Cannabis – From Bench to Bedside

Dr David Potter JP

GW Pharma Ltd

Outline of talk

- Cannabis History and Legal Issues
- GW Pharma – Early Days
- Cannabis Botany and Pharmacology
- Cannabinoid Biosynthesis
- Changes in illicit ‘medicinal’ cannabis
- Licensed medicinal cannabis propagation
- Processing and formulation
- Looking ahead

Medicinal Cannabis History

Recorded medicinal use:

Sumeria (3000 BCE), China (2600 BCE), Egypt (1600 BCE), India (1500 BCE).

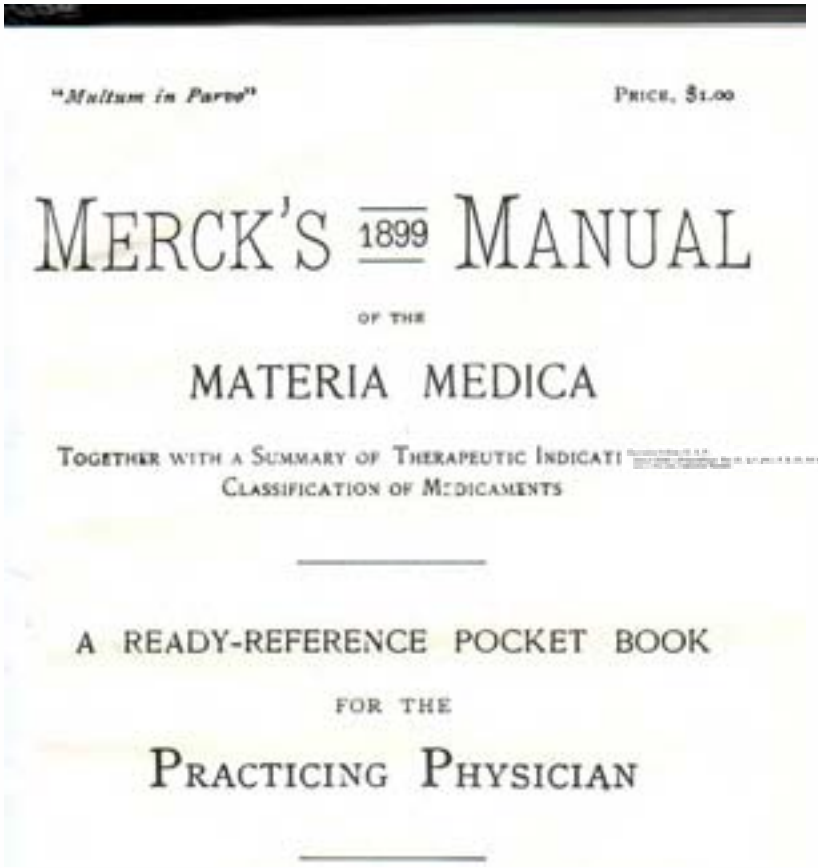
Introduced to Britain from India by Dr WB o'Shaughnessy

His tinctures of cannabis found various uses: -

analgesic, muscle relaxant, anticonvulsant, oxytocic, hypnotic, bronchodilator.

Robson P. 2009 Forbidden Drugs

Cannabis appears in Merck's Manual 1899



"Multum in Parvo" PRICE, $1.00

MERCK'S 1899 MANUAL

OF THE

MATERIA MEDICA

TOGETHER WITH A SUMMARY OF THERAPEUTIC INDICATI
CLASSIFICATION OF MEDICAMENTS

A READY-REFERENCE POCKET BOOK

FOR THE

PRACTICING PHYSICIAN

INDICATIONS.

Pain.—*See also, After-Pains, Anesthesia, Boils, Chest Pains, Colic, Gastralgia, Headache, Hepatalgia, Inflammation, Lumbago, Myalgia, Neuralgia, Neuritis, Odontalgia, Otalgia, Ovarian Neuralgia, Rheumatism, etc. Also lists of Antigesics, Anesthetics and Narcotics.*

Acetanilid.
Acid, Carbolic.
Aconite.
Aconitine.
Ammonium Iodide.
Atropine.
Belladonna.
Camphor, Monobromated.
Camphor-phenol.
Cannabis Indica.
Chloroform.
Chloral Hydrate.
Chloral-Camphor.
Cocaine.
Codeine.

Cannabis Indica—U. S. P.

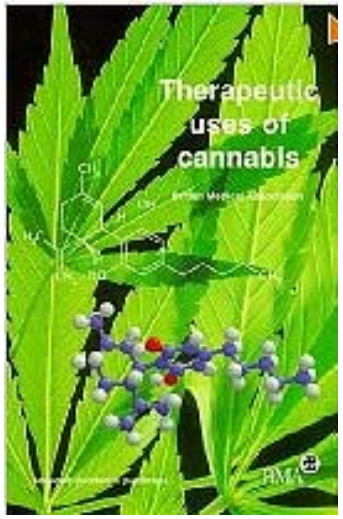
INDIAN HEMP.—*Preparations:* Ext. (D., ¼-1 grn.); F. E. (D., 2–5 min.); Tr. (D., 5–20 min.).—See also, Cannabine Tannate.

Medicinal cannabis: reasons for decline

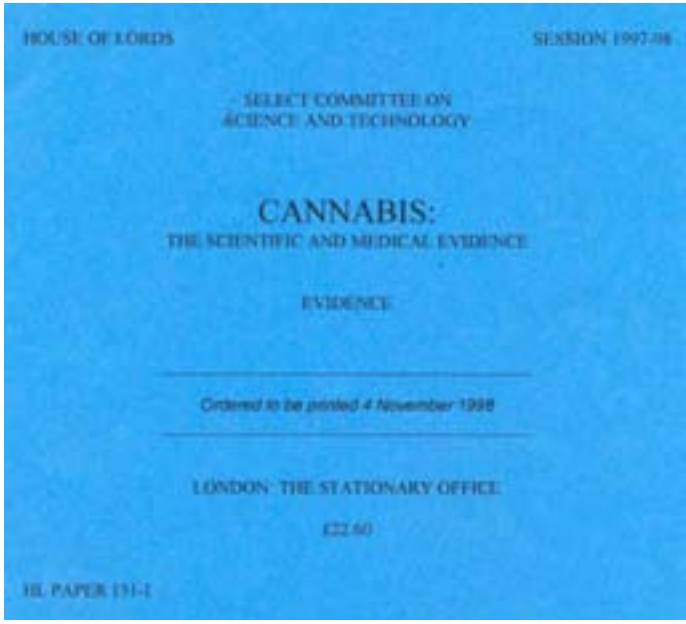
- Variable potency
- Unreliable supply
- Poor stability
- Unpredictable response by oral route
- Lack of clarity over dose
- Increasing emphasis on synthetic drugs
- Increasing concern over recreational use
- Declared Schedule 1 Drug in Misuse of Drugs Act 1971

Notcutt W, 2004. Cannabis in the treatment of chronic pain. In *The Medicinal Use of Cannabis and Cannabinoids*

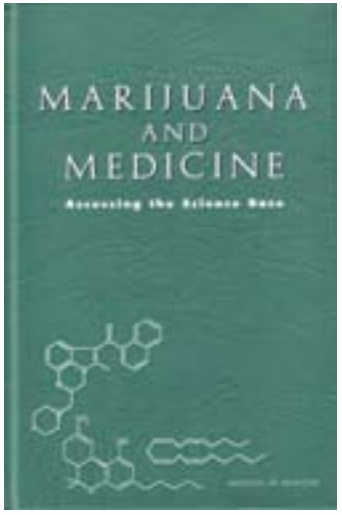
As 20th Century Closes - A growing medical acceptance of cannabis



- **1997 BMA *Therapeutic Uses of Cannabis***

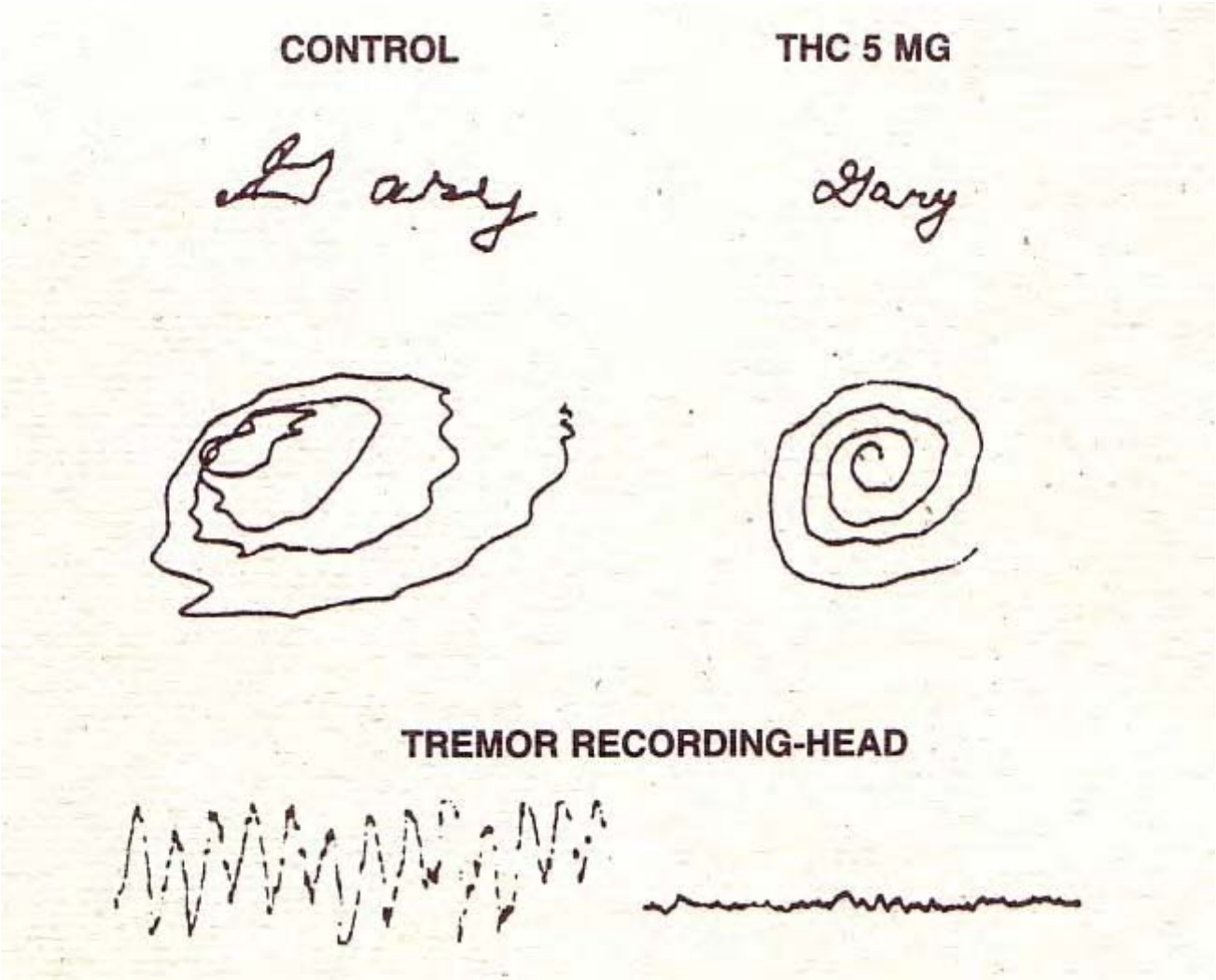


- **1998: UK House of Lords Select Committee Inquiry into Medicinal Cannabis**



- **1999: US Institute of Medicine Report: - *Marijuana and Medicine***

THC effects on tremor in MS
Just one example of proven efficacy



Clifford CB 1983. Tetrahydrocannabinol for tremor in MS. Annals of Neurology; **13**: 13-15

GW Early days – Autumn 1998

Britain's first government-licensed cannabis is to be harvested secretly this week by a specially vetted team of **mature botanists**.

No younger staff were employed because of fears that they might mix business with pleasure.

The crop has been guarded round the clock as hundreds of fully potent plants have reached eight feet in the past four months.

Only GW and the Home Office know the location of the **greenhouse** in southern England.

The Times. 28th December 1998

The first crop is hung to dry

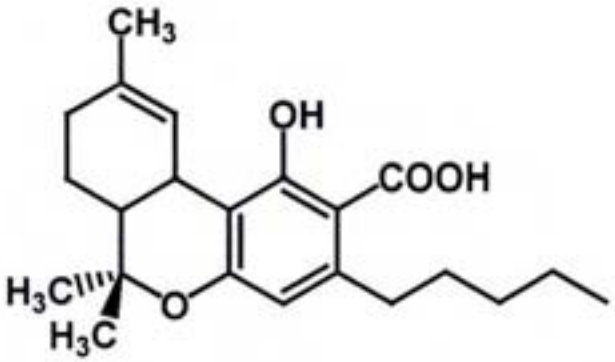
The Guardian. 5th January 1999

Cannabis Pharmacology

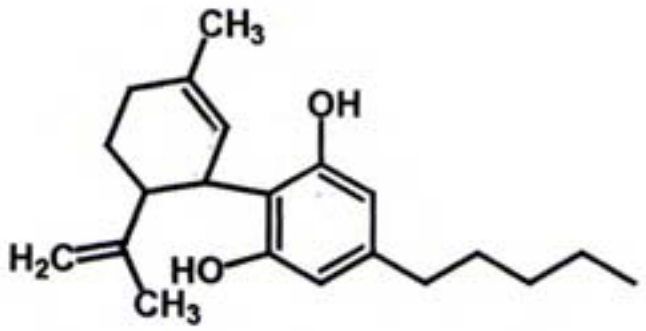
Main active ingredients are the (phyto)cannabinoids - terpenophenolic compounds unique to *Cannabis sativa*

70 identified - *a neglected pharmacological treasure trove* (Mechoulam. *Br J Pharmacol.* 2005 December)

The most studied are THC and CBD



THCA



CBD

Interaction of cannabinoids and cannabinoid receptors

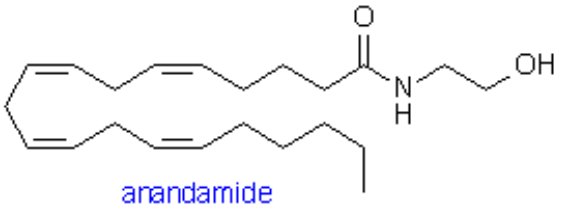
CB_1 cannabinoid receptors found in mammalian brain and CNS (Devane et al 1988), subsequently cloned (Matsuda et al 1990)

Member of superfamiliy of G-protein-linked receptors, through which over half of all known drugs work (Alberts et al 2002)

CB_2 receptors found in spleen macrophages, not CNS (Pertwee 1997), and appear to regulate cytokinine activity altering cell-to-cell communication (Pertwee 2004)

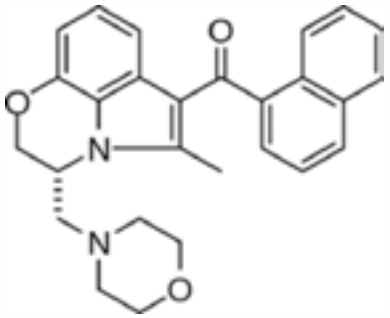
Interaction of cannabinoids and cannabinoid receptors

'Phytocannabinoid' function explained by discovery of '*endocannabinoids*' (Devane et al 1992)

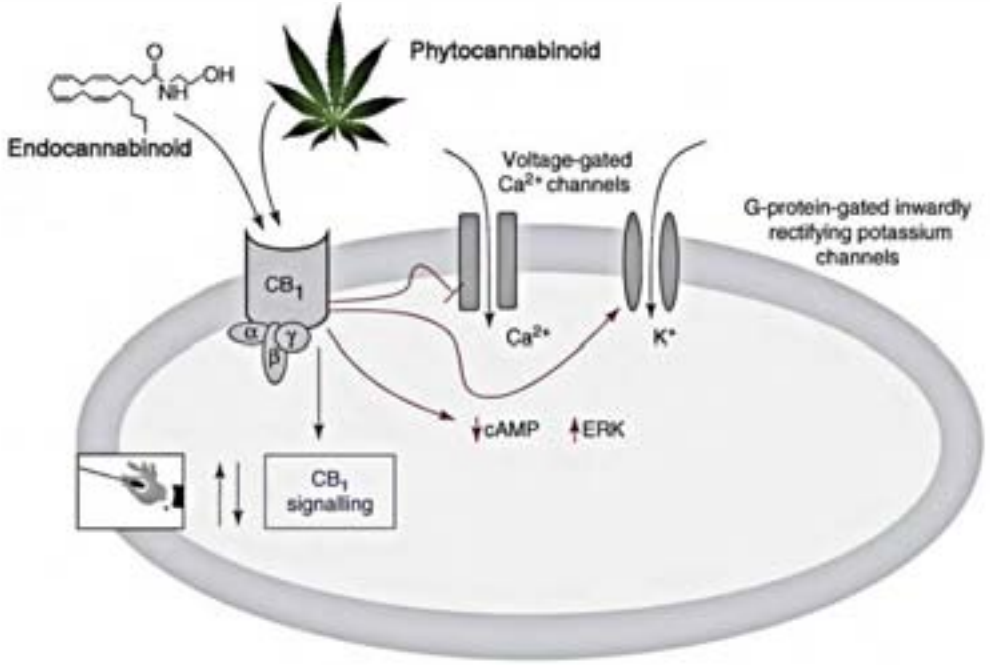


anandamide

Synthetic cannabinoids (eg WIN 55,212-2) can also act as CB receptor agonists



WIN 55,212-2



Interaction of endo or phyto-cannabinoids with the CB_1 receptor in the eukaryote cell and effects on cAMP, ERK activity and ion channelling. (Ross 2007).

Interaction of cannabinoids and cannabinoid receptors

THC interacts with CB_1 receptor causing psychoactivity (BMA 1997)

CBD (cannabidiol) has a weak affinity for CB_1 and CB_2 receptors but has significant levels of antipsychotic effects

CBD and THC equally potent neuroprotective antioxidant properties

β caryophyllene binds to CB_2. Anti-inflammatory in mice (Gertsch 2008)

Monoterpenes also thought to interact with cannabinoids (McPartland and Russo 2001)

Cannabis plant extracts significantly more active than cannabinoids alone (Williamson and Evans 2000)

THC Distribution in Female Cannabis

Seeds	0.0%
Roots	0.0%
Stem	0.3%
Leaves	0.8%
Seeded Female Flowers	6.3%
Unseeded Female Flowers	15.2%

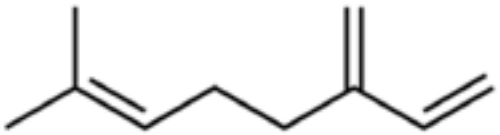
Male (left)
Female (right)

Foliar and floral secondary metabolites

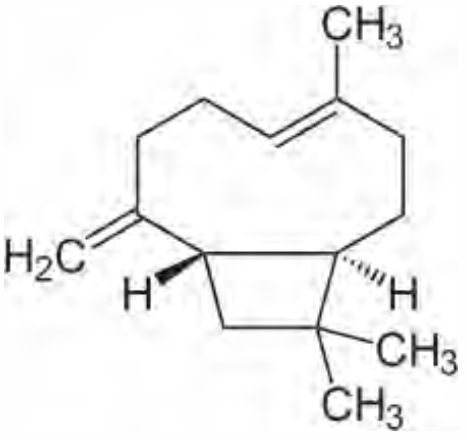
- Foliar and floral tissues differ in terpene profile

- Leaves: - High sesquiterpene (C_{15})
  Low monoterpene (C_{10})

- Flowers: - Low sequiterpene (C_{15})
  High monoterpene (C_{10})



Myrcene $C_{10}H_{16}$

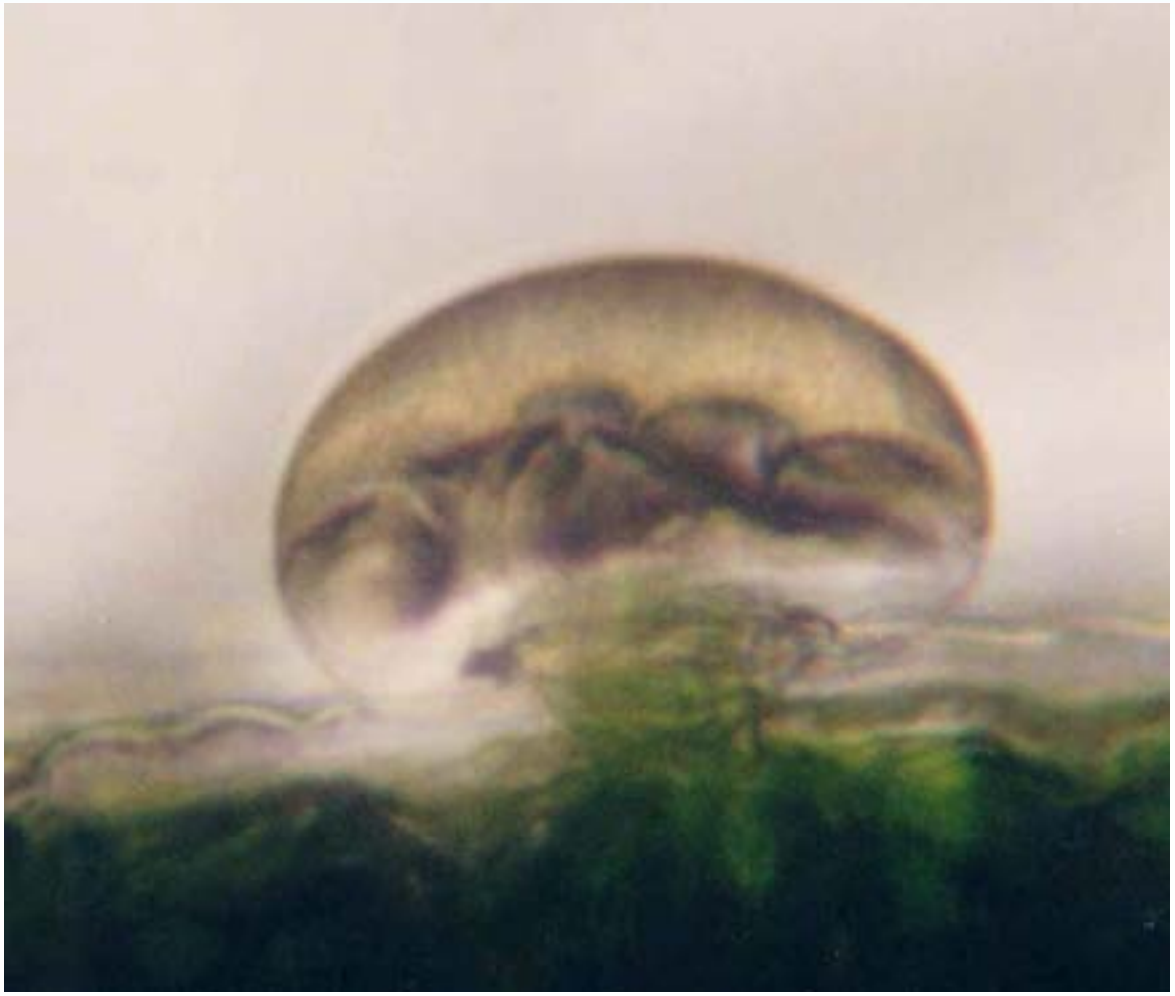


β caryophyllene $C_{15}H_{24}$

Cannabis Trichomes

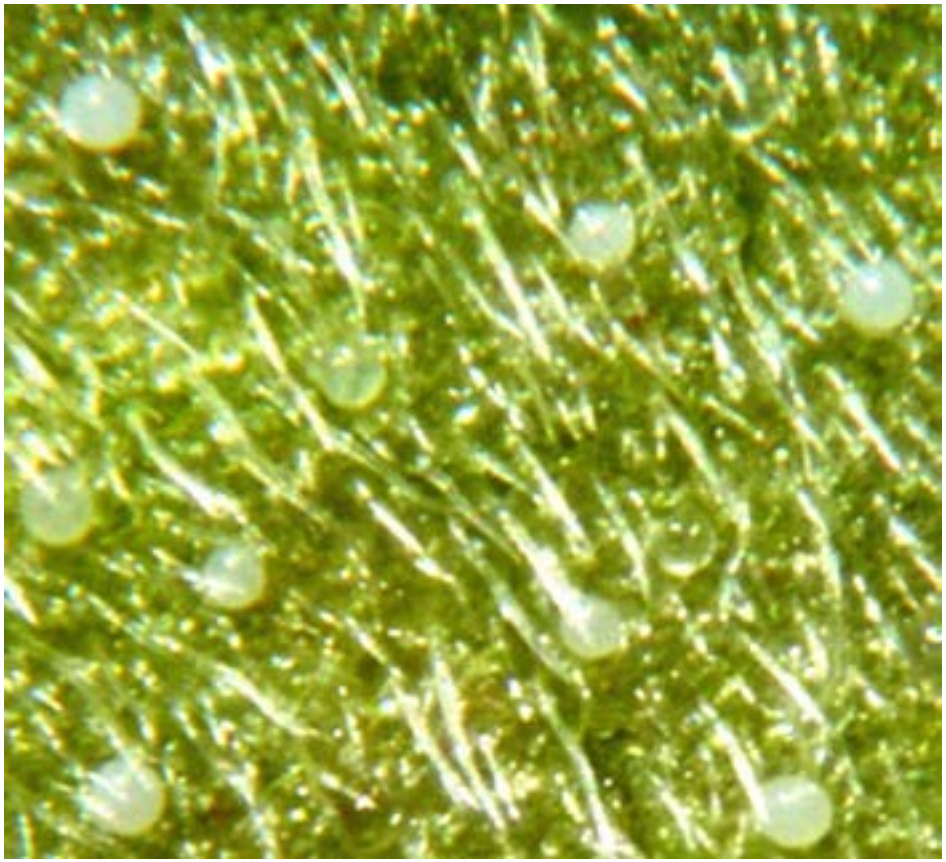
Seven day old cannabis leaf

Capitate Sessile Trichome on *Cannabis sativa* L



Cannabinoids and essential oils are secreted by cells at the base of the trichome, and sequestered in the space above.

Ageing Sessile Trichomes on *Cannabis sativa* L



Sessile trichomes on fresh sample of *Cannabis sativa*

Sessile trichomes on ancient sample of *Cannabis sativa*

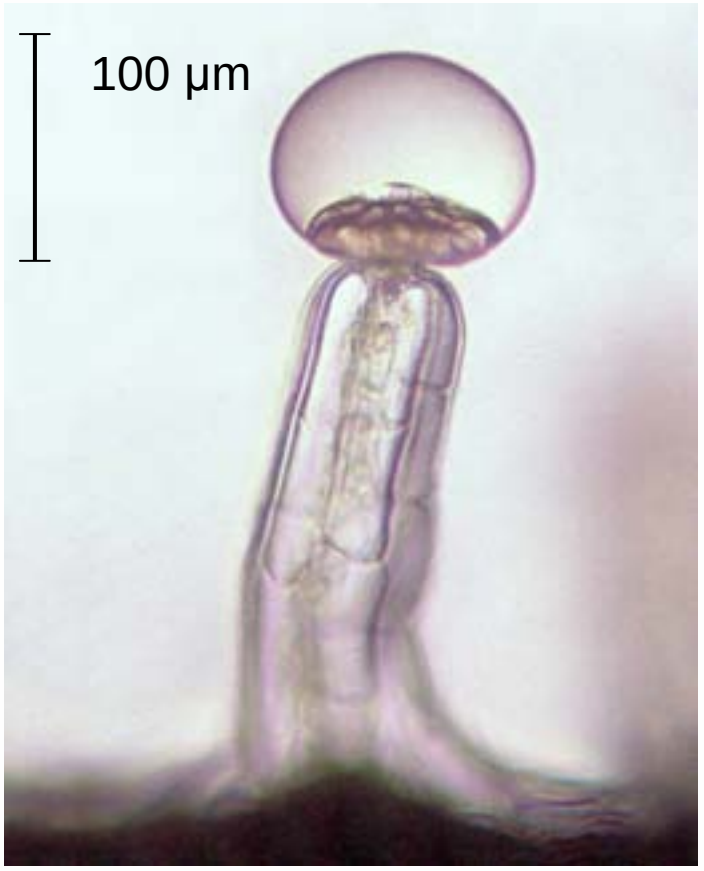
Aged Sessile Trichomes on *Cannabis sativa* L

Russo EB, Jiang HE, Li X, Sutton A, Carboni A, del Bianco F, Mandolino G, Potter DJ, et al

Phytochemical and genetic analyses of ancient cannabis from Central Asia.

J. Exp. Bot. 59, 15, 4171-4182

Capitate stalked trichomes



Typical terpene profile: -

25% bitter sesquiterpenes and 75% fragrant monoterpenes

Likely functions phytophagous predators repellent?

Capitate-Stalked Trichomes on Female Inflorescence

The pubescence of trichomes acts as a garment, insulating the plant tissue.

Infra red and UV light are reflected. The plant is thus protected from heat and sun burn.

Insect movement is slowed, but above all the material tastes repugnant.

David Potter PhD Thesis. Kings College London. May 2009

Insect Entrapment in *Cannabis sativa* L

Beware of the flowers, ('cos I am sure they're going to get you yeah!)

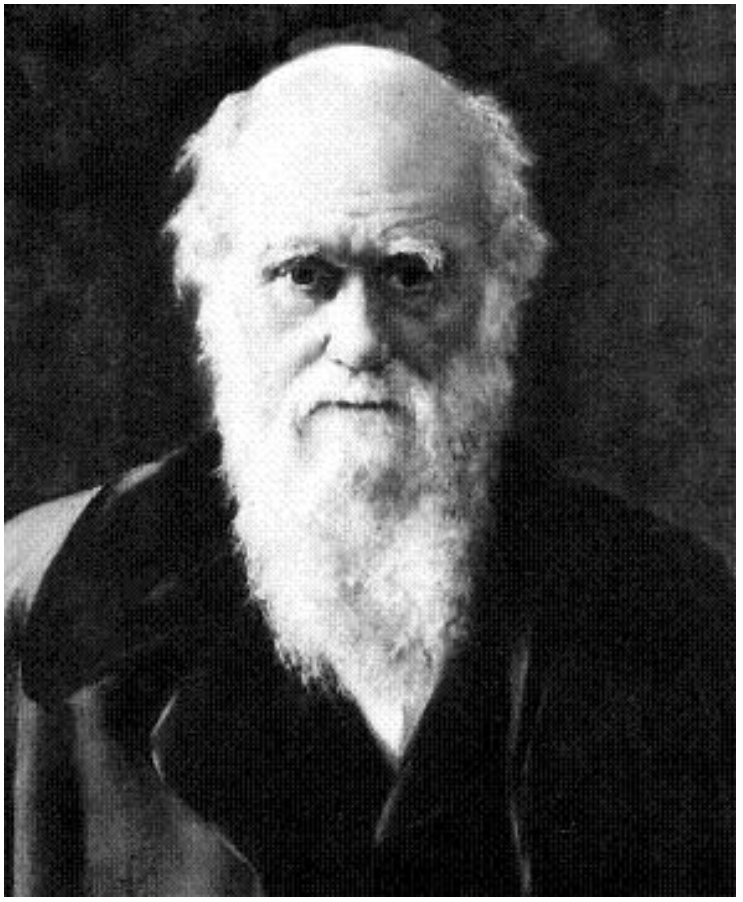
John Otway

Cotton-melon aphid irreversibly glued to cannabis trichomes. The aphid can emit alarm pheromones, warning others. Another possible minor benefit of trichomes but.....

Trichomes – an inherited aid to survival

Individuals having any advantage, however slight, over others, would have the best chance of surviving and procreating their kind.

The Origin of Species



Charles Darwin JP

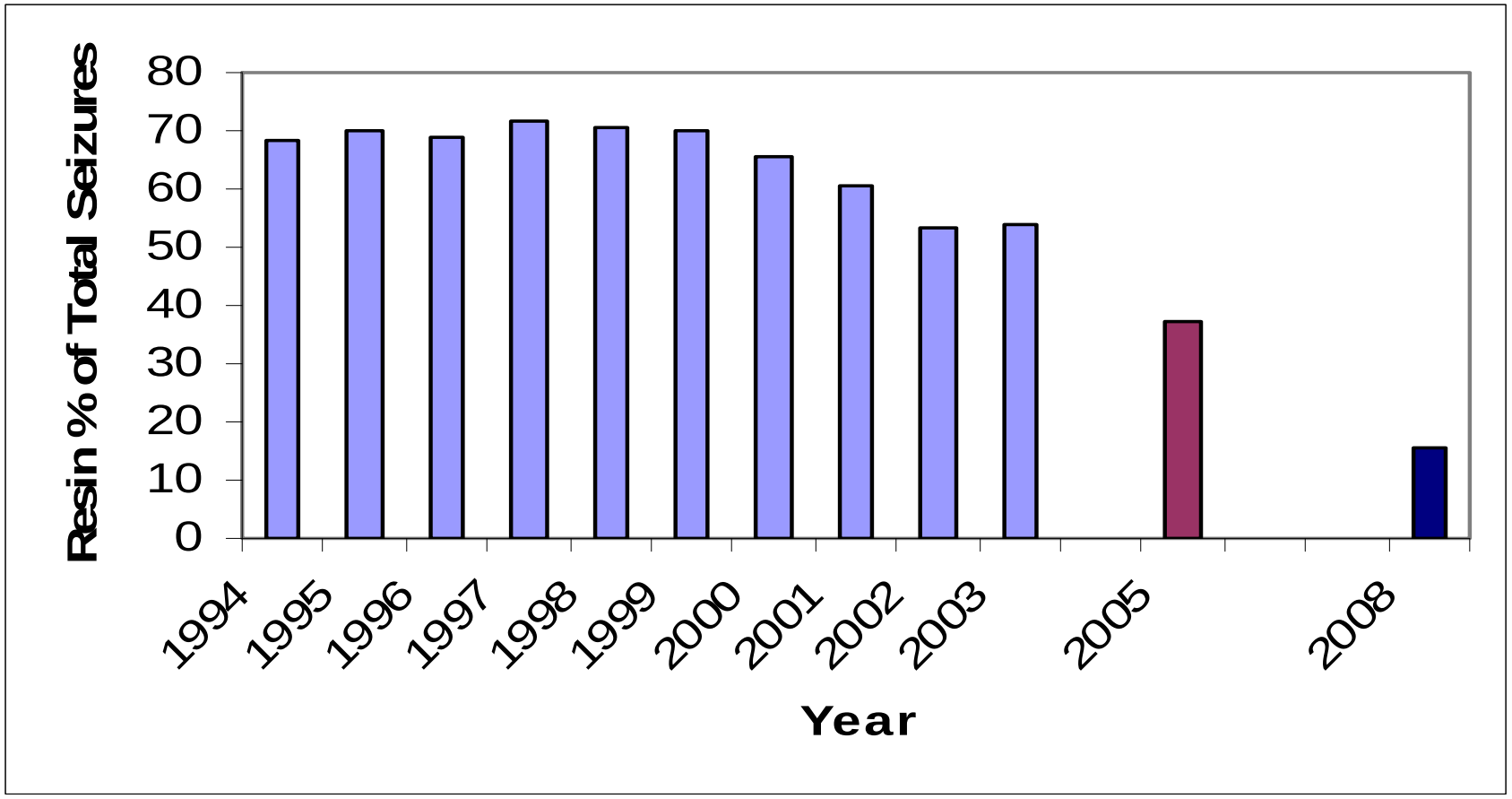
Principal forms of illicit cannabis

Traditional Resin

Sinsemilla (Skunk)

Imported Herbal

Decline in Dominance of Cannabis Resin

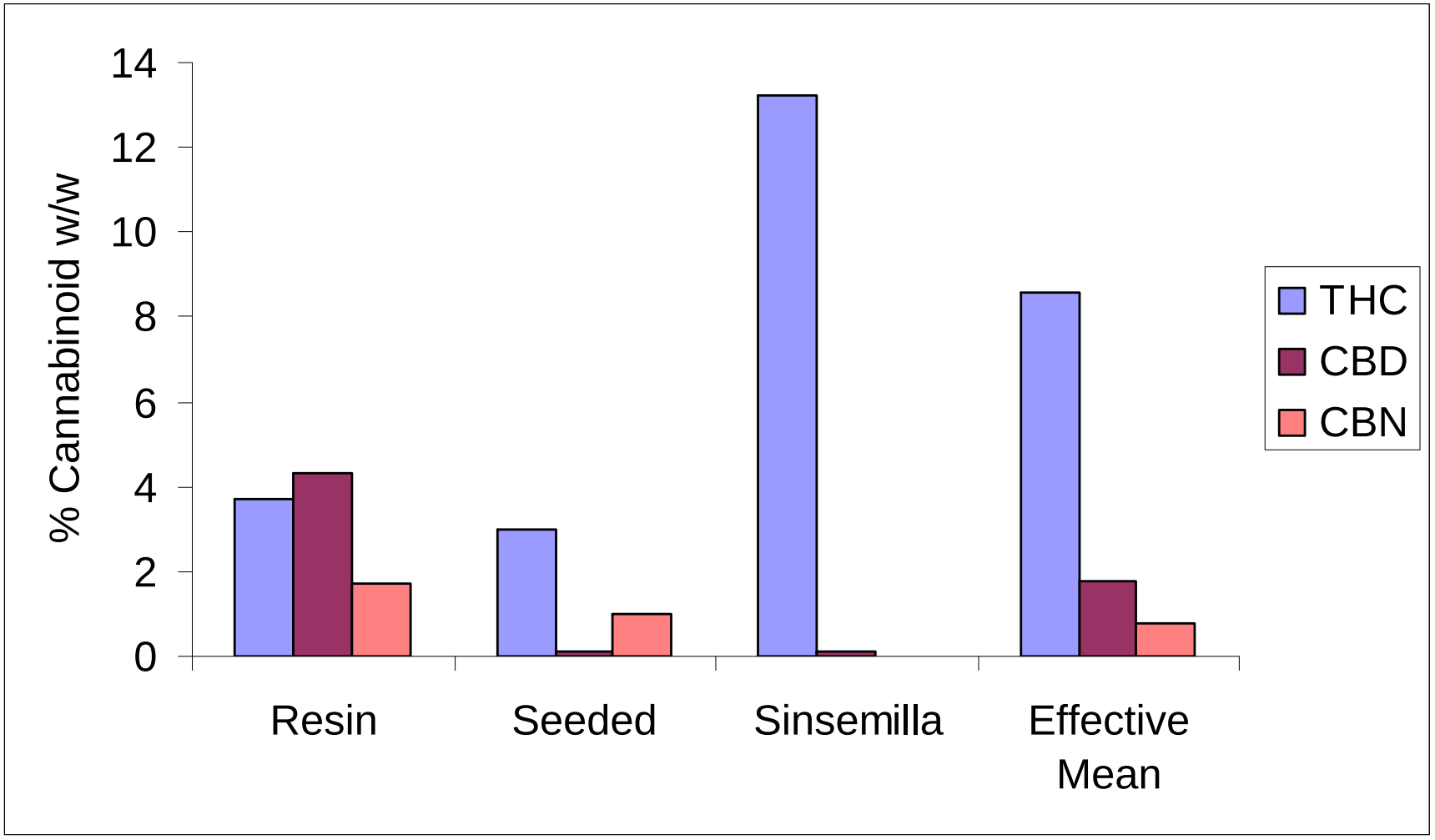


Mwenda et al. Home Office (2005) Findings 265 - Seizures of Drugs in England and Wales 2003

Potter DJ, Clark P and Brown MB. (2008) Journal of Forensic Sciences; 53:1 90-94

King L and Hardwick S (2008) Home Office Cannabis Potency Study

2005 'Effective' potency summary
CBD in decline in illicit UK Cannabis



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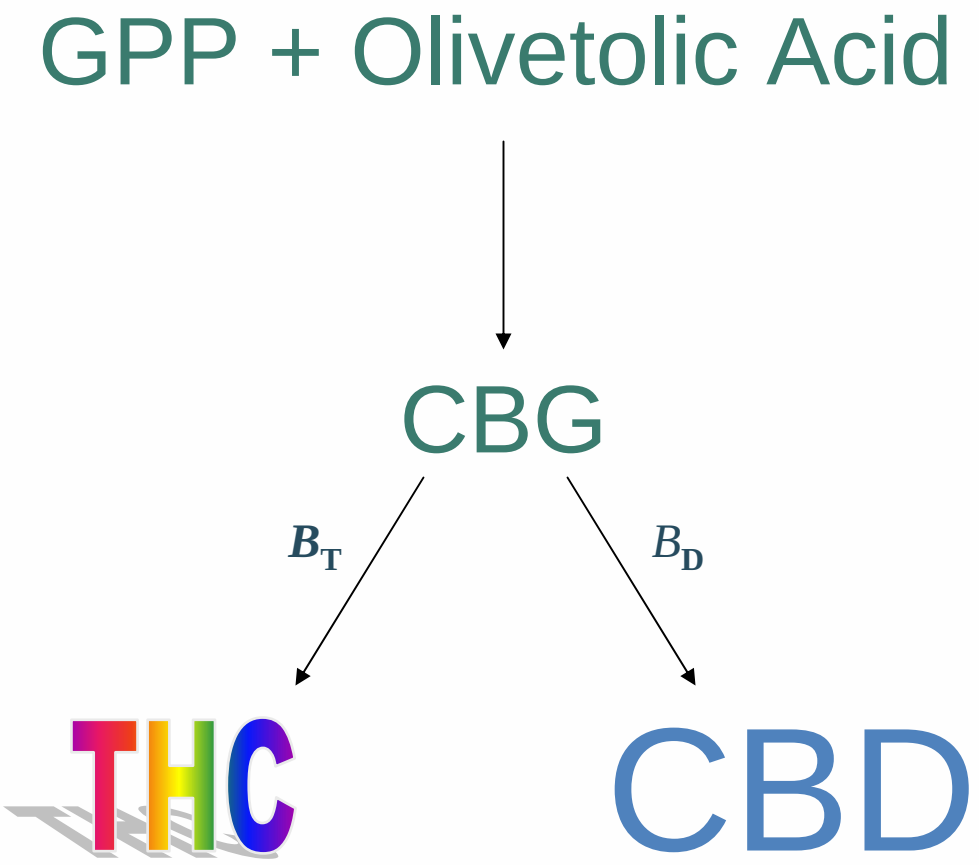
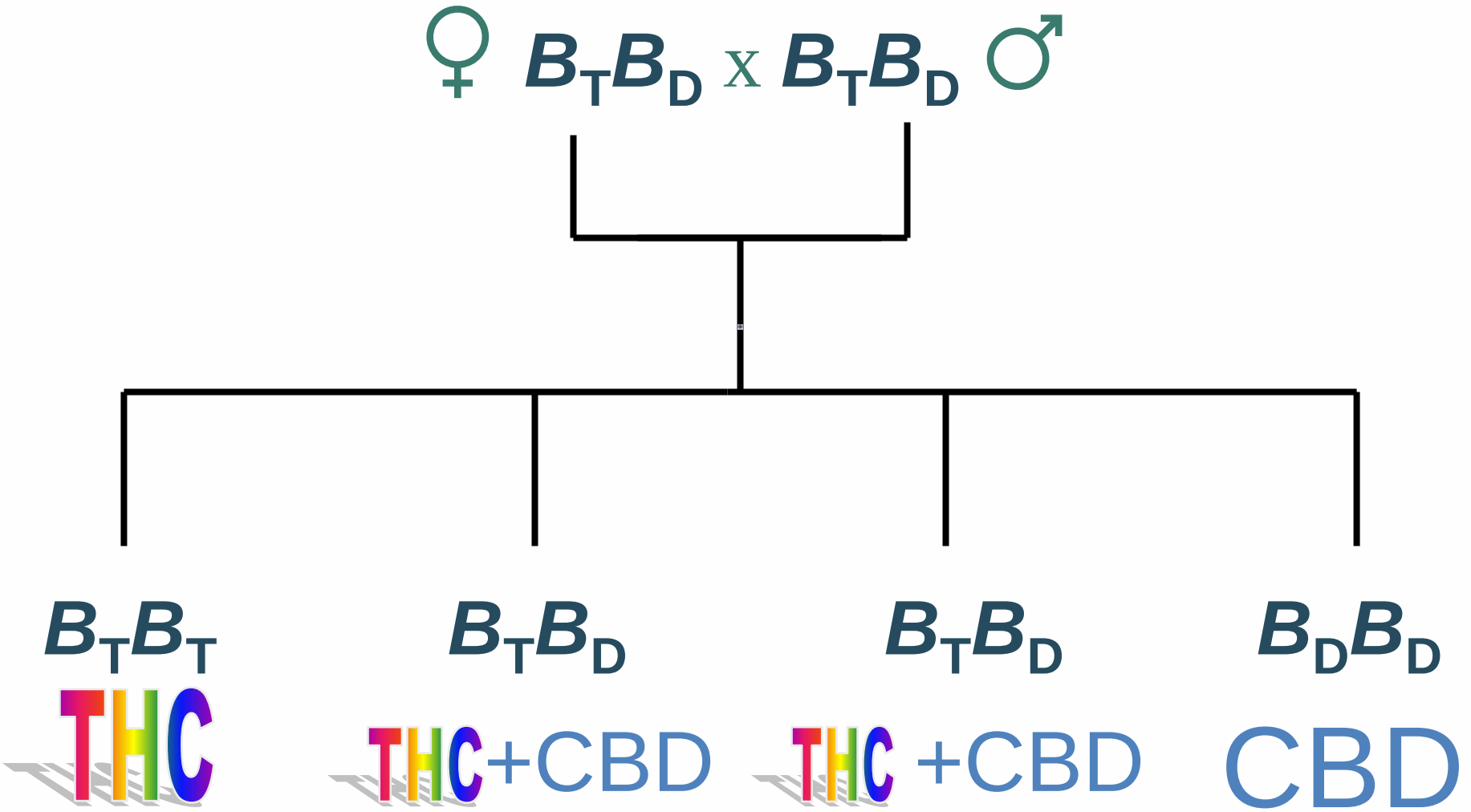
Phyto-Cannabinoid Biosynthesis
'Co-dominant Monogenic Control'



De Meijer, E. P. M. et al. (2003). Genetics **163**: 335-346.
Inheritance of Chemical Phenotype in *Cannabis sativa* L.

Phyto-Cannabinoid Biosynthesis
'Co-dominant Monogenic Control'



Homozygous THC producing B_TB_T genotypes are typically selected for recreational use

THC dominance of commercial cannabis varieties

Examples of Commercial Cannabis Seeds

52 varieties tested.

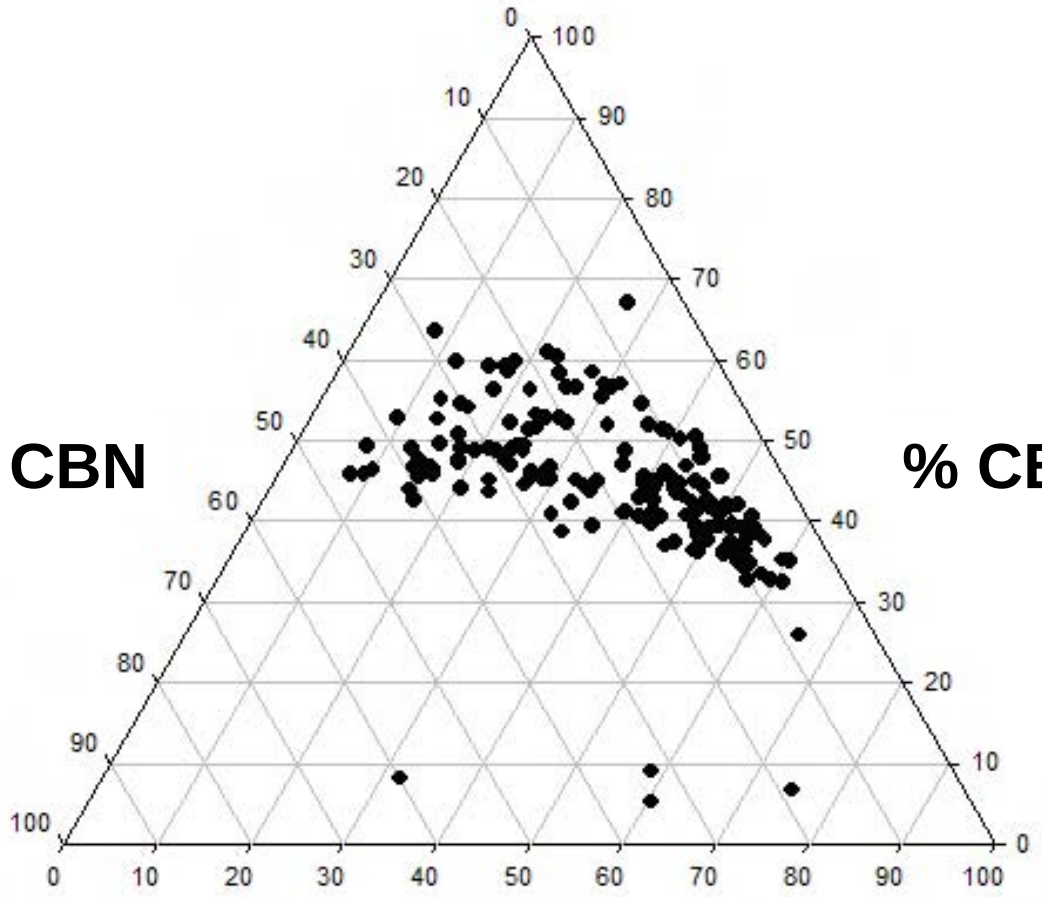
48 entirely THC dominant.

4 produced a few plants with mixed THC / CBD profile

Overall 97% of plants were THC dominant

Varying Cannabinoid Ratios in Resin 2005

Highly variable cannabinoid content and profile



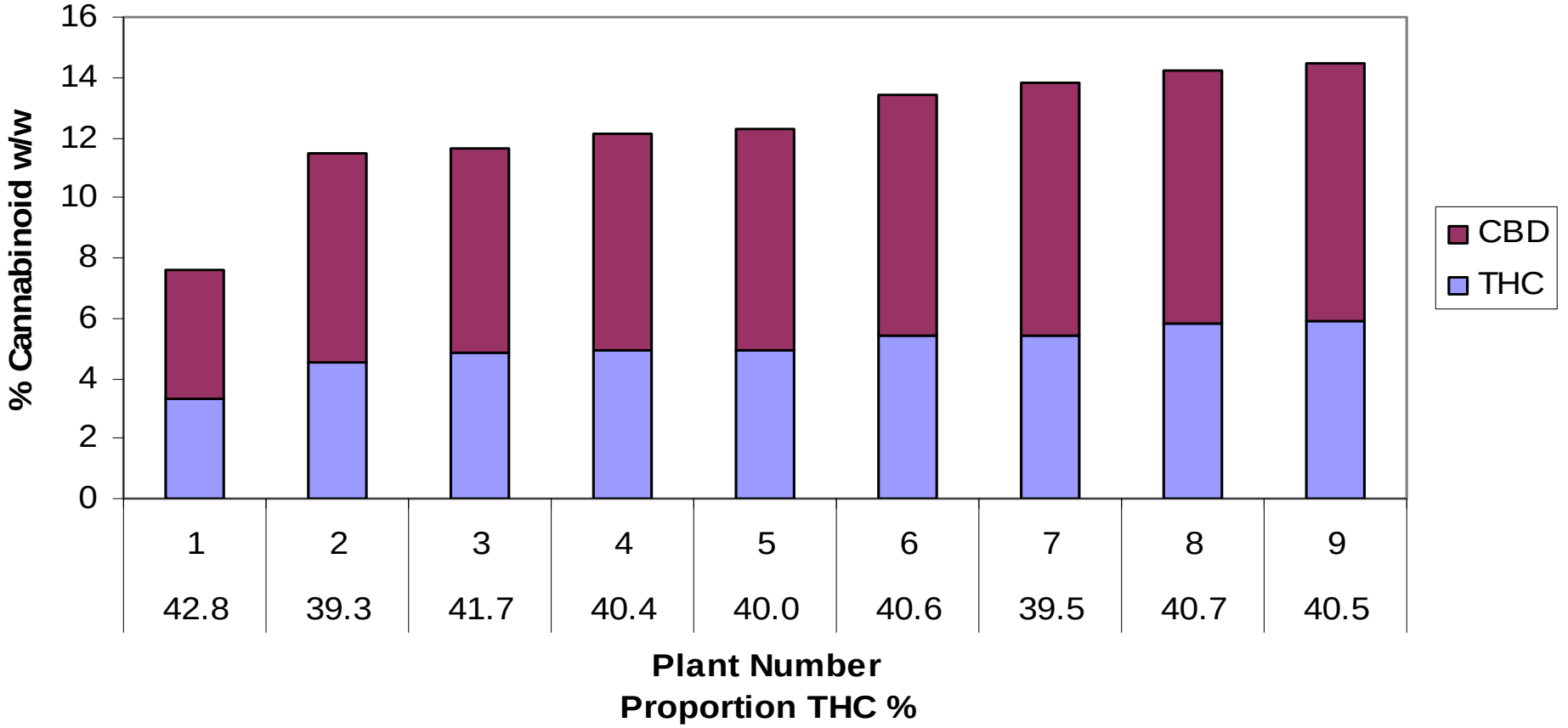
Average Content (median)

THC	**3.5%**
CBD	**4.2%**
CBN	**1.6%**

n = 169

David Potter PhD Thesis. Kings College London. 2009

Variable Cannabinoid Content and Profile of Heterozygous B_TB_D cannabis



Mature floral material was analysed from nine heterozygous plants with mixed CBD/THC profiles. Cannabinoid content and cannabinoid profile were variable

Other Sources of THC+CBD

- Bediol®

  approximately 6 % THC,
  approximately 7.5 % CBD,

  available as

  Cannabis Flos Bediol® **granulate**.

- Smoking not recommended.
- A vaporizer can be used
- Ingestion as herbal tea is also recommended.

http://www.bedrocan.nl/english/products/bediol.html

Advantages and disadvantages of indoor cannabis growing

Sativex is a ‘botanical drug’ : -

A well categorised , multi-component drug extracted from plant sources

Variations in outdoor environment might be expected to affect the balance of these multiple components

The glasshouse offers more environmental control and security.

Optimised Growth Medium and Irrigation

Minimal hand watering for 3 weeks, until roots established

Automated systems apply water through to harvest

Biological Pest Control

Parasitised Aphids

Wasp parasitising white fly

No pesticides

8 - 10 beneficial insect species regularly used

Feltiela with spider mite

Ambleseius with thrip

Mean temperature maintained at 25ºC

Year-round bright light at 50°N

May 2000

Mercury Vapour Lamps,

17 W m^{-2} PAR

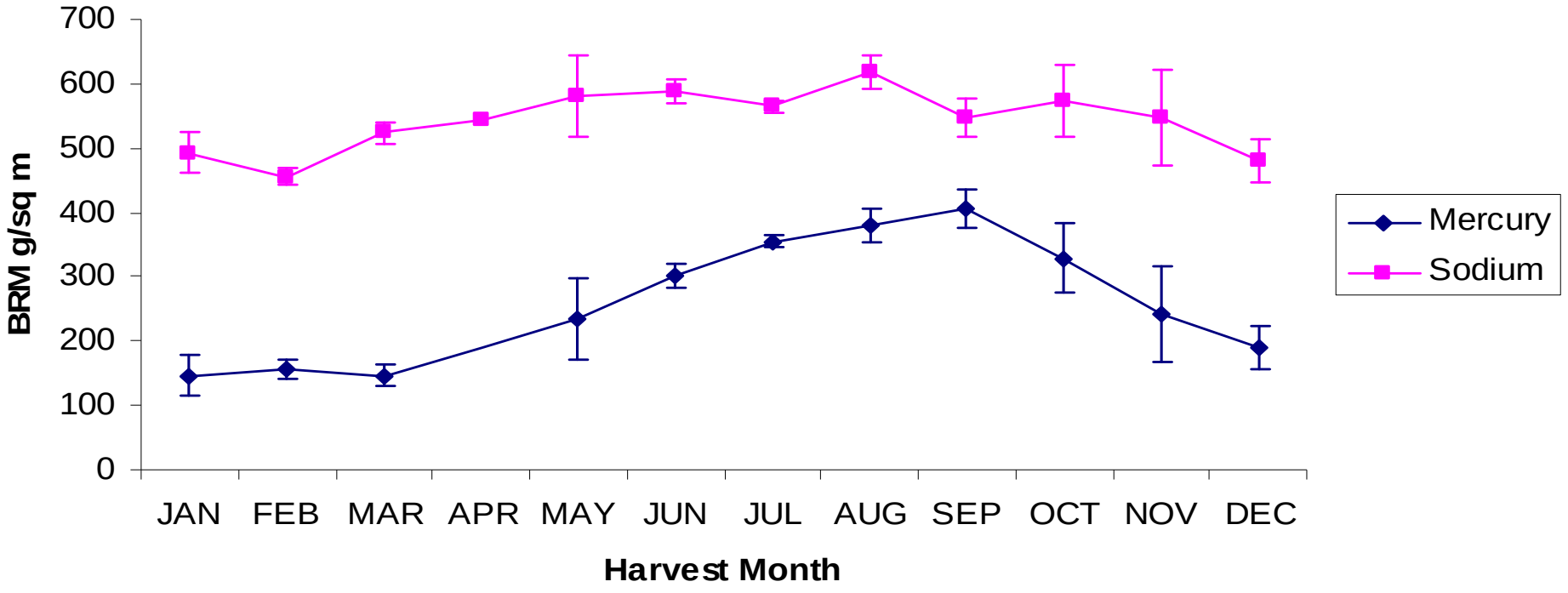
Feb 2010

High Pressure Sodium Lamps,

55 W m^{-2} PAR

Year-round bright light at 50°N

Average THC BRM yields before and after improvements (± sd)
(n ≈ 4 crops per month)



Monthly average yield, significant increase (ANOVA, $p < 0.001$)

Monthly yield uniformity, significant improvement (F-test, $p = 0.013$)

Initial vegetative phase
3 weeks of continual lighting optimum

Inflorescence development after the *critical day length*

1st week in flower

3rd week in flower

6th week in flower

8th week in flower

Flowering phase
8 Weeks of 12 hour days

Cannabis Production in Eleven Weeks

Uniform mature plants awaiting harvest

Strictly controlled growing conditions

- Genetic Stock — Clones (cuttings) of selected chemotypes
- Growth Medium — Bespoke recipe, strictly controlled
- Vegetative Growth — 3 weeks in continuous lighting
- Flowering — 8 weeks in 12 hour days
- Plant Spacing — Identical for all batches
- Temperature — Daily average 25°C (± 2°C)
- Light Intensity — Minimum 75 Wm-2 PAR

Manufacture of API - Overview

Botanical Raw Material (THC and CBD BRM)

Milling

↓

Decarboxylation

↓

CO2 Extraction

↓

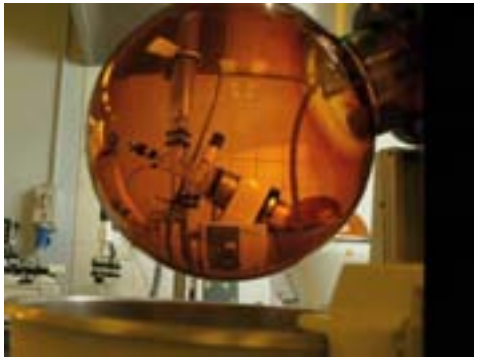
Winterisation

↓

Isolation

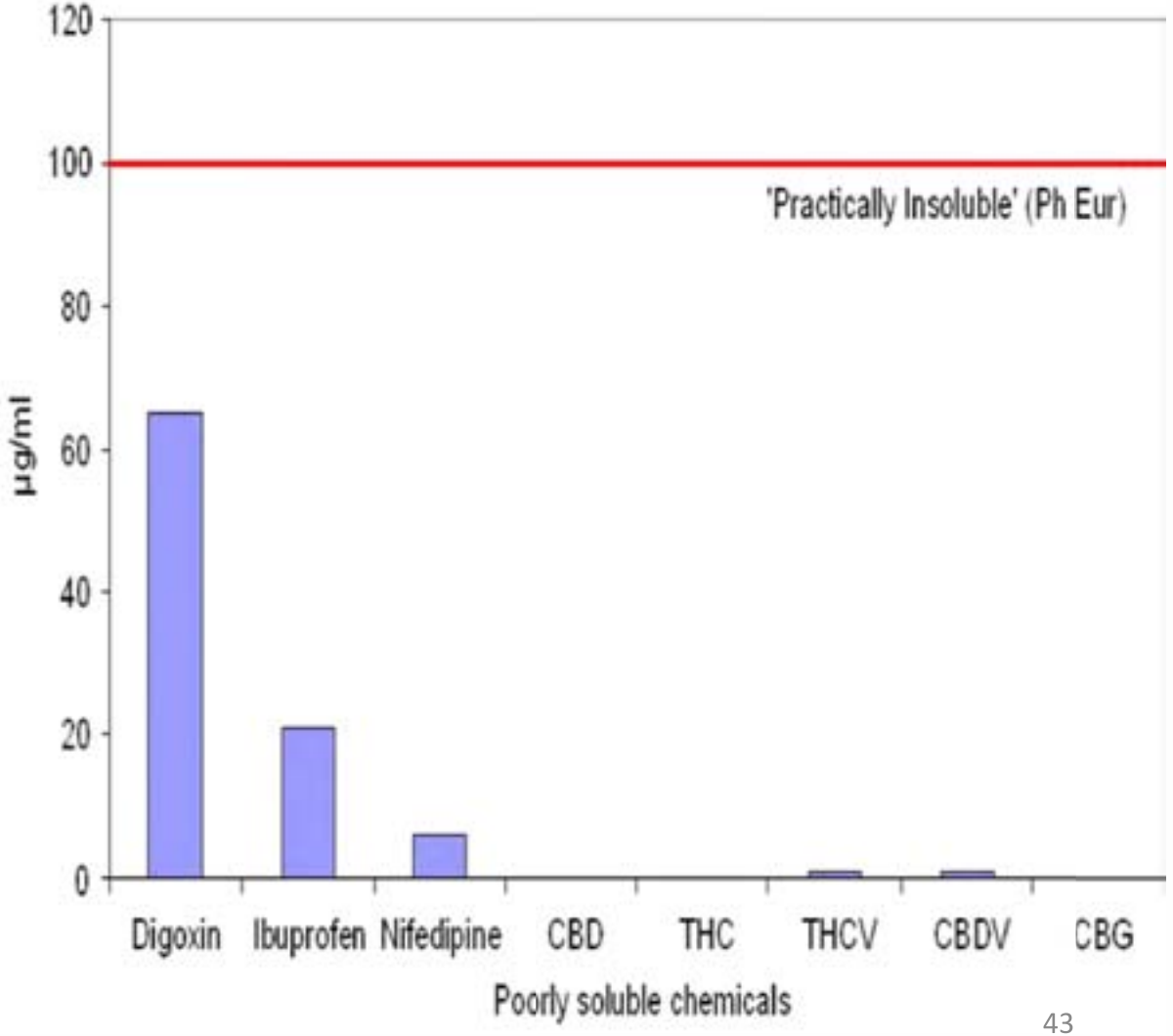
↓

API (THC and CBD BDS)



Development of Formulation - Minimal Aqueous Solubility

BDS 70 - 75% Cannabinoids



Manufacture of Sativex® - Overview



API (THC and CBD BDS) → Bulk Solution → Filling → Packaging → **Sativex Finished Product**


Sativex®

Oro-mucosal spray

(THC) Δ^9-Tetrahydrocannabinol 27mg/ml

(CBD) Cannabidiol 25mg/ml

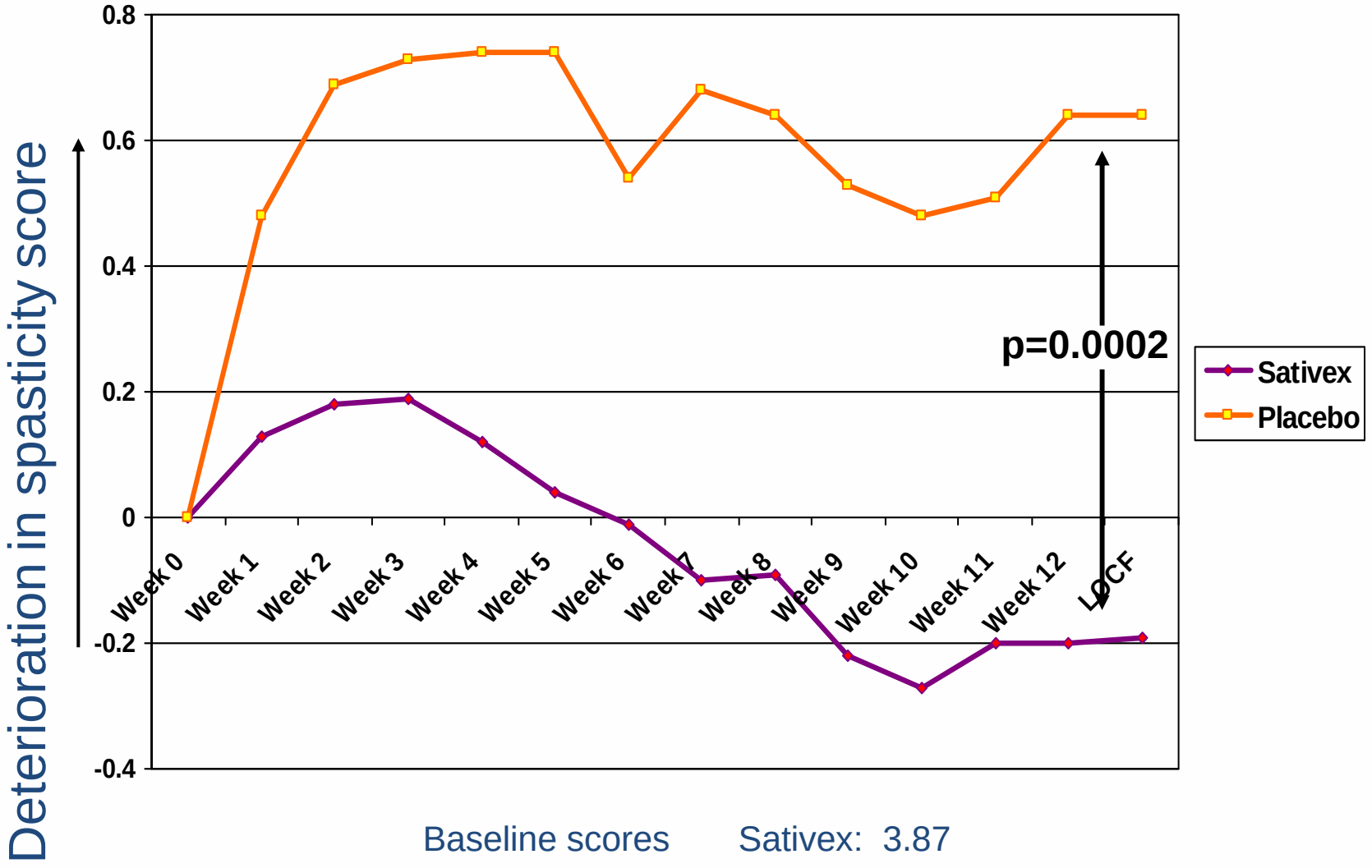
Excipients:

Ethanol Anhydrous

Propylene Glycol

Peppermint Oil

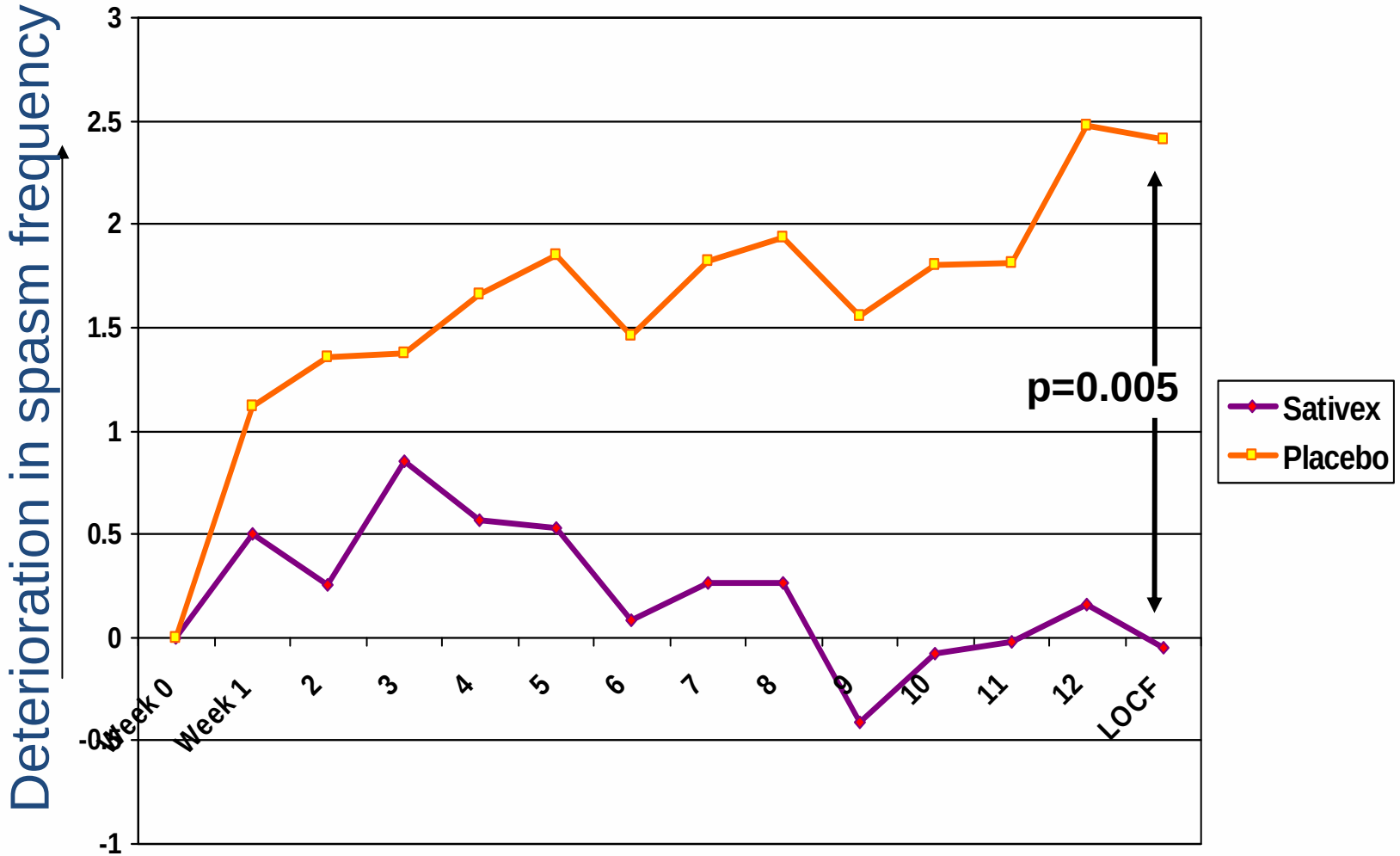
Phase III MS Spasticity Trial: Change in Spasticity scores



Baseline scores Sativex: 3.87

Placebo: 3.92

Phase III MS Spasticity Trial: Change in Spasm scores



Baseline scores: Sativex: 5.61
Placebo: 5.29

Sativex in the clinical setting

- **Approved in Canada for Neuropathic Pain in MS in mid 2005**
  - Extended to Cancer Pain Aug 07
- **UK Named Patient Programme**
  - Since 2004. 2500+ patients
- **Europe**
  - Open access throughout Spain
  - Mutual Recognition Mar 2011
  - Approval expected in Germany, Sweden, Italy, Denmark, Austria and Czech Republic.
- **Rest of the World**
  - Patients in 27 countries
- **USA**
  - Currently undergoing late stage clinical development for treatment of cancer pain

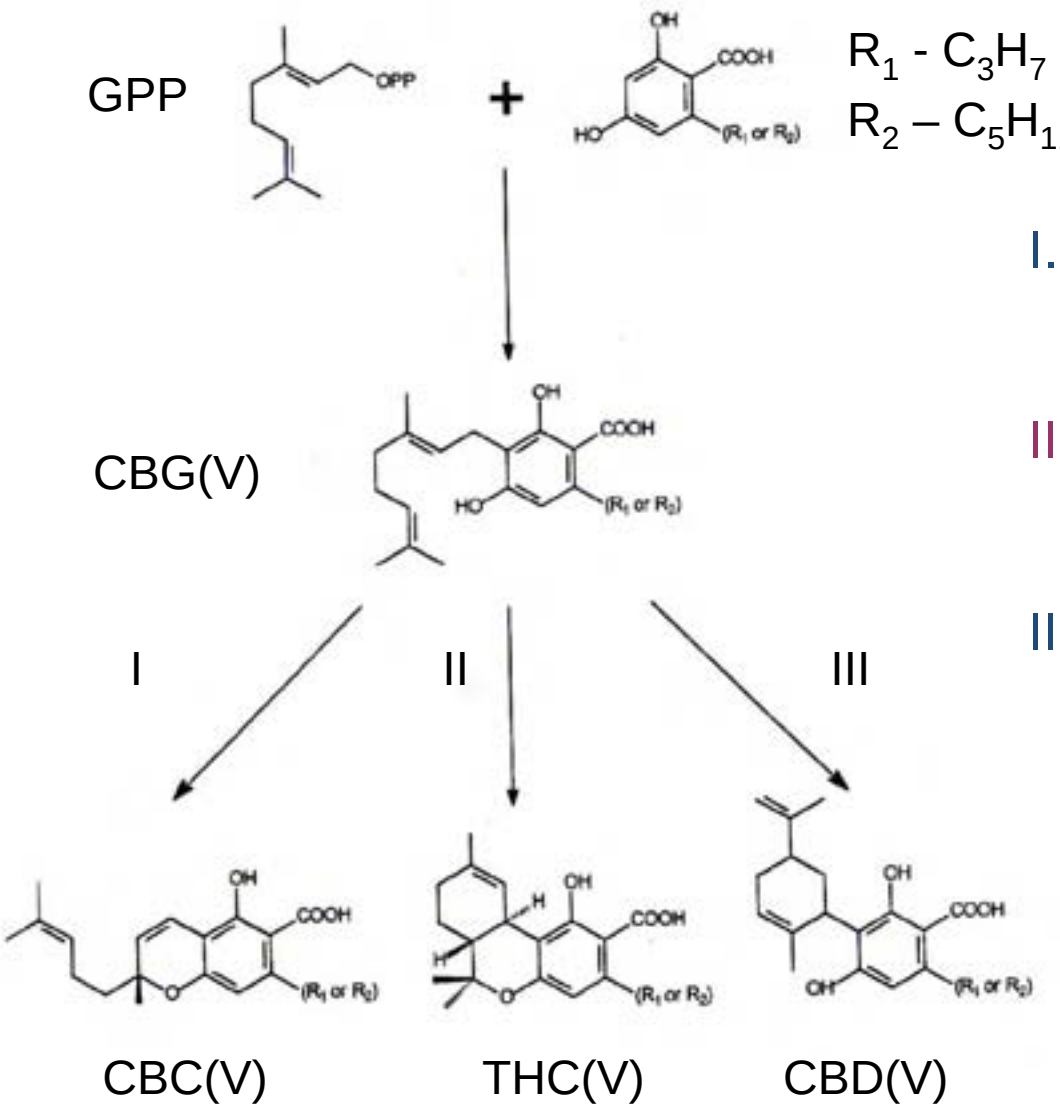
Sativex®

Approved in Canada as adjunctive analgesic in patients with advanced cancer, experiencing moderate/severe pain during highest tolerated dose of strong opioid therapy.

Cancer pain also lead target indication for approval of Sativex in the US.

Two positive Phase II trials supported advance into Phase III studies. These in latter planning stages and to be completed before regulatory submission in US and rest of the world.

Cannabis Biosynthesis



R_1 - C_3H_7 diverinic acid

R_2 – C_5H_{11} olivetolic acid

I. CBC Synthase K_m 23 µM
k_{cat} 0.04 s^{-1}

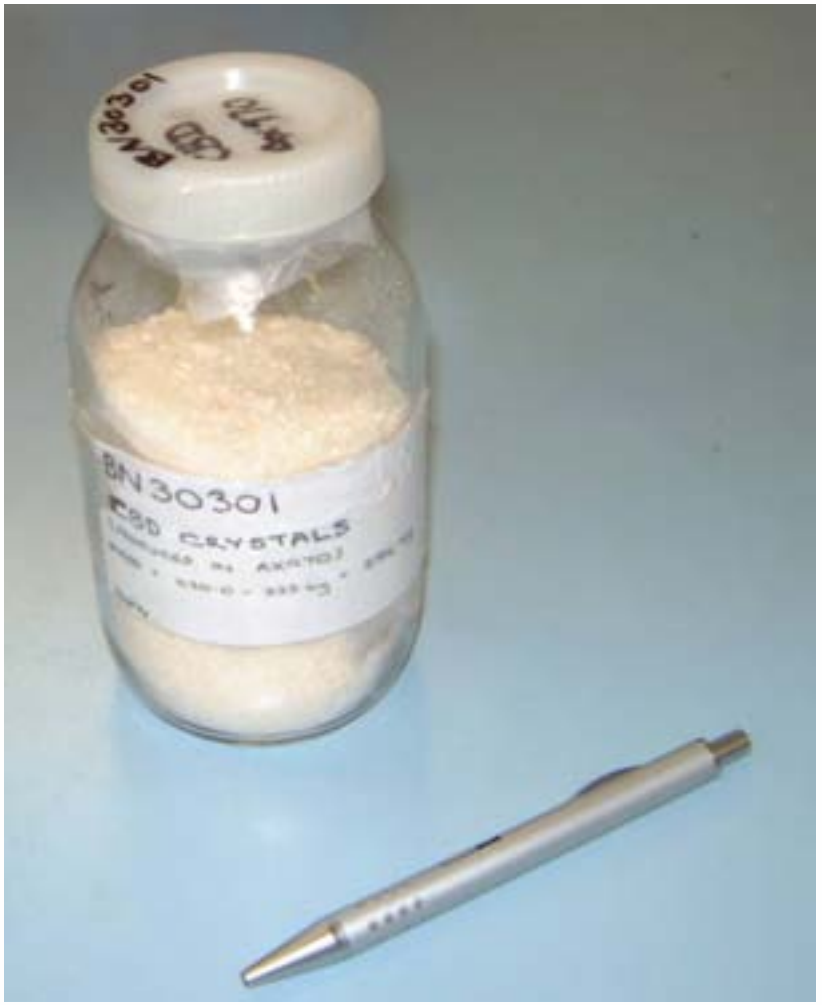
II. THC Synthase K_m 137 µM
k_{cat} 0.20 s^{-1}

III. CBD Synthase K_m 134 µM
k_{cat} 0.19 s^{-1}

Plant breeding is producing chemotypes rich in a range of cannabinoids

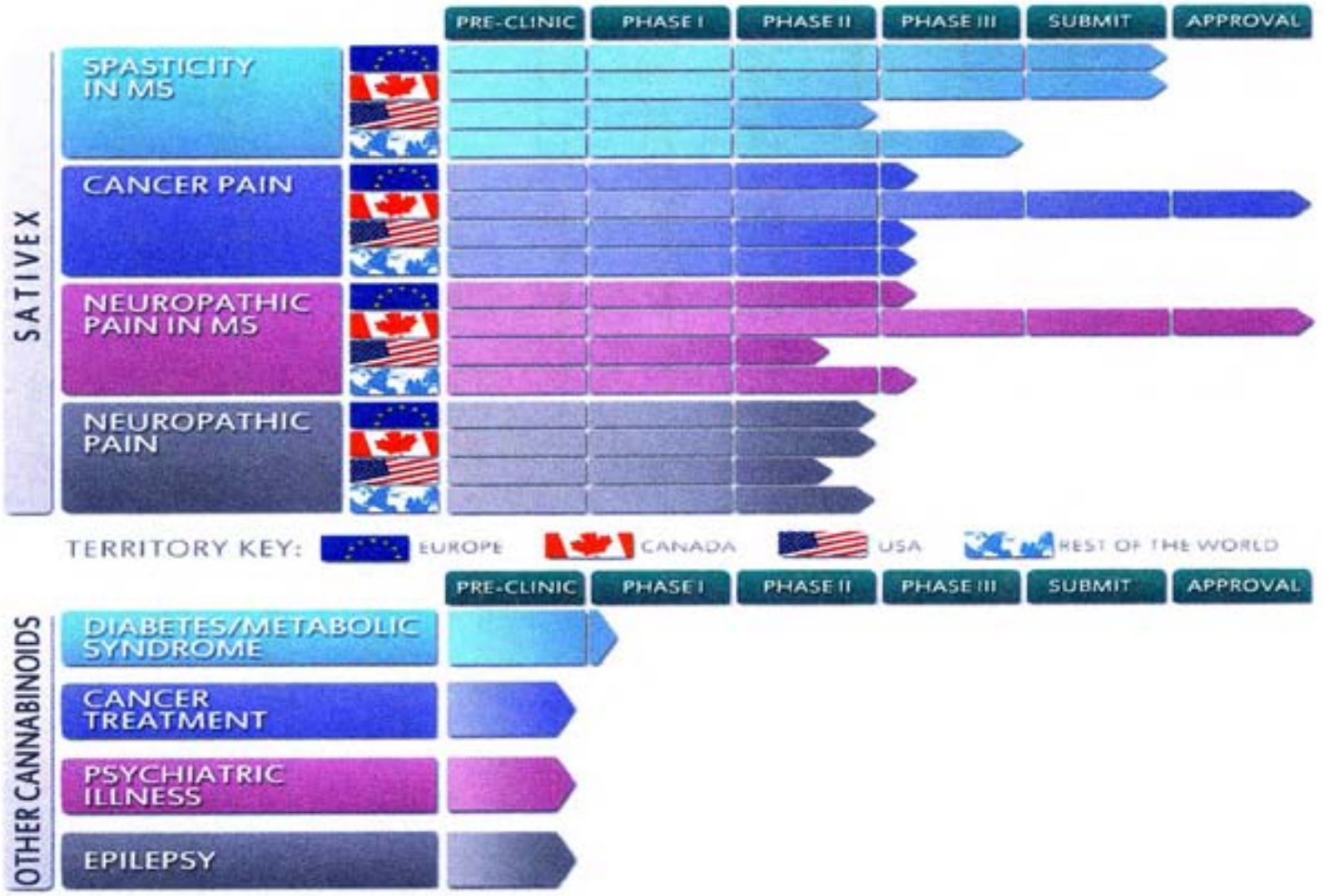
Pure Cannabinoids as NCEs

CBG



CBD

GW Pipeline March 2011



Sativex®: A valuable new medicine

Bedside Relief

BBC Panorama 2001

Cannabis-based medicine has meant a whole new outlook on life because of having a good night's sleep, pain free.

And being able to feed myself breakfast, feed myself lunch, feed myself dinner.

It makes me feel normal which is all I'm asking.