 mg/kg is a suitable dose orally.

Daily doses may be given as a single administration (e.g. a daily tablet for oral consumption or as a single daily injection). Alternatively the compound used may require administration twice or more times during a day. As an example, a D₂-dopamine receptor agonist for treating L-DOPA induced dyskinesia in patients with Parkinson’s disease may be administered as two (or more depending upon the severity of the dyskinesia) daily doses of between 25mgs and 5000mgs in tablet form. A patient receiving treatment may take a first dose upon waking and then a second dose in the evening (if on a two dose regime) or at 3 or 4 hourly intervals thereafter. Alternatively a slow release device may be used to provide optimal doses to a patient without the need to administer repeated doses.

A preferred means of using protein or peptide compounds which enhance D₂-Dopamine receptor activity for the treatment of dyskinesias is to deliver the compound to the brain by means of gene therapy. For instance, gene therapy may be used to increase expression of D₂-Dopamine receptors, increase expression of enzyme(s) responsible for the synthesis of endogenous D₂-Dopamine receptor agonists, decrease expression of a protein which promotes breakdown or desensitisation of D₂-Dopamine receptors or decrease expression of a protein which promotes breakdown of D₂-Dopamine receptor agonists. Therefore according to a fourth aspect of the present invention there is provided a delivery system for use in a gene therapy technique, said delivery system comprising a DNA molecule encoding for a protein which directly or indirectly enhances D₂-Dopamine receptor activity, said DNA molecule being capable of being transcribed to allow the expression of said protein and thereby treating a dyskinesia.

The delivery systems according to the fourth aspect of the invention are highly suitable for achieving sustained levels of a protein which directly or indirectly enhances D₂-Dopamine receptor activity over a longer period of time than is possible for most conventional therapeutic regimes. The delivery system may be used to
induce continuous protein expression from cells in the brain that have been transformed with the DNA molecule. Therefore, even if the protein has a very short half-life as an agent \textit{in vivo}, therapeutically effective amounts of the protein may be continuously expressed from the treated tissue.

Furthermore, the delivery system of the invention may be used to provide the DNA molecule (and thereby the protein which is an active therapeutic agent) without the need to use conventional pharmaceutical vehicles such as those required in tablets, capsules or liquids.

The delivery system of the present invention is such that the DNA molecule is capable of being expressed (when the delivery system is administered to a patient) to produce a protein which directly or indirectly has activity for enhancing D\textsubscript{3}-Dopamine receptor activity. By “directly” we mean that the product of gene expression \textit{per se} has the required activity. By “indirectly” we mean that the product of gene expression undergoes or mediates (e.g. as an enzyme) at least one further reaction to provide a compound effective for enhancing D\textsubscript{3}-Dopamine receptor activity and thereby treating a dyskinesia.

The DNA molecule may be contained within a suitable vector to form a recombinant vector. The vector may for example be a plasmid, cosmid or phage. Such recombinant vectors are highly useful in the delivery systems of the invention for transforming cells with the DNA molecule.

Recombinant vectors may also include other functional elements. For instance, recombinant vectors can be designed such that the vector will autonomously replicate in the cell. In this case, elements which induce DNA replication may be required in the recombinant vector. Alternatively the recombinant vector may be designed such that the vector and recombinant DNA molecule integrates into the genome of a cell. In this case DNA sequences which favour targeted integration (e.g. by homologous recombination) are desirable. Recombinant vectors may also have DNA coding for genes that may be used as selectable markers in the cloning process.
The recombinant vector may also further comprise a promoter or regulator to control expression of the gene as required.

The DNA molecule may (but not necessarily) be one which becomes incorporated in the DNA of cells of the subject being treated. Undifferentiated cells may be stably transformed leading to the production of genetically modified daughter cells (in which case regulation of expression in the subject may be required e.g. with specific transcription factors or gene activators). Alternatively, the delivery system may be designed to favour unstable or transient transformation of differentiated cells in the subject being treated. When this is the case, regulation of expression may be less important because expression of the DNA molecule will stop when the transformed cells die or stop expressing the protein (ideally when the dyskinesia has been treated or prevented).

The delivery system may provide the DNA molecule to the subject without it being incorporated in a vector. For instance, the DNA molecule may be incorporated within a liposome or virus particle. Alternatively the “naked” DNA molecule may be inserted into a subject’s cells by a suitable means e.g. direct endocytotic uptake.

The DNA molecule may be transferred to the cells of a subject to be treated by transfection, infection, microinjection, cell fusion, protoplast fusion or ballistic bombardment. For example, transfer may be by ballistic transfection with coated gold particles, liposomes containing the DNA molecule, viral vectors (e.g. adenovirus) and means of providing direct DNA uptake (e.g. endocytosis) by application of the DNA molecule directly to the brain topically or by injection.
The invention will be further illustrated in the non-limiting Example and figures, in which:

Figure 1 illustrates that the activation of D5 receptors enhances the discharge frequency of subthalamic neurones. Top traces display representative examples of electrical activity and Bottom bar graphs summarise the measurements. **A:** Spontaneous firing frequency was enhanced in the presence of specific agonists in the D1 family, SKF 82958 and SKF 38393 at 10 μM. **B:** The firing response to a step of current (+80 pA for 1 s) was increased in presence of D1 agonists (10 μM).

Figure 2 illustrates that the activation of D5 receptors potentiates plateau potentials and low threshold spikes in subthalamic nucleus. Top traces display representative examples of electrical activity. Bottom bar graphs summarise the measurements. **A:** In about half subthalamic neurones, a short depolarising current pulse (+80 pA) triggers a plateau potential that outlasts the duration of the stimulus. The number of action potentials per plateau potential (PP) was increased by the D5 agonists (10 μM) whereas its duration was not changed. **B:** All subthalamic nucleus neurones respond to the break of a negative step (-80 pA) by a low threshold spike (LTS) with a few action potentials. D5 stimulation increased the number of action potentials fired per LTS without changing the duration of LTS; and

Figure 3 illustrates that a specific antagonist of D5 receptors does not change excitability but prevents the actions of D5 agonists in subthalamic neurones. Top traces display representative examples of electrical activity. Bottom bar graphs summarise the measurements. Data from evoked firing responses and plateau potentials were pooled. **A:** The evoked firing response was not change in the presence of the specific D5 antagonist, SCH23390 (10 μM). **B:** Co-application of a D5 agonist (5 μM) and 10μM of SCH 23390 did not produce significant changes in the number of action potentials per PP.
EXAMPLE

Experiments were performed to demonstrate that the stimulation of D5 receptors will reduce dyskinetic symptoms.

The basis of this study was to demonstrate that stimulation of D5 receptors increases the excitability of subthalamic neurones.

1. Background

Dopamine receptors can be classed into two major families, the D1-like family (which includes D1 and D5 receptors) and the D2-like family (which includes D2, D3 and D4 receptors). The subthalamic nucleus is a key structure in the circuitry responsible for generating movement disorders. These include, but are not limited to, abnormal involuntary movement such as those seen following dopamine replacement therapy in Parkinson's disease (e.g. levodopa-induced dyskinesia), dystonia and tardive dyskinesia observed after neuroleptic therapy. A pathological feature of these hyperkinetic disorders is that the subthalamic nucleus is underactive and is responsible, in part at least, for generating symptoms. Thus, any process, which acts to increase the activity of the subthalamic nucleus, will be valuable in alleviating the symptoms of these hyperkinetic disorders.

Recent reports have established that there is a dopaminergic projection from the substantia nigra pars compacta (SNc) to the subthalamic nucleus (STN). Although, STN neurons express mRNAs of both D1 and D5 dopamine receptors, only D5 dopamine receptors have been detected in the STN by immunocytochemistry. D1 dopamine receptors cannot be found in this structure. Therefore, effects seen with dopamine D1-like agonists in the STN can be assumed to be mediated by the D5 dopamine receptor.

In the current study, we demonstrate that stimulation of the D5 receptor increases the activity of STN and thus represents an action to reduce hyperkinetic / dyskinetic movements.
2. Materials and methods

*Slice preparation:* Experiments were performed on STN neurones in 400 µm-thick coronal slices obtained from 18-22 day-old Wistar rats as described in Baufreton et al (2001) J Neurophysiol 86:75-85.

One slice was transferred to an immersion-type recording chamber and continuously superfused (3.5 ml/min) with Krebs' solution bubbled with 95% O₂ and 5% CO₂ at room temperature (~20°C). The Krebs' solution contained (in mM): 124 NaCl, 26 NaHCO₃, 3.6 KCl, 1.3 MgCl₂, 2.4 CaCl₂, 1.25 HEPES, and 10 glucose (pH 7.4).

Recordings were made using the blind patch-clamp technique in the whole-cell configuration. Current-clamp mode was used. Pipette solution contained (in mM): 120 K gluconate, 10 KCl, 10 NaCl, 11 EGTA, 10 HEPES, 1 CaCl₂, 2 ATP-Mg, and 0.4 Na-GTP. The osmolarity of the intrapiette solution was between 280 and 300 mOsm and its pH adjusted to 7.25. Electrodes were pulled from borosilicate thin-glass capillaries (GC150F-15, Harvard Apparatus, Edenbridge, UK) on a vertical puller (PP-830, Narishige, Japan) and had a resistance of ~12 MΩ. Signals were recorded using an Axopatch-1D amplifier (Axon Instruments, Foster City, CA) with the amplifier filter set at 5 kHz and continuously stored on videotape. Quantification was made on portions of signals digitised at 2.5 KHz using a Digidata 1200B (Axon Instruments, USA) and pClamp 6.01 software (Axon Instruments, USA). Access resistance (~ 20 MΩ) was regularly monitored. Junction potential was measured according to the method described by Neher (1992) and voltage error corrected off line.

All drugs were purchased from Sigma (St. Louis, MO, USA) except SKF 38393, which was purchased from Tocris (Bristol, UK). They were prepared as stock-solutions and stored at ~80°C. The solvent was water, except in the case of SKF 82958 (prepared in ethanol). Drugs, diluted in the oxygenated Krebs' solution, were
delivered by means of a multi-barrel gravity-feed system (HSSE-2, ALA Scientific Instruments Inc, Sega Electronique, FR) composed of 2 capillaries positioned just above the patch pipette allowing up to 7 solutions to be tested successively. The minimal duration of a single application was 2 s. Shorter applications produced inconsistent results, presumably because of the time taken by drugs to reach the recorded neurone in the slice. Final dilution of the solvent was always kept below 0.002.

*Experimental protocol:* After about 7 min of observation in voltage and current clamp, recording was initiated (or the cell was discarded if its behaviour appeared not to be typical; cf. Baufreton et al., 2001). Current-clamp mode in the presence of antagonists of fast synaptic transmission (GABA_A receptor antagonist, bicuculline 10 μM; NMDA receptors antagonist, APV, 40 μM; AMPA/kainate receptors antagonist, CNQX, 10 μM) was then used throughout except for shortly checking seal parameters in voltage clamp.

Three drugs were used: SKF82958 and SKF38393 to stimulate D5 receptors (5 or 10 μM), and SCH 23390 to block D5 receptors (10 μM or 20 μM). Electrical activity after 3 to 5 min of application of a drug (test) was compared to pre-test (control) electrical activity. Results obtained with the 2 agonists were pooled since no difference in their actions was noticed.

Electrical activity was assayed by measuring at least one of the following three parameters:

1. spontaneous firing frequency (at zero current level) or evoked firing frequency induced by a long stimulation (±80 pA / 1000ms, the membrane being held around -70 mV by current injection).

2. firing frequency during and length of plateau potential (PP, induced by a depolarising current pulse, +80 pA / 200 ms, membrane held around -70 mV)

3. firing frequency during and length of low threshold spike (LTS, induced by an hyperpolarising current pulse, -80 pA / 1 s, membrane held around -70 mV).
Parameter (1) delineates the firing of neurones in the single-spike firing mode at resting membrane potential whereas parameters (2) and (3) describes the firing properties of neurones at hyperpolarized membrane potential, usually found in the burst-firing mode (Beurrier et al., 1999 J Neurosci 19:599-609). In each condition (control or test), any parameter value was obtained from the mean of at least 4 measures.

3. Results

The present study is based on a total of 25 neurones. Of these neurones, 75% were found to be responsive to stimulation of the D5 receptor (see Table 1). Activation of D5 receptors significantly increased the frequency of spontaneous and evoked firing of STN neurones (see Fig. 1). Furthermore, activation of D5 receptors significantly increased plateau potentials and low threshold spikes in subthalamic neurones (see Fig. 2). Blockade of D5 receptors had no significant action when applied alone, but antagonised the effect of D5 receptor stimulation in the responsive neurones (Fig. 3).

4. Summary

- STN neurones display functionally active post-synaptic dopamine D5 receptors.

- D5 receptor activation potentiates the firing frequency of subthalamic neurones in a way that would reduce dyskinesia.

- This is a D5 receptor specific effect since it was no longer found following blockade of the D5 receptor.

Conclusions

L-DOPA-induced dyskinesia, and other dyskinesias, are associated with a reduced activity of subthalamic neurones. These data demonstrate that activation of D5 dopamine receptors alleviates this underactivity by increasing the firing frequency
of subthalamic neurons and thus reduces or removes dyskinesia.

Therefore, D5 dopamine receptor agonists are demonstrated to be useful in the treatment of L-DOPA-induced dyskinesia and other dyskinesias such as dystonia and tardive dyskinesia.
<table>
<thead>
<tr>
<th>D5 stimulation (n=12)</th>
<th>1</th>
<th>2</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Number of action potential / PP</strong></td>
<td><strong>Duration of PP (in ms)</strong></td>
<td><strong>Number of action potential / LTS</strong></td>
<td><strong>Duration of LTS (in ms)</strong></td>
</tr>
<tr>
<td><strong>Firing frequency of spontaneous activity (in Hz)</strong></td>
<td><strong>P=0.06 ; t =-2.5</strong></td>
<td><strong>38.9 ± 4.4</strong></td>
<td><strong>656.4 ± 114.6</strong></td>
</tr>
</tbody>
</table>

**D5 blockade (n=5)**

| **Firing frequency of evoked activity** (in Hz) | **P=0.55 ; t=-0.6** | **27.2** | **79** | **104.2** | **10.4** | **11.4** | **16.2** | **15.2** | **28** | **20** | **5** |

**D5 stimulation + blockade (n=4)**

| **Firing frequency of evoked activity** (in Hz) | **P = 0.62 ; t=0.5** | **22** | **12.7** | **17.2** | **66** | **40.7** | **20** | **18** | **4** |

* Tendencies will be significant with an increased sampling. So it will be considered to be significant.

** Values from evoked firing responses and plateau potentials.
CLAIMS

1. The use of a compound which enhances D₅-dopamine receptor activity, or activation, for the manufacture of a medicament for the treatment of dyskinesia.

2. The use according to claim 1, wherein the compound is a D₅-dopamine receptor agonist.

3. The use according to claim 2 wherein the D₅-Dopamine receptor agonist is 2-amino-6, 7-dihydroxytetralin or a functional analogue thereof.

3. The use according to claim 2 wherein the D₅-Dopamine receptor agonist is one of:

   (+/-)6-chloro-7, 8-dihydroxy-3-allyl-phenyl-2,3,4,5-tetrahydro-1H-3benzazepine++ + hydrobromide (SKF-82958);

   (+/-)1-phenyl-2,3,4, 5-tetrahydro-(1H)-3-benzazepine-7,8-diol hydrochloride (SKF-38393);

   R(+)6chloro-7, 8-dihydroxy-1-phenyl-2,3,4,5-tetrahydro-1H-3-benzazepine hydrobromide (SKF-81297);

   trans-10,11-dihydroxy-5,6,6a,7,8,12b hexahydrobenzo[a] phenanthridine (Dihydrexidine); or

   (-)4,6,6a,7,8,12b-hexahydro-7 methyl-indolo[4,3-ab]phenanthridine (CY 208-243)

5. The use according to claim 2, wherein the compound is a selective D₅-dopamine receptor agonist.

6. The use according to claim 1 wherein the compound is an inhibitor of MAO-B or COMT.
7. The use according to any preceding claim, for the treatment of dyskinesia associated with movement disorders.

8. The use according to claim 7, for the treatment of dyskinesia associated with parkinsonism.

9. The use according to claim 8 wherein the parkinsonism is idiopathic Parkinson’s disease or post-encephalitic parkinsonism.

10. The use according to claim 8 wherein the parkinsonism results from head injury, the treatment of schizophrenia, drug intoxication or manganese poisoning.

11. The use according to any one of claims 1 - 6 for the treatment of dyskinesia associated with Huntington’s disease, idiopathic torsion dystonia, tardive dyskinesia or off-dystonia in Parkinson’s disease.

12. The use according to any one of claims 1 - 6 for the treatment of dyskinesia which arises as a side-effect of a therapeutic agent.

13. The use according to claim 12 for the treatment of dyskinesia associated with agents used to treat movement disorders.

14. The use according to claim 12 or 13 wherein the agent is L-DOPA, chloro-APB or apomorphine.

15. The use according to claim 12 wherein the agent is used to treat parkinsonism.

16. The use according to any preceding claim for prophylactic treatment of dyskinesia.
17. A method for the treatment of dyskinesia comprising administering to a person or animal in need of such treatment a therapeutically effective amount of a compound which enhances D_2-Dopamine receptor activity, or activation.

18. The method according to claim 17 comprising administering a compound or compounds as defined in any one of claims 1 to 16.
FIG. 1

A  Spontaneous activity
-63 mV
-59 mV
20 mV
1 s

B  Evoked activity
Control
-79 mV
+ D5 stimulation
-79 mV
20 mV
250 ms

Discharge frequency (%)

(n=5) n.s.

control  D5 agonist

(n=6) ###

control  D5 agonist
FIG. 3

A  Control

B  Control: D5 stimulation

+ D5 blockade

D5 stimulation + D5 blockade

nb of action potentials (%)

(n=5)
n.s.

control  D5 antagonist

nb of action potentials (%)

(n=4)
n.s.

control  D5 agonist + D5 antagonist