Abstract: The present invention relates to methods of treating or preventing addiction and relapse use of addictive agents, and treating or preventing addictive or compulsive behaviour and relapse practice of an addictive behaviour or compulsion, by administering a peroxisome proliferator-activated receptor gamma (PPAR) agonist, alone or in combination with another therapeutic agent, such as, for example, an opioid receptor antagonist or an antidepressant. The present invention also includes pharmaceutical compositions for treating or preventing addiction or relapse that include a PPAR agonist and one or more other therapeutic agents, as well as unit dosage forms of such pharmaceutical compositions, which contain a dosage effective in treating or preventing addiction or relapse. The methods and compositions of the invention are useful in the treatment or prevention of addiction to any agent, including alcohol, nicotine, marijuana, cocaine, and amphetamines, as well as compulsive and addictive behaviours, including pathological gambling and pathological overeating.

Title: COMPOSITIONS AND METHODS FOR PROPHYLAXIS AND TREATMENT OF ADDICTIONS
COMPOSITIONS AND METHODS FOR PROPHYLAXIS AND TREATMENT OF ADDICTIONS

CROSS-REFERENCE TO RELATED APPLICATION

This application claims the benefit under 35 U.S.C. § 119(e) of U.S. Provisional Patent Application No. 60/91 1,201 filed April 11, 2007, where this provisional application is incorporated herein by reference in its entirety.

BACKGROUND

Technical Field

The present invention is directed generally to the treatment or prevention of addictions using PPARγ agonists, alone or in combination with other therapeutic agents.

Description of the Related Art

The World Health Organization (WHO) defines substance addiction as using a substance repeatedly, despite knowing and experiencing harmful effects. Substance addiction is a chronic, relapsing disease characterized by a loss of control over drug use, compulsive drug seeking and craving for a substance, use that persists despite negative consequences, and physical and/or psychological dependence on the substance. Substance addiction typically follows a course of tolerance, withdrawal, compulsive drug taking behaviour, drug seeking behaviour, and relapse. Substance abuse and addiction are public health issues with significant social and economic impact on both the addict and society by playing a major role in violent crime and the spread of infectious diseases. Addictive substances include alcohol, caffeine, nicotine, cannabis (marijuana) and cannabis derivatives, opiates and other morphine-like opioid agonists such as heroin, phencyclidine and phencyclidine-like compounds, sedative ipnotics such as benzodiazepines and...
barbiturates and psychostimulants such as cocaine, amphetamines and amphetamine-related drugs such as dextroamphetamine and methylamphetamine.

Alcohol is one of the most commonly abused substances at a global level. Additionally, alcoholism leads to serious liver and cardiovascular disease and generates dependence resulting in severe mental disorders, social problems and adverse consequences including the division of families, tragic accidents and the reduction of work performance. According to the WHO, alcohol consumption is responsible for 20-30% of oesophageal and liver cancer, liver cirrhosis, homicides, epilepsy, and motor vehicle accidents worldwide. Globally, alcohol abuse leads to about 1.8 million deaths per year. Compulsive behaviour towards the consumption of alcohol is a core symptom of the disorder. In recent years several approaches have been investigated to help alcoholic patients to not only control alcohol drinking but also alcohol cravings and relapse (Monti et al., 1993; Volpicelli et al. 1992; O’Brien et al. 1997).

Medications such as naltrexone, acamprosate, ondansetron, disulfiram, gamma hydroxybutyrate (GHB), and topiramate tested for their potential therapeutic effect on alcohol abuse belong to several classes (Volpicelli et al. 1992; O’Brien et al. 1997). Few of these pharmacotherapeutics, such as naltrexone, acamprosate, and disulfiram, have been proven to be of a certain utility and approved for the treatment of alcoholism. Among these medications, the non-selective opioid antagonist naltrexone is currently considered the pharmacological gold standard. However, despite some promising results none of these medications, including naltrexone, is of sufficient efficacy in alcoholism and prognosis remains poor.

Nicotine is one of the most widely used addictive drugs, and nicotine abuse is the most common form of substance abuse. The WHO estimates that there are 1.25 billion smokers worldwide, representing one third of the global population over the age of 15. The WHO further estimates that 5 million deaths occur each year as a direct result of tobacco use, making nicotine abuse the largest single preventable cause of death worldwide. In industrialized countries, 70-90% of lung cancer, 56-80% of chronic respiratory disease, and 22% of cardiovascular disease
instances are attributed to nicotine addiction. Cigarette smoking is associated with 430,000 deaths a year in the US alone and is estimated to cost the nation 80 billion dollars yearly in health care costs. Tobacco use accounts for one third of all cancers, including cancer of the lung, mouth, pharynx, larynx, esophagus, cervix, kidney, ureter, and bladder. The overall rates of death from cancer are twice as high among smokers as among nonsmokers. Smoking also causes lung diseases such as chronic bronchitis and emphysema; exacerbates asthma symptoms; and increases the risk of heart disease, including stroke, heart attack, vascular disease, and aneurysm. An estimated 20% of the deaths from heart disease are attributable to smoking. Expectant women who smoke are at greater risk than nonsmokers for premature delivery, spontaneous abortion, and infants with decreased birth weight.

Nicotine use results in increased levels of the neurotransmitter dopamine, which activates the reward pathways to regulate feelings of pleasure and to mediate the desire to consume nicotine. Symptoms associated with nicotine withdrawal include craving, irritability, anger, hostility, aggression, fatigue, depression, and cognitive impairment, which lead the abuser to seek more nicotine. Environmental conditioning factors and exposure to psychological stress represent additional factors motivating nicotine use in smokers. Repeated nicotine use results in the development of tolerance, requiring higher doses of nicotine to produce the same initial stimulation.

Most therapies developed for nicotine addiction have shown only moderate success in preventing relapse, leading to a high failure rate in attempts to quit smoking. Treatments include the use of nicotine replacement products, anti-depressants, anti-hypersensitives, and behavioural therapy.

The National Institute on Drug Abuse estimates that 72 million Americans, about one third of the population, have tried marijuana. Acute effects of marijuana use include memory and learning problems, distorted perception, difficulty problem solving, loss of coordination, and increased heart rate. Long term abuse can cause the same respiratory problems observed in tobacco smokers, such as daily cough, phlegm production, increased risk of lung infections, and an increased
chance of developing cancer of the head, neck and lungs. Depression, anxiety, and job-related problems have been associated with marijuana use. Long term marijuana use can result in addiction with compulsive use that interferes with daily activities. Cravings and withdrawal symptoms, such as irritability, increased aggression, sleeplessness, and anxiety make it difficult for addicts to stop using marijuana. There are no pharmaceutical treatments available for treating marijuana addiction and relapse.

According to the WHO, an estimated 13 million people abuse opiates world wide, including 9 million heroin addicts. More than 25% of opiate abusers die from suicide, homicide, or an infectious disease, such as HIV and hepatitis, within 10-20 years of becoming addicted. Tolerance and physical dependence can develop within two to three days.

The goals for treatment of opiate addiction, as with other types of substance addictions, are to discontinue the use of the opiate while minimizing painful withdrawal symptoms and preventing relapse. Current treatments involve replacing the addictive drug with a substitution of an opioid receptor agonist or mixed agonist/antagonist. An alternative approach consists of the use of an opioid receptor antagonist to block the effect of the agonist. Antagonists provide no relief from pain or other withdrawal symptoms; rather, they can precipitate withdrawal, and their therapeutic use was associated with increased accidental opioid agonists overdosing and increased lethality. Use of agonists with a lower affinity for the receptors results in the least severe withdrawal symptoms, but it can lead to a dependence on the substitute opiate. Also, many substitution therapies take 3-6 months, allowing time for addicts to stop treatment midway.

Psychostimulants, such as cocaine and amphetamines, cause euphoria, increased alertness, and increased physical capacity in humans. These substances first increase dopamine transmission, but long term drug usage results in a reduction of dopamine activity, leading to dysregulation of the brain reward system and dysporia. The WHO estimates 33 million people around the world abuse amphetamines.
Chronic cocaine abuse can result in hyperstimulation, tachycardia, hypertension, mydriasis, muscle twitching, sleeplessness, extreme nervousness, hallucinations, paranoia, aggressive behaviour, and depression. Cocaine overdose may lead to tremors, convulsions, delirium, and death resulting from heart arrhythmias and cardiovascular failure. Desipramine, amantadine and bromocriptine have been shown to decrease cocaine withdrawal symptoms.

Amphetamine withdrawal symptoms include EEG changes, fatigue, and mental depression. Tolerance develops over time and may be associated with tachycardia, auditory and visual hallucinations, delusions, anxiety reactions, paranoid psychosis, exhaustion, confusion, memory loss, and prolonged depression with suicidal tendencies. Current treatments for amphetamine addiction include phenothiazines, haloperidol, and chlorpromazine for hallucinations, but potential side effects of these drugs include postural hypotension and severe extrapyramidal motor disorders.

In the past, treatment for substance addictions focused on behavioural therapy, but dependence on many of these highly addictive substances is hard to break. In particular, addictions to alcohol, cocaine, and heroin are considered chronic, relapsing disorders. Also, concurrent abuse of multiple substances, such as nicotine, heroin, cocaine and alcohol, is common.

The long-lasting, chronic nature of many addictions and high rates of recidivism present a considerable challenge for the treatment of drug and alcohol addiction, such that understanding of the neurobiological basis of relapse has emerged as a central issue in addiction research. Emotional and environmental factors (conditioning stimuli) were listed among the main causes of relapse. For example, it is known that specific stress conditions such as loss of work and economic difficulties, or stimuli predictive of the presence of alcohol previously associated with its use such as a bottle of the preferred wine and a bar-like environment, may strongly facilitate relapse in detoxified former alcoholics.
Two major theoretical positions exist to explain the persistence of addictive behaviour and vulnerability to relapse associated with drug and alcohol addiction, homoeostatic hypotheses and conditioning hypotheses.

Homeostatic hypotheses relate relapse risk to neuroadaptive changes and disruption of neuroendocrine homeostasis that are thought to underlie anxiety, mood dysregulation and somatic symptoms that accompany acute withdrawal, and that can persist for considerable periods of time during what has been referred to as the "protracted withdrawal" phase. This view, therefore, implicates alleviation of discomfort and negative affect as a motivational basis for relapse.

Conditioning hypotheses are based on observations that relapse is often associated with exposure to drug-related environmental stimuli. This view holds that specific environmental stimuli that have become associated with the rewarding actions of a drug by means of classical conditioning can elicit subjective states that trigger resumption of drug use. The homeostatic and conditioning hypotheses are not mutually exclusive. In fact, homeostatic and conditioning factors are likely to exert additive effects in that exposure to drug-related environmental stimuli may augment vulnerability to relapse conveyed by homeostatic disturbances.

Clearly, there is a need in the art for new methods for treating and preventing addiction and the relapse use of addictive agents. The present invention meets these needs by providing methods and pharmaceutical combinations useful in treating and preventing addiction and recidivism.

BRIEF SUMMARY

The present invention is directed generally to the use of PPARγ agonists, alone or in combination with one or more additional therapeutic agents, for the treatment and prevention of addictions and relapse to addictive use or behavior. Accordingly, the present invention provides methods and related compositions, unit dosage forms, and kits useful for the treatment and prevention of addictions, and for the treatment and prevention of relapse use of addictive agents or practice of addictive or compulsive behaviours.
In one embodiment, the present invention includes a method of treating or preventing an addiction, comprising determining that a subject has or is at risk of developing an addiction, and providing to the subject an amount of an agonist of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) effective for the treatment or prevention of the addiction.

In a related embodiment, the present invention provides a method of treating or preventing an addiction, comprising providing to a subject having an addiction a peroxisome proliferator-activated receptor gamma (PPARγ agonist) and an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of the addiction.

In certain embodiments of the methods of treating or preventing addiction of the present invention, the PPARγ agonist is a thiazolidinedione (TZD). In particular embodiments, the TZD is pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone, or darglidazone. In certain embodiments, the additional therapeutic agent is an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT₂A/₂C antagonist, or a cannabinoid-1 (CB1) receptor antagonist. In particular embodiments, the opioid antagonist is naltrexone or nalmefene. In particular embodiments, the antidepressant is fluoxetine, mirtazapine, or bupropion. In particular embodiments, the antiepileptic is topiramate, levetiracetam, or gabapentin. In one embodiment, the CRF-1 receptor antagonist is antalarmin. In another embodiment, the selective serotonin-3 (5-HT3) antagonist is ondansetron. In particular embodiments, the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant. In one embodiment, the mixed opioid agonist/antagonist is buprenorphine.

In certain embodiments of the methods of the present invention, the subject is addicted to an addictive agent, or at risk for relapse use of an addictive agent. In particular embodiments, the addictive agent is alcohol, nicotine, marijuana,
a marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, or a psychostimulant. In certain embodiments, the opioid agonist is selected from the group consisting of: morphine, methadone, fentanyl, sufentanil and heroin. In certain embodiments, the psychostimulant is cocaine, amphetamine or an amphetamine derivative. In addition, the subject may be addicted to more than one addictive agent, and the pharmaceutical compositions, unit dosage forms, and kits may be useful for treating or preventing addiction or relapse use of more than one addictive agent.

In other embodiments of the present invention, the subject is addicted to an addictive or compulsive behavior or at risk for relapse practice of an addictive or compulsive behaviour. In particular embodiments, the addictive or compulsive behavior is pathological gambling, pathological overeating, pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking. In addition, the subject may be addicted to more than one addictive or compulsive behaviour, and the pharmaceutical compositions, unit dosage forms, and kits may be useful for treating or preventing addiction or relapse use of more than one addictive or compulsive behaviour.

In particular embodiments of any of the methods of the present invention, the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist. In one embodiment, the opioid antagonist is naltrexone. In another embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In other particular embodiments of any of the methods of the present invention, the addictive agent is nicotine and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.
In further particular embodiments of any of the methods of the present invention, the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.

In other particular embodiments of any of the present invention, the subject is addicted to two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist. In certain embodiments, the opioid antagonist is naltrexone or nalmefene. In other embodiments, the mixed opioid partial agonist/antagonist is buprenorphine.

In further related embodiments, the present invention provides a method of preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour. In certain embodiments, the subject has undergone physiological withdrawal from the addictive agent during the period of abstinence from, or limited or reduced use of, the addictive agent or due to no longer being exposed to an effective amount of the anti-addiction treatment. The anti-addiction treatment may be an anti-addiction drug or may be a non-pharmacologic therapy such as counseling, psychotherapy or hypnosis therapy.

In a related embodiment, the present invention includes a method of preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour, and also providing to the subject an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective prevention of the relapse use or practice. In certain embodiments, the subject has undergone physiological withdrawal from the addictive agent during the period of abstinence from, or limited
or reduced use of, the addictive agent or due to no longer being exposed to an effective amount of the anti-addiction treatment.

In another related embodiment, the present invention provides a method of treating relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour. In certain embodiments, the subject has undergone physiological withdrawal from the addictive agent during the period of abstinence from, or limited or reduced use of, the addictive agent or due to no longer being exposed to an effective amount of the anti-addiction treatment.

In a further embodiment, the present invention includes a method of treating relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour, and also providing to the subject an additional therapeutic agent wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment of the relapse use or practice. In certain embodiments, the subject has undergone physiological withdrawal from the addictive agent during the period of abstinence from, or limited or reduced use of, the addictive agent or due to no longer being exposed to an effective amount of the anti-addiction treatment.

In another related embodiment, the present invention provides a method of preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject, wherein the subject previously reduced or eliminated use of the addictive agent or practice of the addictive or compulsive behaviour in response to treatment with an effective amount
of an anti-addiction treatment, and wherein the subject is no longer exposed to an
effective amount of the anti-addiction treatment. In certain embodiments, the subject
is no longer exposed to an effective amount of an anti-addiction agent because the
subject has become conditioned to the anti-addiction agent. In certain embodiments,
the subject is no longer exposed to an effective amount of the anti-addiction
treatment because the subject has reduced or eliminated exposure to the anti-
adiction treatment.

In a related embodiment, the present invention provides a method of
preventing relapse use of an addictive agent or practice of an addictive or
compulsive behaviour, comprising providing an effective amount of a peroxisone
proliferator-activated receptor gamma (PPARγ agonist) to a subject, wherein the
subject previously reduced or eliminated use of the addictive agent or practice of the
addictive or compulsive behaviour in response to treatment with an effective amount
of an anti-addiction treatment, and wherein the subject is no longer exposed to an
effective amount of the anti-addiction treatment, and also providing to the subject an
additional therapeutic agent, wherein each of the PPARγ agonist and the additional
therapeutic agent contribute to the effective prevention of the relapse use or practice.
In certain embodiments, the subject is no longer exposed to an effective amount
of an anti-addiction agent because the subject has become conditioned to the anti-
adiction agent. In certain embodiments, the subject is no longer exposed to an
effective amount of the anti-addiction treatment because the subject has reduced or
eliminated exposure to the anti-addiction treatment.

In additional embodiments, the present invention includes a method of
treating relapse use of an addictive agent or practice of an addictive or compulsive
behaviour, comprising providing an effective amount of a peroxisone proliferator-
activated receptor gamma (PPARγ agonist) to a subject, wherein the subject
previously reduced or eliminated use of the addictive agent or practice of the
addictive or compulsive behaviour in response to treatment with an effective amount
of an anti-addiction treatment, and wherein the subject is no longer exposed to an
effective amount of the anti-addiction treatment. In certain embodiments, the subject
is no longer exposed to an effective amount of an anti-addiction agent because the subject has become conditioned to the anti-addiction agent. In certain embodiments, the subject is no longer exposed to an effective amount of the anti-addiction treatment because the subject has reduced or eliminated exposure to the anti-addiction treatment.

In a further embodiment, the present invention includes a method of treating relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisone proliferator-activated receptor gamma (PPARγ agonist) to a subject, wherein the subject previously reduced or eliminated use of the addictive agent or practice of the addictive or compulsive behaviour in response to treatment with an effective amount of an anti-addiction treatment, and wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment, and also providing to the subject an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment of the relapse use or practice.

In certain embodiments, the subject is no longer exposed to an effective amount of an anti-addiction agent because the subject has become conditioned to the anti-addiction agent. In certain embodiments, the subject is no longer exposed to an effective amount of the anti-addiction treatment because the subject has reduced or eliminated exposure to the anti-addiction treatment.

In particular embodiments of any of the methods of treating or preventing relapse use or practice of the present invention, the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

In particular embodiments of any of the methods of treating or preventing relapse use or practice of the present invention, the relapse use or relapse practice is stress-induced.

In another embodiment, the present invention provides a method of reducing one or more symptoms associated with physiological withdrawal from an addictive agent, comprising providing an effective amount of a peroxisome prolifera-
proliferator-activated receptor gamma (PPARγ) agonist to a subject undergoing physiological withdrawal from an addictive agent.

In a related embodiment, the present invention provides a method of reducing one or more symptoms associated with physiological withdrawal from an addictive agent, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent to a subject undergoing physiological withdrawal from an addictive agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to reducing one or more symptoms associated with physical withdrawal from the addictive agent.

In particular embodiments of methods of reducing one or more symptoms associated with physiological withdrawal from an addictive agent according to the present invention, the PPARγ agonist is a thiazolidinedione (TZD). In certain embodiments, the TZD is pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone or darglidazone. In certain embodiments, the additional therapeutic agent is an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, or a cannabinoid-1 (CB1) receptor antagonist.

In another embodiment, the present invention includes a pharmaceutical composition, comprising a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of an addiction. In certain embodiments, the PPARγ agonist is a thiazolidinedione (TZD). In certain embodiments, the TZD is pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone or darglidazone.

In one embodiment, the pharmaceutical composition is effective in the treatment of an addiction to an addictive agent. In particular embodiments, the
addictive agent is alcohol, nicotine, marijuana, a marijuana derivative, an opioid
agonist, a benzodiazepine, a barbiturate, or a psychostimulant.

In another embodiment, the pharmaceutical composition is effective in
the treatment of an addiction to an addictive or compulsive behaviour. In particular
embodiments, the addictive or compulsive behavior is pathological gambling,
pathological overeating, pathological use of electronic devices, pathological use of
electronic video games, pathological use of electronic communication devices,
pathological use of cellular telephones, addiction to pornography, sex addiction,
obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent
explosive disorder, kleptomania, pyromania, trichotillomania, compulsive
overexercising, and compulsive overworking.

In certain embodiments of pharmaceutical compositions of the present
invention, the additional therapeutic agent is an opioid antagonist, a mixed opioid
partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a
corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3
(5-HT3) antagonist, a 5-HT2A/2C antagonist, and a cannabinoid-1 (CB1) receptor
antagonist. In one embodiment, the opioid antagonist is naltrexone or nalmefene. In
one embodiment, the antidepressant is fluoxetine, mirtazapine, or bupropion. In one
embodiment, the antiepileptic is selected from the group consisting of: topiramate,
levetiracetam, and gabapentin. In one embodiment, the CRF-1 receptor antagonist
is antalarmin. In one embodiment, the selective serotonin-3 (5-HT3) antagonist is
ondansetron. In one embodiment, the cannabinoid-1 (CB1) receptor antagonist is
rimonabant or tanarabant. In one embodiment, the mixed opioid agonist/antagonist
is buprenorphine.

In a particular embodiment of a pharmaceutical composition of the
present invention, the addictive agent is alcohol and the additional therapeutic agent
is an opioid antagonist or a mixed opioid antagonist/partial agonist. In one
embodiment, the opioid antagonist is naltrexone. In one embodiment, the mixed
opioid partial agonist/antagonist is buprenorphine.
In a particular embodiment of a pharmaceutical composition of the present invention, the addictive agent is nicotine and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.

In a particular embodiment of a pharmaceutical composition of the present invention, the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.

In a particular embodiment of a pharmaceutical composition of the present invention, the addictive agent comprises two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist. In one embodiment, the opioid antagonist is naltrexone or nalmefene. In one embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In a particular embodiment of a pharmaceutical composition of the present invention, the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

In further related embodiments, the present invention includes unit dosage forms of a pharmaceutical composition adapted for the treatment of an addiction, wherein said unit dosage form comprises a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent, wherein said unit dosage form comprises the PPARγ agonist and the additional therapeutic agent in a combined amount effective in the treatment of an addiction, and wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of the addiction. In particular embodiments, the PPARγ agonist is a thiazolidinedione (TZD). In certain embodiments, the TZD is pioglitazone, rosiglitazone, cigitazone, troglitazone, englitazone, rivoglitazone or darglidazone. In certain embodiments, the additional therapeutic agent is an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, and a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, and a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist.
antagonist, or a cannabinoid-1 (CB1) receptor antagonist. In one embodiment, the opioid antagonist is naltrexone or nalmefene. In one embodiment, the antidepressant is fluoxetine, mirtazapine, or bupropion. In one embodiment, the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin. In one embodiment, the CRF-1 receptor antagonist is antalarmin. In one embodiment, the selective serotonin-3 (5-HT3) antagonist is ondansetron. In one embodiment, the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant. In one embodiment, the mixed opioid agonist/antagonist is buprenorphine.

In one particular embodiment of a unit dosage form of the present invention, the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

In another related embodiment, the present invention includes a kit useful for the treatment or prevention of an addiction, comprising: a first container comprising a peroxisome proliferator-activated receptor gamma (PPARγ) agonist; and a second container comprising an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment of prevention of an addiction. In particular embodiments, the PPARγ agonist is a thiazolidinedione (TZD). In certain embodiments, the TZD is pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone or darglidazone. In certain embodiments, the additional therapeutic agent is an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, or a cannabinoid-1 (CB1) receptor antagonist. In one embodiment, the opioid antagonist is naltrexone or nalmefene. In one embodiment, the antidepressant is fluoxetine, mirtazapine, or bupropion. In one embodiment, the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin. In one embodiment, the CRF-1 receptor antagonist is antalarmin. In one embodiment, the selective serotonin-3 (5-HT3) antagonist is ondansetron. In one embodiment, the
cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant. In one embodiment, the mixed opioid agonist/antagonist is buprenorphine.

In one particular embodiment of a kit of the present invention, the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

In one particular embodiment of a kit of the present invention, the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist. In one embodiment, the opioid antagonist is naltrexone. In one embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In one particular embodiment of a kit of the present invention, the addictive agent is nicotine and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.

In one particular embodiment of a kit of the present invention, the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant. In one embodiment, the antidepressant is bupropion.

In one particular embodiment of a kit of the present invention, the addictive agent comprises two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist. In one embodiment, the opioid antagonist is naltrexone or nalmefene. In one embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In a further embodiment, the present invention includes a kit comprising one or more unit dosage forms of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and one or more unit dosage forms of nicotine. In one embodiment, the one or more unit dosage forms of nicotine comprise two or more different amounts of nicotine. In one embodiment, the PPARγ agonist is a thiazolidinedione (TZD). In one embodiment, the TZD is pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone or darglidazone.

In additional embodiment, the present invention includes a method of preventing a subject from becoming addicted, or reducing the likelihood that a subject will become addicted, to an addictive therapeutic agent, comprising providing
to a subject in need thereof an addictive therapeutic agent, and an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist), wherein the effective amount of the PPARγ agonist is an amount effective in preventing the subject from becoming addicted, or reducing the likelihood that the subject will become addicted, to the addictive therapeutic agent. In particular embodiment, this method further comprises providing to the subject an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to preventing the subject from becoming addicted, or reducing the likelihood that the subject will become addicted, to the addictive therapeutic agent. In one embodiment, the addictive therapeutic agent is an opioid agonist.

In certain embodiments of the methods of the present invention, the subject is addicted to an addictive agent, or at risk for relapse use of an addictive agent. In various embodiments, the pharmaceutical compositions, unit dosage forms, and kits of the present invention are useful for the treatment or prevention of addiction to an addictive agent or relapse use of an addictive agent. In particular embodiments, the addictive agent is alcohol, nicotine, marijuana, a marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, or a psychostimulant. In certain embodiments, the opioid agonist is selected from the group consisting of: morphine, methadone, fentanyl, sufentanil and heroin. In certain embodiments, the psychostimulant is cocaine, amphetamine or an amphetamine derivative. In addition, the subject may be addicted to more than one addictive agent, and the pharmaceutical compositions, unit dosage forms, and kits may be useful for treating or preventing addiction or relapse use of more than one addictive agent.

In other embodiments of the present invention, the subject is addicted to an addictive or compulsive behavior or at risk for relapse practice of an addictive or compulsive behaviour. In various embodiments, the pharmaceutical compositions, unit dosage forms, and kits of the present invention are useful for the treatment or prevention of addiction to an addictive or compulsive behaviour or relapse use of an addictive or compulsive behaviour. In particular embodiments, the addictive or compulsive behavior is pathological gambling, pathological overeating,
pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking. In addition, the subject may be addicted to more than one addictive or compulsive behaviour, and the pharmaceutical compositions, unit dosage forms, and kits may be useful for treating or preventing addiction or relapse use of more than one addictive or compulsive behaviour.

BRIEF DESCRIPTION OF THE SEVERAL VIEWS OF THE DRAWINGS

Figure 1 is a graph depicting the effect of acute administration of 10.0 or 30.0 mg/kg of pioglitazone (Pio 10 and Pio 30, respectively) on alcohol intake in Marchigian Sardinian alcohol-preferring (msP) rats. Controls were treated with vehicle only (Veh). Values represent the mean ± sem of alcohol intake. Significant difference from controls is indicated: * p<0.05.

Figure 2 is a graph depicting the effect of acute administration of 0.25 mg/kg of naltrexone (Ntx) alone or in combination with 10.0 or 30.0 mg/kg of pioglitazone (Pio 10 and Pio 30, respectively) on alcohol intake in msP rats. Controls were treated with drugs' vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake. Significant difference from controls is indicated: ** p< 0.01 and * p<0.05.

Figures 3A-3D are graphs demonstrating the effect of subchronic administration of 10.0 or 30.0 mg/kg of pioglitazone (Pio 10 and Pio 30, respectively) on alcohol intake in msP rats. Controls were treated with drug vehicle (Veh). The values shown in Figures 3A-3C represent the mean ± sem of daily alcohol intake measured at: 2 hours (Figure 3A); 8 hours (Figure 3B); and 24 hours (Figure 3C) from the beginning of the dark phase of the daily light/dark cycle. Figure 3D shows daily food intake measured at 24 hour intervals. Significant difference from controls is indicated: * p<0.05.
Figures 4A-4D are graphs demonstrating the effect of subchronic administration of 0.25 mg/kg of naltrexone (Ntx) alone or in combination with 10.0 or 30.0 mg/kg of pioglitazone (Pio 10 and Pio 30, respectively) on alcohol intake in msP rats. Controls were treated with drug vehicle (Veh + Veh). The values shown in Figures 4A-4C represent the mean ± sem of daily alcohol intake measured at: 2 hours (Figure 4A); 8 hours (Figure 4B); and 24 hours (Figure 4C) from the beginning of the dark phase of the daily light/dark cycle. Figure 4D shows daily food intake measured at 24 hour intervals. Significant difference from controls is indicated: *p<0.05.

Figure 5 is a bar graph depicting the effect of pioglitazone on yohimbine-induced reinstatement of ethanol seeking behaviour. Compared to extinction (Ext), yohimbine elicited a significant reinstatement of responding that was markedly reduced by pre-treatment with 10.0 and 30.0 mg/kg of pioglitazone (Pio 10 and 30, respectively). Values represent the mean (±SEM) number of responses at the active lever. Significant difference from controls (pioglitazone vehicle; Veh) is indicated: **P<0.05.

Figure 6 is a bar graph depicting the lack of effect of pioglitazone on cue-induced reinstatement of ethanol seeking behaviour. Values shown represent the mean (±SEM) number of responses at the active or inactive levers.

Conditioning: responses of the last 10% alcohol (filled circle) and water (open circle) session of the discrimination phase. Extinction (Ext): responses during the last day of this phase. Reinstatement: responses in rats exposed to stimuli predictive of alcohol (S+/CS+) or water (S7CS−) availability. Significant difference from Ext is indicated: **P<0.01.

Figure 7 is a graph depicting the effect of treatment with ciglitazone 5.0 (Cig 5) or 20.0 mg/kg (Cig 20) or its vehicle (Veh) on FR1 ethanol self-administration in Wistar rats. Each lever response resulted in the delivery of 0.1 ml of 10% ethanol. Significant difference from controls (Veh) is indicated: *P<0.05.

Figure 8 is a graph the effect of administration of 7.5 or 15.0 mg/kg of rosiglitazone (Ros) on alcohol intake in msP rats. Controls were treated with the
drug vehicle (Veh). Values represent the mean ± sem of alcohol intake (g/kg) at the indicated time points. Significant difference from controls is indicated: ‘‘<0.01 and ‘p<0.05.

Figures 9A and 9B are graphs depicting the effect of pre-treatment with the PPARγ antagonist GW9662 on pioglitazone induced reduction of ethanol drinking. Figure 9A represents the effect of GW9662 (GW) given alone (1.0 and 5.0 mg/kg) on ethanol intake in msP rats. Figure 9B describes the effect of pre-treatment with GW9662 on animals injected with 30 mg/kg pioglitazone (Pio) or its vehicle. The control group received vehicles of both drugs (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: ‘‘p<0.01.

Figure 10 is a graph depicting the effect of pre-treatment with the PPARγ antagonist GW9662 given ICV on pioglitazone-induced reduction of ethanol drinking. MsP rats received 5.0 µg/rat of GW9662 (GW) alone, 30 mg/kg of pioglitazone (Pio) alone or their combination. Control group received vehicles of both drugs (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: ‘p<0.05 and ‘‘p<0.01.

Figure 11 is a graph depicting the effect of naltrexone (Ntx) on yohimbine-induced reinstatement of alcohol seeking. Compared to extinction (Ext) yohimbine elicited a significant reinstatement of responding that was not modified by pre-treatment with 0.25 and 1.0 mg/kg of naltrexone. Values represent the mean (±SEM) number of responses at the active lever. Difference from controls (0.0) was not significant.

Figure 12 is a graph depicting the effect of naltrexone (Ntx) on cue-induced reinstatement of alcohol seeking. Values represent the mean (±SEM) number of responses at the active lever. Conditioning: responses of the last 10% alcohol (filled circle) and water (open circle) session of the discrimination phase. Extinction (Ext): responses during the last day of this phase. Reinstatement responses in rats exposed to stimuli predictive of alcohol (S7CS +) or water (S7CS −)
availability. Treatment with 0.25 and 1.0 mg/kg of naltrexone significantly reduced cue-induced reinstatement of alcohol-seeking; **p < 0.01.

Figures 13A and 13B are graphs depicting the effect of naltrexone (ntx) plus pioglitazone (Pio) combination on: yohimbine-induced reinstatement of alcohol seeking (Figure 13A) or cue-induced reinstatement of alcohol seeking (Figure 13B). Compared to extinction (Ext), yohimbine elicited a significant reinstatement of responding. The combination of Naltrexone (1.0 mg/kg) plus pioglitazone (10 and 30 mg/kg) significantly inhibited yohimbine-induced reinstatement of alcohol seeking (Figure 13A). Treatment with 1.0 mg/kg of naltrexone in combination with Pioglitazone (10.0 and 30.0 mg/kg) also significantly reduced cue-induced reinstatement of alcohol-seeking. Conditioning: responses of the last 10% alcohol (filled circle) and water (open circle) session of the discrimination phase. Extinction (Ext): responses during the last day of this phase. Reinstatement responses in rats exposed to stimuli predictive of alcohol (S+/CS+) or water (S7CS−) availability. Values represent the mean (±SEM) number of responses at the active lever. Significant difference from Ext is indicated: *P < 0.05, **p < 0.01.

Figure 14 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 3 mg/kg of fluoxetine alone or their combination on alcohol intake in msP rats. Controls were treated with the drug vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: *p < 0.05 and **p < 0.01.

Figure 15 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 5 mg/kg of mirtazapine alone or their combination on alcohol intake in msP rats. Controls were treated with the drug vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: *p < 0.05.

Figure 16 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 30 mg/kg of topiramate alone or their combination on alcohol intake in msP rats. Controls were treated with the drug vehicles (Veh+Veh).
Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: *p<0.05.

Figure 17 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 100 mg/kg of levetiracetam (Leve) alone or their combination on alcohol intake in msP rats. Controls were treated with vehicles alone (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: *p<0.05 and **p<0.01 .

Figure 18 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 30 mg/kg of gabapentin alone or their combination on alcohol intake in msP rats. Controls were treated with vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: **p<0.01 and *p<0.05.

Figure 19 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 1.0 mg/kg of ondansetron alone or their combination on alcohol intake in msP rats. Controls were treated with the drug vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: **p<0.01 and *p<0.05.

Figure 20 is a graph depicting the effect of administration of 10 mg/kg of pioglitazone (Pio) alone or 15 mg/kg of antalarmin alone or their combination on alcohol intake in msP rats. Controls were treated with vehicles (Veh+Veh). Values represent the mean ± sem of alcohol intake (g/kg). Significant difference from controls is indicated: *p<0.05 and **p<0.01 .

Figure 21 is a graph depicting the effect of administration of 10 and 30 mg/kg of pioglitazone (Pio) on an alcohol withdrawal score in Wistar rats. Controls received oral administration of alcohol vehicle. Values represent the mean ± sem of total withdrawal score. Significant difference from controls is indicated: **p<0.01 .

Figures 22A and 22B are graphs depicting the effect of treatment with 10.0 or 30.0 mg/kg pioglitazone (10 or 30, respectively) or its vehicle (veh) on FR5 cocaine self-administration in Wistar rats. Figure 22A shows the number of rewards at the active lever, with each five lever response resulting in the delivery of one
reward (0.25 mg/0.1 ml of cocaine). Figure 22B shows the number of responses at the left inactive lever. Significant difference from controls (Veh) is indicated: **p<0.01.

Figure 23A and 23B are graphs depicting the effect of treatment with pioglitazone (30.0 mg/kg) or its vehicle (veh) on FR5 nicotine self-administration in Wistar rats. Figure 23A shows the number of rewards at the active lever, with each five lever response resulting in the delivery of 0.25 mg/0.03 ml of nicotine. Figure 23B shows the number of responses at the left inactive lever. Significant difference from controls (Veh) is indicated: *p<0.05.

DETAILED DESCRIPTION

The present invention is based, in large part, upon the finding described herein that peroxisome proliferator-activated receptor gamma (PPARγ) agonists are useful in the treatment and prevention of addictions and relapse use of an addictive agent or behaviour. Accordingly, the present invention provides methods and related compositions, formulations, unit dosage forms and kits for the treatment and prevention of addiction and relapse use, which include one or more PPARγ agonists, alone or in combination with one or more additional therapeutic agents in which each of the PPARγ agonist and the additional therapeutic agent(s) contribute to the effective treatment or prevention of the addiction.

As demonstrated in the accompanying Examples, a variety of different thiazolidinediones (TZDs) were shown to reduce intake of an addictive agent in various models of addiction. For example, each of the TZDs, pioglitazone, ciglitazone, and rosiglitazone, significantly reduced ethanol consumption in rat models of alcohol addiction (Examples 1, 3, 7, and 8). The effect was evident for both acute and subchronic administration of a TZD (Examples 1 and 2). In addition, TZDs were shown to reduce cocaine use in a rat model of cocaine addiction (Example 23) and a rat model of nicotine addiction (Example 24). This effect of the PPARγ agonists was shown to be mediated by the activation of PPARγ receptors using two different PPARγ agonists (Examples 9 and 10). In addition, an
observational study of human patients using pioglitazone for the treatment of diabetes confirmed that this PPARγ agonist was efficacious in reducing ethanol abuse (Example 22). These data establish that PPARγ agonists may be used to treat and prevent addiction to a variety of different addictive agents.

In addition, the accompanying Examples demonstrate that PPARγ agonists used in combination with a variety of different therapeutic agents substantially reduced intake of an addictive agent. For example, it is shown that acute or subchronic treatment with the TZD, pioglitazone, enhanced the inhibitor action of the opioid antagonist, naltrexone, on ethanol intake (Examples 2 and 4). These data demonstrate that the use of a PPARγ agonist in combination with an opioid antagonist would have increased, e.g., additive or synergistic, efficacy in treating or preventing addiction.

In addition to reducing use of an addictive agent, PPARγ agonists were also able to reduce or prevent relapse use, or reinstatement, of addictive agents. As described in Example 5, treatment with pioglitazone significantly reduced stress-induced reinstatement of alcohol use. Interestingly, however, it did not significantly reduce cue-induced reinstatement of alcohol use (Example 6). In contrast, the opioid antagonist, naltrexone, reduced cue-induced reinstatement of alcohol use, but not stress-induced reinstatement of alcohol use (Examples 12 and 11). The data support the concept that the combination of a PPARγ agonist and an opioid antagonist would have an enhanced ability to prevent relapse use of an addictive agent, since such a combination would prevent both stress-induced and cue-induced relapse use. In fact, treatment with the combination of the PPARγ agonist, pioglitazone, and the opioid antagonist, naltrexone, resulted in a significantly reduced reinstatement of both stress-induced and cue-induced alcohol use (Example 13).

PPARγ agonists also worked synergistically with other classes of therapeutic agents in reducing or preventing addiction and relapse use. For example, the TZD, pioglitazone, used in combination with a variety of different classes of antidepressants, including fluoxetine and mirtazapine, worked
synergistically in reducing ethanol consumption in an animal model of ethanol addiction (Examples 14 and 15). Anti-epileptics, including topiramate, levetiracetam, and gabapentin, showed synergism in combination with a TZD in reducing ethanol intake (Examples 16-18), and antiemetics, including the serotonin-3 (5-HT3) receptor selective antagonist, ondansetron, and the corticotrophin releasing factor 1 receptor selective antagonist, antalarmin, also showed synergism in combination with a TZD in reducing alcohol consumption (Examples 19 and 20).

Interestingly, the experiments described in the accompanying Examples also showed that PPARγ agonists significantly reduced withdrawal symptoms in alcohol-addicted animals (Example 21).

In summary, the present invention demonstrates that treatment with PPARγ agonists represent a novel pharmacological approach for the treatment and prevention of addiction, since it reduces addictive agent consumption and recidivism associated to stress exposure.

In addition, considering that the physiopathology of addiction has features (i.e., drug craving, compulsive behaviour triggered by drug desire, withdrawal, relapse behaviour, neurological damages, cognitive impairment) common to all drugs of abuse it is reasonable to think that PPARγ agonists will be useful also for the treatment of dependence to other addictive agents or behaviours, including, e.g., opiates (morphine, heroin, methadone), psychostimulants (cocaine, methamphetamine, and amphetamine related compounds in general), nicotine, gamma hydroxybutyrate (GHB), phencyclidine, and phencyclidine derivatives, etc.

PPARγ agonists showed efficacy also in combination with opioid antagonists; co-administration of the two drugs resulted in additivity with regard to the effect on ethanol drinking and expanded the efficacy of the opioid antagonist on stress induced reinstatement. In a co-administration regimen, it is of particular significance to note the neuroprotective anticonvulsant and withdrawal-reducing effect of TDZs, especially during the early treatment phase. In fact, opioid antagonists do not result in any amelioration of withdrawal symptoms and this, in general, may
contribute to the early treatment drop out and low compliance often reported with these drugs.

The ability of TDZs to normalize hepatic function may also have positive consequences in exploitation of a combined treatment approach. In fact, the clinical condition of alcoholic patients is, in general, compromised, especially during the early detoxification phase. Thus, rapid recovery and amelioration from a pathological state could improve treatment retention.

A. Methods of Treating and Preventing Addictions Using PPARγ Agonist(s)

Thus, the present invention includes methods of treating or preventing an addiction, comprising providing one or more PPARγ agonists to a subject having an addiction or at risk for developing an addiction. In various embodiments, the subject is addicted to an addictive agent or behaviour, including, but not limited to, any of the addictive agents and behaviours described herein. The subject may be physically or physiologically dependent on the substance or behaviour; the subject may be psychologically dependent; or the subject may be both physically and psychologically dependent. The subject may be addicted to one or more than one addictive agent or behaviour.

As used herein, unless the context makes clear otherwise, "treat," and similar word such as "treatment," "treating" etc., is an approach for obtaining beneficial or desired results, including and preferably clinical results. Treatment can involve optionally either the reducing or amelioration of a disease or condition, (e.g., addiction, relapse use, withdrawal), or the delaying of the progression of the disease or condition (e.g., addiction relapse use, withdrawal).

As used herein, unless the context makes clear otherwise, "prevent," and similar word such as "prevention," "preventing" etc., is an approach for preventing the onset or recurrence of a disease or condition, (e.g., addiction, relapse use, withdrawal) or preventing the occurrence or recurrence of the symptoms of a disease or condition, or optionally an approach for delaying the onset or recurrence
of a disease or condition or delaying the occurrence or recurrence of the symptoms of a disease or condition.

Generally, a subject is provided with an effective amount of a PPARγ agonist. As used herein, an "effective amount" or a "therapeutically effective amount" of a substance, e.g., a PPARγ agonist, is that amount sufficient to affect a desired biological or psychological effect, such as beneficial results, including clinical results. For example, in the context of treating addiction using the methods of the present invention, an effective amount of a PPARγ agonist is that amount sufficient to cause the subject to reduce or discontinue use of an addictive agent.

According to certain embodiments of the present invention, a subject is provided with a PPARγ agonist alone, while in other embodiments, a subject is provided with a PPARγ agonist in combination with an additional therapeutic agent. It is understood that the effective amount of either or both of a PPARγ agonist and an additional therapeutic agent may be different when either is provided alone than when provided in combination. For example, when the PPARγ agonist and the additional therapeutic agent act synergistically, then a lower amount of the PPARγ agonist, a lower amount of the additional therapeutic agent, or lower amounts of both the PPARγ agonist or the additional therapeutic agent may be required to achieve the same therapeutic effect that would be provided by either the PPARγ agonist or the additional therapeutic agent alone. In other embodiments, the same amount of the PPARγ agonist and the additional therapeutic agent are used to provide an enhanced therapeutic effect relative to the therapeutic effect provided by either the PPARγ agonist or the additional therapeutic agent alone. As another example, data in the Examples below indicate that patients addicted to alcohol and treated with the PPARγ agonist pioglitazone exhibit decreased depression, and treatment of addicted patients with a combination of a PPARγ agonist and an antidepressant agent in accordance with the present invention may provide an enhanced antidepressive therapeutic effect as part of the treatment of the addictive disorder.

The subject may be any animal, including a mammal, and, particularly, a human.
In one aspect of the invention, the subject is first determined or diagnosed to have an addiction, or to be at risk of developing an addiction, by diagnostic testing, observation or analysis by a medical care provider. An effective amount of a PPARγ agonist, or an effective amount of a PPARγ agonist and one additional therapeutic agent, are then provided to the subject for treatment or prevention of the addiction. In another aspect of the invention, the subject is first determined or diagnosed to have an addiction, or to be at risk of developing an addiction, by diagnostic testing, observation or analysis by a medical care provider, but the subject has not been diagnosed or determined to have diabetes or other insulin disorder. An effective amount of a PPARγ agonist, or an effective amount of a PPARγ agonist and one additional therapeutic agent, are then provided to the subject for treatment or prevention of the addiction. The dosage of the PPARγ agonist, or the PPARγ agonist and the one additional therapeutic agent, may be specifically determined by the medical practitioner for treatment or prevention of the addiction rather than for any other disorder or disease.

In particular aspects, the subject is provided with a PPARγ agonist, alone or in combination with an additional therapeutic agent for the primary purpose of treating or preventing an addiction. In related aspects of the methods of the present invention, the subject has not previously been provided with a PPARγ agonist for the treatment or prevention of any disease or disorder other than an addiction. In particular, in certain embodiments, the subject has not previously been provided with a PPARγ agonist for the treatment of insulin resistance or diabetes. In a further related embodiment, the subject has not been diagnosed with insulin resistance or diabetes.

In various embodiments of the present invention, the subject may be provided with any PPARγ agonist, including any of the specific PPARγ agonists described below. In particular embodiments, the PPARγ agonist is a TZD, including any of the TZDs described below. In certain embodiments, the TZD is pioglitazone, ciglitazone, rosiglitazone or trogalitazone.
In particular embodiments, the subject is suffering from or at risk for addiction to any physically addictive agent or addictive or compulsive behaviour, including, e.g., any of those described below. In particular embodiments, the subject is addicted to alcohol, cocaine, nicotine, marijuana, an opiate or other opioid agonist or methamphetamine or other psychostimulant, or phencyclidine and phencyclidine derivatives.

In particular embodiments, a subject is considered at risk of addiction or relapse to use of an addictive agent or practice of an addictive behaviour when the subject has previously been addicted to the same or a different addictive agent or addictive or compulsive behaviour. In certain embodiment, the subject is considered at risk of addiction or relapse to use of an addictive agent or practice of an addictive behaviour when the subject is psychologically addicted to an addictive agent or addictive or compulsive behaviour, even if the subject is no longer physically addicted.

In certain embodiments, the subject is addicted to or at risk of becoming addicted to a therapeutic agent provided to the patient to treat a disease or disorder, e.g., a pain medication. In a related embodiment, the subject may be at risk of abusing an addictive therapeutic agent, such as a pain medication. Abusing an addictive therapeutic agent, in certain embodiment, is understood to indicate using the agent for a reason different than or in addition to its prescribed use. In such a situation, a subject may be provided with both an addictive therapeutic agent and a PPARγ agonist, alone or in combination with an additional therapeutic agent. For example, a subject suffering from pain, or at risk of pain, may be provided with an opioid agonist and a PPARγ agonist or TZD, e.g., pioglitazone, to both provide analgesia and prevent or treat addiction to the opioid agonist. Because PPARγ agonists have been shown to reduce neuropatic pain and inflammatory responses (see, e.g., Oliveira A. et al., Antinociceptive and antiedematogenic activities of fenofibrate, an agonist of PPAR alpha, and pioglitazone, an agonist of PPAR gamma, *Eur J Pharmacol*. 561(1-3):194-201 (2007)), the PPARγ agonist may add to or enhance the analgesic affect of the opioid agonist.
In various embodiments, the subject is provided with the PPARγ agonist at the same time that the subject is using an addictive agent, after the subject has discontinued use of an addictive agent, or before the subject begins using an addictive agent.

1. Addictive Agents

The term addiction is used to describe a recurring compulsion by an individual to engage in some specific activity, despite harmful consequences to the individual's health, mental state or social life. The term is often reserved for drug addictions, but it is sometimes applied to other compulsions, such as problem gambling, and compulsive overeating. Factors that have been suggested as causes of addiction include genetic, biological/pharmacological and social factors.

The medical community now makes a careful theoretical distinction between physical or physiological dependence (characterized by symptoms of withdrawal) and psychological dependence (sometimes referred to simply as addiction). Addiction is now narrowly defined as "uncontrolled, compulsive use." If there is no harm being suffered by, or damage done to, the patient or another party, then clinically it may be considered compulsive, but to the definition of some it is not categorized as "addiction". In practice, the two kinds of addiction (physiological dependence and psychological dependence) are not always easy to distinguish. Addictions often have both physical and psychological components.

Physical dependence (or drug dependence) refers to a state resulting from habitual use of a drug, where negative physical withdrawal symptoms result from abrupt discontinuation. Examples of addictive agents for which a user may develop a physical dependence include nicotine, opioids, barbiturates, benzodiazepines, alcohol, i.e., ethyl alcohol, GHB, and methaqualone.

Commonly abused stimulants such as cocaine or amphetamine class drugs are not believed to cause significant physical dependence. However, their potential for extreme physiological addiction can compel the user to consume
amounts which become physically damaging, but life-threatening withdrawal effects have not been observed.

As used herein, addictive agents includes any and all agents to which a subject can become addicted, either physically or psychologically, or both. As noted above, addiction includes addiction to chemical entities, such as drugs, e.g., ethyl alcohol, nicotine, or cocaine, as well as addiction to other behaviours, e.g., pathological gambling, pathological overeating, pathological use of electronic devices, e.g., BlackBerry®, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking.

Addictive agents include addictive recreational drugs, as well as addictive medications. Examples of addictive agents include, but are not limited to, alcohol, e.g., ethyl alcohol, gamma hydroxybutyrate (GHB), caffeine, nicotine, cannabis (marijuana) and cannabis derivatives, opiates and other morphine-like opioid agonists such as heroin, phencyclidine and phencyclidine-like compounds, sedative hypnotics such as benzodiazepines, methaqualone, mecloqualone, etoqualone and barbiturates and psychostimulants such as cocaine, amphetamines and amphetamine-related drugs such as dextroamphetamine and methylamphetamine. Other examples include LSD, psilocybin, extasy and other hallucinogens. Examples of addictive medications include, e.g., benzodiazepines, barbiturates, and pain medications including alfentanil, allylprodine, alphaprodine, anileridine benzylmorphine, bezitramide, buprenorphine, butorphanol, clonitazene, codeine, cyclazocine, desomorphine, dextromoramide, dezocine, diampromide, dihydrocodeine, dihydromorphone, dimenoxadol, dimephtanol, dimethyethylthiambutene, dioxaphetyl butyrate, dipipanone, eptazocine, ethoheptazine, ethylmethylthiambutene, ethylmorphine, etonitazene fentanyl, heroin, hydrocodone, hydromorphone, hydroxyphenazocine, isomethadone, ketobemidone, levallophan, levorphanol, levophencyclidine, lofenantil, meperidine, meptazinol, metazocine,
methadone, metopon, morphine, myrophine, nalbuphine, narceine, nicomorphine, norlevorphanol, normethadone, nalorphine, normorphine, norpipanone, opium, oxycodone, OXYCONTIN®, oxymorphone, papaveretum, pentazocine, phenadoxone, phenomorphan, phenazocine, phenoperidine, piminoine, piritramide, propheptazine, promedol, properidine, propiram, propanoxyphene sufentanil, tramadol, tilidine, salts thereof, mixtures of any of the foregoing, mixed µ-agonists/antagonists, and the like.

In certain embodiments, a subject may be addicted to an opioid agonist. The terms "opioid agonist," "opioid" and "opiate" are used interchangeably herein and are used to designate a group of drugs that are, to varying degrees, opium- or morphine-like in their properties. Their main use is for pain relief. These agents work by binding to opioid receptors, which are found principally in the central nervous system and the gastrointestinal tract. Opiates are also addictive agents. Opiates include alfentanil, allylprodine, alpaphrodine, anileridine, apomorphine, benzylmorphine, beta-hydroxy 3-methylfentanyl, bezitramide, carfentanil, clonitazene, codeine, desomorphine, dextromoramide, diacetylmorphine (heroin), diapramiprole, dihydrocodeine, dihydroetorphine, dihydromorphone, dimenoxadol, dimepheptanol, dimethylthiambutene, dioxaphetylbutyrate, dipipanone, etazocine, ethoheptazine, ethylmethylthiambutene, ethylmorphine, etonitazene, etorphine, fentanyl, hydrocodone, hydromorphone, hydroxypethidine, isomethadone, ketobemidone, LMM, levorphanol, levophenacymorph, lofenatin, meperidine, metapone, metazocine, methadone, methadyl acetate, metopon, morphine, myrophine, narceine, nicomorphine, norlevorphanol, normethadone, normorphine, norpipanone, opium, oxycodone, oxymorphone, papaverine, phenadoxone, phenomorphan, phenoperidine, piminoine, piritramide, propheptazine, promedol, properidine, propoxyphene, remifentanil, sufentanil, thebaine, tilda, and tramadol.

Naturally occurring opiates include codeine, morphine, noscapine, papaverine, and thebaine. Semi-synthetic opioids include diacetylmorphine, hydrocodone, hydromorphone, levorphanol, metapone, nalorphine, naloxone, naltrexone, oxycodone, oxymorphone, and tramadol. Synthetic opioids include
ethoheptazine, fentanyl, levorphanol, meperidine, methadone, phenazocine, propoxyphene and sufentanil.

Three broad classifications of opiates are phenanthrenes, phenylheptylamines, and phenylpiperidines. Examples of phenanthrenes include codeine, etorpine, hydrocodone, hydromorphone, morphine, oxycodone, and oxymorphone. Examples of phenylheptylamines include dimeheptanol, dimenoxadol, dipipanone, isomethadone, methadone, methadyl acetate, and propoxyphene. Examples of phenylpiperidines include alfentanyl, alphaprodine, beta-promedol, carfentanyl, fentanyl, lofentanil, meperidine, properidine, and sufentanil.

Specific psychostimulants include, by way of example, amphetamine, cocaine, dextroamphetamine, methamphetamine, pemoline, and methylenedioxymethamphetamine.

While a subject may be addicted to a single addictive agent or behaviour, frequently subject is addicted to two or more addictive agents or behaviours. Addiction to two or more addictive agents or addictive behaviours is referred to as polyaddiction.

2. PPARγ Agonists

Peroxisome proliferator-activated receptors (PPARs) are ligand-activated transcription factors of the nuclear hormone receptor superfamily. At present three distinct PPAR isoforms, namely PPARα, PPARβ/δ and PPARγ, have been identified (Breidert et al., 2002; Feinstein et al. 2003). The PPARα receptor isoform is highly expressed in the liver and kidney and it regulates fatty acid catabolism; the PPARβ/δ is ubiquitously expressed and is involved in the regulation of different cellular processes including adipocytes, keratinocytes and oligodendrocytes differentiation. Finally, PPARγ receptors are predominantly expressed in adipose tissue and macrophages, where they are involved in adipocyte differentiation, regulation of sugar and lipid homeostasis and control of inflammatory
The endogenous ligands of PPAR receptors belong to various classes of unsaturated fatty acid compounds that include leukotrienes, retinoic acid metabolites and prostaglandins. For example, the PPARγ receptor is mainly located in the cytoplasmatic fraction and is activated by the 15-deossi-Δ12-14-prostaglandin J2 (Burstein 2005; Cernuda-Morollon, et al., 2002).

Recent studies have also shown that, in addition to various peripheral tissues, PPARβ/δ and PPARγ receptors are expressed in neurons, and oligodendrocytes (but not in astrocytes) of the central nervous system (CNS). The exact role of these receptors in the brain is not well understood yet (Kainu et al. 1994).

It is known that activation of PPARγ mediates neuroprotective responses against excitotoxic process and inflammatory damages (Butcher et al. 2002). Activation of these receptors is also associated with improvement of cognitive performances, and has protective potential against epileptic insults (Yu et al. 2008).

In 1997 a new class of drugs, the thiazolidinediones (TZDs), was developed in Japan, originally as anti-oxidants. Certain of these compounds were then approved for the clinical treatment of insulin resistance and type 2 diabetes.

At the molecular level, TDZs bind with high affinity and activate PPARγ receptors; this has been proposed as the major mechanism through which these molecules exert their therapeutic effects. At present, two TDZ compounds are used clinically to treat humans, pioglitazone (Actos®) and rosiglitazone (Avandia®). Pioglitazone and methods for synthesizing and formulating pioglitazone and pioglitazone compositions are further described in US Patents 4,687,777, 5,965,584 and 6,150,383, the disclosure of each of which is hereby incorporated by reference. Other compounds (i.e., ciglitazone, troglitazone, aleglitazar, muraglitazar, tesaglitazar, and ragaglitazar, etc.) are under development. Suitable PPARγ agonists for use in the present invention include selective PPARγ agonists such as
ciglitazone, troglitazone, pioglitazone, rosiglitazone, englitazone, rivoglitazone and
darglidazone.

An additional class of PPARγ agonists are the dual-acting PPAR agonists. Dual-acting PPAR agonists are a novel group of compounds that activate nuclear transcription factors. By activating both PPARα and PPARγ receptors, they simultaneously reduce atherogenic triglycerides, raise cardioprotective HDI levels, and improve insulin resistance. Examples of dual-acting PPAR agonists that may be suitable for use in the present invention include tesaglitazar, aleglitazar, muraglitazar, and ragaglitazar.

Additional PPARγ agonists that may be used according to the present invention include, but are not limited to, those described in the following patents and patent applications: U.S. Patent Nos. 6,294,580, 7,067,530, 6,582,738, 6,794,154, 4,812,570, 4,775,687, 4,725,610, 4,582,839, and 4,572,912; and U.S. Patent Application Publication Nos. US2002/006942, US2007/0299047, US2004/0077525, and US2008/0045580, the disclosures of which are hereby incorporated by reference. Examples of dual PPARγ agonists that may be used according to the present invention include, e.g., those described in U.S. Patent Application Nos. 2007/037882, US2006/0270722, US2006/0211749, US2006/0167045, and US2005/0014833, the disclosures of which are hereby incorporated by reference.

B. Methods of Treating and Preventing Addiction Using PPARγ Agonist(s) in Combination with Other Therapeutic Agents

As demonstrated in the accompanying Examples, PPARγ agonists may be effectively used in combination with one or more additional therapeutic agents to treat or prevent addiction, including addiction to one or more of the addictive agents described infra and compulsive or addictive behaviour. Accordingly, the present invention includes methods of treating or preventing an addiction, comprising providing to a subject addicted to an addictive agent one or more PPARγ agonist(s) and one or more additional therapeutic agent(s), in which each of the PPARγ agonist(s) and the additional therapeutic agent(s) contribute to the effective
treatment or prevention of the addiction. In one embodiment, a subject is provided with or administered one PPARγ agonist and one additional therapeutic agent. In another embodiment, a subject is addicted to two or more addictive agents. As demonstrated by the Examples below, the combination of a PPARγ agonist and another therapeutic agent may have advantageous additive or synergistic efficacy in treating or preventing addiction or relapse use of an addictive agent. In some embodiments, the additional agent is another anti-addiction agent.

The PPARγ agonist and the additional therapeutic agent may be administered at the same time (i.e., concurrently), or either may be administered before the other (i.e., sequentially). In general, both the PPARγ agonist and the additional therapeutic agent are present in the subject at the same time for a duration of time and at levels sufficient to provide a therapeutic benefit to the subject, i.e., in the treatment or preventing of an addiction or the prevention of a relapse use (or reinstatement) of an addictive agent or compulsive or addictive behaviour. The PPARγ agonist and the additional therapeutic agent may be administered by the same or different routes of administration. Typically, the PPARγ agonist and the additional therapeutic agent are each provided to a subject according to a standard route of administration of a commercially available or other pharmaceutical composition. In one embodiment, the PPARγ agonist and the additional therapeutic agent are co-administered using a composition comprising both agents.

The additional therapeutic agent provided in combination with a PPARγ agonist may be any therapeutic agent that contributes to an aspect of the effective treatment or prevention of the addiction. For example, the additional therapeutic agent may be a drug used to treat an addiction or a drug used to alleviate side-effects associated with physiological withdrawal from an addictive agent. In addition, the additional therapeutic agent may be any drug that affects brain serotonin neurotransmission, such as selective serotonin reuptake inhibitors (SSRIs), and tricyclic and tetracyclic serotonin and norepinephrine reuptake inhibitors (SNRIs) as described below, and serotonin agonists such as sumatriptan, ergonovine, dihydroergotamine and buspirone. In certain embodiments, the additional
therapeutic agent is an opioid antagonist, including mixed opioid partial agonist/antagonists, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist such as mianserin, mirtazapine and ketanserin, or a cannabinoid-1 (CB1) receptor antagonist, including but not limited to those therapeutic agents specifically described infra.

In one embodiment, the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist. In a particular embodiment, the opioid antagonist is naltrexone. In another embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In one embodiment, the addictive agent is alcohol, and the additional therapeutic agent is topiramate or levetiracetam.

In one embodiment, the addictive agent is nicotine and the additional therapeutic agent is an antidepressant. In a particular embodiment, the antidepressant is bupropion.

In one embodiment, the addictive agent is cocaine, and the additional therapeutic agent is buprenorphine.

In one embodiment, the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant. In a particular embodiment, the antidepressant is bupropion.

In one embodiment, the subject is addicted to two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist. In a particular embodiment, the mixed opioid partial agonist/antagonist is buprenorphine.

In particular embodiments, a subject is provided with a combination of: pioglitazone and naltrexone; ciglitazone and naltrexone; rosiglitazone and naltrexone; englitazone and naltrexone; rivoglitazone and naltrexone; darglidazone and naltrexone; pioglitazone and fluoxetine; ciglitazone and fluoxetine; rosiglitazone and fluoxetine; englitazone and fluoxetine; rivoglitazone and fluoxetine; darglidazone and fluoxetine; pioglitazone and mirtazapine; ciglitazone
and mirtazapine; rosiglitazone and mirtazapine; englitazone and mirtazapine; rivoglitazone and mirtazapine; darglidazone and mirtazapine; pioglitazone and topiramate; ciglitazone and topiramate; rosiglitazone and topiramate; englitazone and topiramate; rivoglitazone and topiramate; darglidazone and topiramate; pioglitazone and levetiracetam; ciglitazone and levetiracetam; rosiglitazone and levetiracetam; englitazone and levetiracetam; rivoglitazone and levetiracetam; darglidazone and levetiracetam; pioglitazone and gabapentin; ciglitazone and gabapentin; rosiglitazone and gabapentin; englitazone and gabapentin; rivoglitazone and gabapentin; darglidazone and gabapentin; piolitazone and ondansetron; ciglitazone and ondansetron; rosiglitazone and ondansetron; englitazone and ondansetron; rivoglitazone and ondansetron; darglidazone and ondansetron; pioglitazone and antalarmin; ciglitazone and antalarmin; rosiglitazone and antalarmin; englitazone and antalarmin; rivoglitazone and antalarmin; darglidazone and antalarmin.

For treatment of alcohol addiction, combinations to be administered in accordance with the present invention include a PPARγ agonist and an opioid agonist or a mixed opioid antagonist/partial antagonist, a PPARγ agonist and an antidepressant, a PPARγ agonist and a CB1 receptor antagonist/inverse agonist, a PPARγ agonist and varenicicline, a PPARγ agonist and acamprosate, and a PPARγ agonist and disulfiram.

For treatment of a psychostimulant addiction, combinations to be administered in accordance with the present invention include, e.g., a PPARγ agonist and an antidepressant or a PPARγ agonist and a partial opioid agonist/antagonist, e.g., buprenorphine.

For treatment of nicotine addiction, combinations to be administered in accordance with the present invention include, e.g., a PPARγ agonist and an antidepressant, a PPARγ agonist and nicotine (as a replacement, in an oral, transcutaneous or other conventional formulation), a PPARγ agonist and an opioid antagonist, a PPARγ agonist and a CB1 receptor antagonist/inverse agonist, and a PPARγ agonist and varenicicline.
For treatment of polysubstance addiction, combinations to be administered in accordance with the present invention include, e.g., a PPAR\(\gamma\) agonist and an opioid agonist or a mixed opioid antagonist/partial antagonist.

For treatment of gambling addiction, combinations to be administered in accordance with the present invention include, e.g., a PPAR\(\gamma\) agonist and an antidepressant or a PPAR\(\gamma\) agonist and an agent affecting dopamine neurotransmission, e.g., a direct or indirect dopamine antagonist.

The effective amount of either or both of a PPAR\(\gamma\) agonist and an additional therapeutic agent may be reduced when administered in combination that when either is provided alone. For example, when the PPAR\(\gamma\) agonist and the additional therapeutic agent act additively or synergistically, then a lower amount of the PPAR\(\gamma\) agonist, a lower amount of the additional therapeutic agent, or lower amounts of both the PPAR\(\gamma\) agonist or the additional therapeutic agent may be required to achieve the same therapeutic effect that would be provided by either the PPAR\(\gamma\) agonist or the additional therapeutic agent alone.

a. Opioid Antagonists

An opioid antagonist acts on one or more opioid receptors. At least three types of opioid receptors, mu, kappa, and delta opioid receptors, have been reported, and opioid antagonists are generally classified by their effects on the opioid receptors. Opioid antagonists may antagonize central receptors, peripheral receptors or both. Naloxone and naltrexone are commonly used opioid antagonist drugs that are competitive that bind to the opioid receptors with higher affinity than agonists, but that do not activate the receptors. This effectively blocks the receptor, preventing the body from responding to opiates and endorphins.

Many opioid antagonists are not pure antagonists but also produce some weak opioid partial agonist effects, and can produce analgesic effects when administered in high doses to opioid-naive individuals. Examples of such compounds include nalorphine, and levallorphan. However, the analgesic effects from these drugs are limited and tend to be accompanied by dysphoria, most likely
due to action at the kappa opioid receptor. Since they induce opioid withdrawal
effects in people who are taking, or have previously used, opioid full agonists, these
drugs are considered to be antagonists.

Naloxone is one example of an opioid antagonist that has no partial
agonist effects. Instead, it is a weak inverse agonist at mu opioid receptors, and is
used for treating opioid overdose.

Specific examples of opioid antagonists that may be used according to
the invention include alvimopan, binaltorphimine, buprenorphine, cyclazocine,
cyclorphan, cypridime, dinicotinate, beta-funaltrexamine, levallorphan,
methylnaltrexone, nalbuphine, nalide, nalmefene, nalmexone, nalorphine, nalorphine
dinicotinate, naloxone, naloxonazine, naltrendol, naltrexone, naltrindole, oxilorphan,
and pentazocine.

b. Antidepressents

Antidepressents are drugs used to treat depression. The three
neurotransmitters believed to be involved in depression are serotonin, dopamine,
and norepinephrine. Certain types of antidepressants increase the levels of one or
more of these neurotransmitters in the brain by blocking their reabsorption.

Several different classes of antidepressants have been identified,
including selective serotonin reuptake inhibitors (SSRIs), tricyclic and tetracyclic
serotonin and norepinephrine reuptake inhibitors (SNRIs), norepinephrine reuptake
inhibitors (NRIs), norepinephrine and dopamine reuptake inhibitors (NDRIs),
azaspiroines, monoamine oxidase inhibitors (MAOIs), and atypical antidepressants.

SSRIs include, e.g., cericlamine, citalopram, clomipramine,
cyanothiepin, dapoxetine, duloxetine, escitalopram, femoxetine, fluoxetine,
fluvoxamine, ifoxetine, imipramine, indalpine, indeloxazine, litoxetine, lofepramine,
mianserine, milnacipran, mirtazapine, nefazadone, nortriptyline, paroxetine,
sertaline, sibutramine, tomoxetine, trazodone, venlafaxine, and zimeldine.

Amitriptyline, amoxapine, butriptyline, clomipramine, demexiptilnine,
desipramine, dibenzepin, dimetacrine, dothiepin, doxepin, imipramine, iprindole,
lofepramine, maprotiline, melitracen, metapramine, mianserin, mirtazpine, nortriptyline, propizepine, protriptyline, quinupramine, setiptiline, tianeptine, and trimipramine are all tricyclic and tetracyclic antidepressants.

SNRIs include, *e.g.*, amoxapine, atomoxetine, biciadine, desipramine, desvenlafaxine, duloxetine, maprotiline, milnacipran, nefazodone, reboxetine, sibutramine, and venlafaxine.

Nisoxetine, nortriptyline, reboxetine, talsupram, and tomoxetine are all examples of NRIs.

NDRIs include, *e.g.*, bupropion, hydroxybupropion, and tesofensine. Azaspirones include, *e.g.*, buspirone, gepirone, ipsapirone, tandospirone, and tiaspirone. Buspirone is an anxiolytic (partial agonist at 5-HT1 autoreceptors) that may be provided with an anti-depressant such as an SSRI.

Specific MAOIs include, *e.g.*, amiflamine, brofaromine, clorgyline, alpha-ethyltryptamine, iprocluzide, iproniazid, isocarboxazid, mebanazine, moclobemide, nialamide, pargyline, phenelzine, pheniprazine, pirlindole, safrazine, selegiline, toloxatone, and trancypromine.

Atypical antidepressants include, *e.g.*, amesergide, aminepine, benactyzine, bupropion, clozapine, fezolamine, levoprotoline, lithium, medifoxamine, mianserin, minaprine, olanzapine, oxaflozane, oxitiptan, rolipram, teniloxazine, tofenacin, trazodone, tryptophan, and viloxazine.

c. Antiepileptics

The anticonvulsants, also called anti-epileptic drugs (AEDs) are a diverse group of drugs used in prevention of the occurrence of epileptic seizures and bipolar disorders. AEDs suppress the rapid and excessive firing of neurons that begins a seizure and/or prevents the spread of the seizure within the brain and offer protection against possible excitotoxic effects that may result in brain damage. Many anticonvulsants block sodium channels, calcium channels, AMPA receptors, or NMDA receptors.
Anti-epileptic agents include, but are not limited to, benzodiazepines, barbiturates, valproates, GABA agents, iminostilbenes, hydantoins, NMDA antagonists, sodium channel blockers and succinamides.

Benzodiazepines include, e.g., alprazolam, chlordiazepoxide, cholrazepate, clobazam, clonazepam, diazepam, halazepam, lorazepam, oxazepam, and prazepam.

Barbiturates used as anti-epileptics include, e.g., amobarbital, mepobarbital, methylphenobarbital, pentobarbital, phenobarbital, and primidone.

Valproates used as anti-epileptics include, e.g., sodium valporate, valproic acid, valproate semisodium, and valpromide.

Anti-epileptic GABA agents include, e.g., gabapentin, losigamone, pregabalin, retigabine, rufinamide, and vigabatrin.

Carbamazepine and oxcarbazepine are examples of iminostilbenes. Hydantoins include, e.g., fosphenytoin sodium, mephenytoin, and phenytoin sodium.

NMDA antagonists such as harkoseramide are used as anti-epileptics. Sodium channel blockers such as lamotrigine are also anti-epileptic agents.

Succinimides include, e.g., ethosuximide, methsuximide, and phensuximide.

Other anti-epileptic drugs include acetazolamide, briveracetam, CBD cannabis derivative, clomthiazole edisilate, divalproex sodium, felbamate, isovaleramide, lacosamide, lamotrigine, levetiracetam, methanesulphonamide, talampanel, tiagabine, topiramate, safinamide, seletracetam, soretolide, stiripentol, sultiam, valrocemide, and zonisamide.

d. Antiemetics

Anti-emetics are drugs effective against vomiting and nausea. Anti-emetics are typically used to treat motion sickness and the side effects of opioid analgesics, general anaesthetics, and chemotherapy.
Classifications of anti-emetics include, e.g., 5-hydroxytryptamine 3 (5-HT3) receptor antagonists, histamine receptor antagonists, dopamine receptor antagonists, muscarinic receptor antagonists, acetyl choline receptor antagonists, cannabinoid receptor antagonists, limbic system inhibitors, NK-1 receptor antagonists, corticosteroids, tachykinin antagonists, GABA agonists, cannabinoids, benzodiazepines, anticholinergics, and substance P inhibitors.

5-HT3 receptor antagonists include, e.g., alosetron, azasetron, bemesetron, cilansetron, dolasetron, granisetron, indisetron, itasetron, ondansetron, palonosetron, propisetron, ramosetron, renzapride, tropisetron, and zatosetron.

Corticosteroid anti-emetics include dexamethasone and methylprednisolone.

Limbic system inhibitors include alprazolam, lorazepam, and midazolam.

Dopamine receptor antagonists include diphenhydramine, dronabinol, haloperidol, metoclopramide, and prochlorperazine.

NK-1 receptor antagonists used as an anti-emetic include aprepitant and morpholine, and an example of a GABA agonist is propofol.

Thiethylperazine is a type of histamine receptor antagonist.

Cannabinoid receptor antagonists used as anti-emetics include dronabinol, nabilone, rimonabant, tanarabout, and tetrahydrocannabinol.

Examples of other anti-emetics include acetylleucine, monoethanolamine, alizapride, benzquinamide, bietanautine, bromopride, buclizine, chlorpromazine, clebopride, cyclizine, dimenhydrinate, dipheniodol, domperidone, dranisetron, meclizine, methalltal, metopimazine, oxypendyl, pipamazine, piprinhydrinate, scopolamine, thioproperzaine, and trimethobenzamide.

e. Cannabinoid Receptor Antagonists

The cannabinoid receptors are a class of the G-protein coupled receptor superfamily. Their ligands are known as cannabinoids. There are currently two known subtypes, CB1 which is expressed mainly in the brain, but also in the
lungs, liver, and kidney, and CB2, which is mainly expressed in the immune system and in hematopoietic cells. It is also believed that there are novel cannabinoid receptors that is, non-CB1 and non-CB2, which are expressed in endothelial cells and in the CNS. Cannabinoid receptor antagonists may be selective for either the CB1 or CB2 receptor. The present invention contemplates the use of either or both CB1 and CB2 receptor antagonists.

Addictive agents {e.g., alcohol, opiates, Delta(9)-tetrahydrocannabinol (Delta(9)-THC) and psychostimulants, including nicotine} elicit a variety of chronically relapsing disorders by interacting with endogenous neural pathways in the brain. In particular, they share the common property of activating mesolimbic dopamine brain reward systems, and virtually all abused drugs elevate dopamine levels in the nucleus accumbens. Cannabinoid-1 (CB1) receptors are expressed in this brain reward circuit and modulate the dopamine-releasing effects of Delta(9)-THC and nicotine.

Rimonabant (SR141716), a CB1 receptor antagonist, blocks both the dopamine-releasing and the discriminative and rewarding effects of Delta(9)-THC in animals. Although CB1 receptor blockade is generally ineffective in reducing the self-administration of cocaine in rodents and primates, it reduces the reinstatement of extinguished cocaine-seeking behaviour produced by cocaine-associated conditioned stimuli and cocaine priming injections. Similarly, CB1 receptor blockade is effective in reducing nicotine-seeking behaviour induced by re-exposure to nicotine-associated stimuli. In human clinical trials, rimonabant was shown to block the subjective effects of Delta(9)-THC in humans and prevents relapse to smoking in ex-smokers.

Other examples of cannabinoid receptor CB1 antagonists include SR141716A (rimonabant), rosanabant, taranabant and CP-945598.

C. Methods of Treating and Preventing Relapse

Relapse use, or reinstatement, refers to the process of returning to the use of alcohol or another addictive agent or the practice of an addictive behaviour
after a period of abstinence from, or limited or reduced use of, an addictive agent or practice of an addictive behaviour. In certain situations, relapse use of an addictive agent refers to the return to use of an addictive agent by a subject who has undergone physical withdrawal from the addictive agent. Typically, the subject will have undergone physical withdrawal from the addictive agent during a period of non-use or limited or reduced use of the addictive agent. In one embodiment, relapse use occurs in a subject who has previously undergone a treatment regime with an effective amount of an anti-addiction agent to reduce or eliminate use of an addictive agent, but who is no longer using an effective amount of the anti-addiction agent. Anti-addictive agents include any and all agents used to treat or prevent addiction or withdrawal symptoms.

Alcoholism, like many other addictions, is a chronic relapsing disorder characterized by high recidivism rates. Two major factors triggering relapse behaviour are stress and environmental conditioning experiences (O'Brien et al. 1997; Monti et al. 1993; Shaham et al. 1995), which probably facilitate relapse to alcohol-seeking via distinct brain mechanisms. For example, activation of the mesolimbic dopamine system via an opioid-dependent mechanism (or via direct alterations in dopamine transmission in the basolateral nucleus of amygdala) seems to mediate the effect of drug-associated cues (Liu and Wiess 2002; Ciccocioppo et al. 2001), and, extrahypothalamic CRF within the bed nucleus of the stria terminalis and median raphe nucleus is likely to mediate stress-induced reinstatement of drug-seeking behaviour (Erb et al. 1998; Shaham et al. 1995; Lê et al. 2000).

Several lines of evidence suggest that molecular mechanisms underlying relapse to addiction are common to different classes of drugs of abuse. Drug craving and loss of control over drug taking behaviour associated to relapse are under the direct influence of stress and environmental conditioning stimuli; the two major factors affecting resumption to drug use.

Chronic drug abuse produces neuroadaptive changes not only within systems implicated in the acute reinforcing effects of ethanol, but also within other motivational systems, notably brain stress-regulatory mechanisms. Stress has an
established role in the initiation and maintenance of drug abuse, and is a major
determinant of relapse in abstinent individuals (Brown et al. 1995; Marlatt et al. 1985; McKay et al. 1995; Wallace 1989). The significance of stress in drug-seeking
behaviour has also been amply documented in the animal literature. Physical,
social, and emotional stress can facilitate acquisition or increase self-administration
of cocaine (Goeders et al. 1995; Haney et al. 1995; Ramsey and VanRee 1993;
Ahmed and Koob 1997), heroin, (Shaham and Stewart 2004), and ethanol (Nash et
al. 1998; Mollenauer et al. 1993; Blanchard et al. 1987; Higley et al. 1991)) in
rodents and nonhuman primates. Stressful stimuli have also been shown to elicit
reinstatement of cocaine, heroin, and ethanol-seeking behaviour in drug-free animals
following extinction (Ahmed and Koob 1997; Shaham 1993; Shaham and Stewart
1995; Le et al. 1998) and these findings provide experimental support for a role of
stress in relapse.

Traditionally, stress-related drug-seeking behaviour has been thought
to be mediated via activation of the hypothalamic-pituitary-adrenal (HPA) axis.
However, growing evidence suggests that the non-neuroendocrine corticotropin-
releasing factor (CRF) system in the central nucleus of the amygdala (CeA) may play
a significant independent role in the regulation of addictive behaviour associated with
stress. The CeA is rich in CRF immunoreactive cell bodies, terminals, and
receptors, and this neuronal CRF system has been implicated in the mediation of
behavioural and emotional responses to stressful stimuli (Dunn and Berridge 1990;
Koob et al. 1994). For example, immobilization stress elevates extracellular CRF
levels in the CeA (Merlo Pich et al. 1995; Merali et al. 1998) while intra-CeA injection
of the CRF receptor antagonist, α-helical CRF9-41 , reduces behavioural signs of
anxiety produced by social and environmental stressors (Heinrichs et al. 1992;
Swiergiel et al. 1993). Anxiety and stress-like symptoms are central to drug and
alcohol withdrawal syndromes. Considering the evidence on a role of CRF neurons
in the CeA in the regulation of emotional and anxiogenic effects of stress, it is likely
that anxiogenic and stress-like consequences of withdrawal from drugs of abuse
may be mediated by the CRF system in the CeA as well.
Changes in the regulation of the activity of the CRF system within the CeA may represent a critical neuroadaptive mechanism responsible for the development of dependence and compulsive drug-seeking behaviour.

The data discussed above identify neuroadaptive changes in brain circuitries and perturbations in stress systems as an important element in compulsive drug-seeking behaviour and dependence. Another important factor in the long-lasting addictive potential of drugs of abuse is the conditioning of their rewarding actions with specific environmental stimuli. Environmental cues repeatedly associated with the subjective effects of drugs of abuse including alcohol can evoke drug craving (Childress et al. 1988; Ehrman et al. 1992; Monti et al. 1993; Pomerleau et al. 1983; Stormark et al. 1995) or elicit automatic behavioural responses (Miller and Gold 1994; Tiffany and Carter 1998) that ultimately may lead to relapse. Learned responses to drug-related stimuli may, therefore, contribute critically to the high rates of relapse associated with cocaine and other drug addiction.

Data from operant response-reinstatement models developed to investigate drug-seeking behaviour associated with exposure to drug-related environmental cues in rats indicate that discriminative stimuli predictive of cocaine (Weiss et al. 2000), ethanol (Katner et al. 1999; Katner and Weiss 1999), or heroin (Gracy et al. 2000) availability reliably elicit strong recovery of extinguished drug-seeking behaviour in the absence of further drug availability. The response-reinstating effects of these stimuli show remarkable resistance to extinction with repeated exposure and, in the case of cocaine, can still be observed after several months of forced abstinence. Additionally, in the case of ethanol, drug-seeking behaviour induced by ethanol-predictive discriminative stimuli was found to be enhanced in genetically alcohol-preferring P rats compared to Alcohol Nonpreferring (NP) and nonselected Wistar rats (Weiss and Ciccocioppo 1999). This observation demonstrates that genetic predisposition toward heightened ethanol intake is reflected also by a greater susceptibility to the motivating effects of ethanol cues (i.e., enhanced drug-seeking under conditions where behaviour is not directly reinforced by ethanol itself). Together, these findings strongly support the
hypothesis that learned responses to drug-related stimuli are a significant factor in long-lasting vulnerability to relapse.

In humans, relapse risk involves multiple determinants that are likely to interact. For example, exposure to drug cues may augment vulnerability to relapse imparted by protracted withdrawal symptoms resulting from neuroadaptative changes in dependent individuals. Interactive effects exacerbating relapse risk may also exist between the motivating effects of stress and drug-related cues. Recent work addressing these issues has confirmed that additive interactions between the response-reinstating effects of ethanol-associated cues and stress can indeed be demonstrated, and that these effects are enhanced in rats with a history of ethanol dependence (Liu and Weiss 2000).

In experimental laboratories, reinstatement of drug seeking is obtained with administration of the α-2 adrenoreceptor antagonist yohimbine, which, increasing brain noradrenaline cell firing and release, acts as a pharmacological stressor. Footshock stress and yohimbine-induced reinstatement of drug-seeking behaviours both represent valid experimental models to investigate stress-induced alcohol relapse (Lee et al. 2004; Lê et al. 2000).

As shown in the accompanying Examples, PPARγ agonists significantly reduce stress-induced relapse use of an addictive agent (Example 5). In addition, in human patients, pioglitazone, a TZD, consistently reduced OCDS score (Example 22). Obsession for alcohol and the urge to drink (which are measured by OCDS scale) are the major predictors of relapse. These data indicate, therefore, that pioglitazone has anti-relapse properties.

Interestingly, the results showed that pioglitazone did not significantly prevent relapse elicited by conditioning factors. Interestingly, various reports have shown that the nonselective opiate receptor antagonist naltrexone reduces the urge to drink elicited by presentation of alcohol cues in human alcoholics (Monti et al. 1993) and decreases the efficacy of an alcohol cue to reinstate extinguished responding at a previously drug-paired lever in rats (Katner et al. 1999). However,
naltrexone does not reduce relapse behaviour elicited by stress (Le A.D. Psychopharmacology 1998).

These findings suggest that the use of a combination of pioglitazone and naltrexone should result in a synergistic action to reduce relapse behaviour elicited by both stress and conditioning factors.

Accordingly, the present invention provides treatment methods and drug combinations that protect individuals from the effects of more than a single environmental risk factor (i.e., stress and environmental conditioning factors).

In one embodiment, the present invention provides a method of treating or preventing stress-induced relapse use of an addictive agent, comprising providing a PPARγ agonist to a subject who has undergone physiological withdrawal from an addictive agent.

In a related embodiment, the invention includes a method of treating or preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who previously reduced or eliminated use of an addictive agent or practice of an addictive or compulsive behaviour in response to exposure to an effective amount of another anti-addiction treatment, wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment. The anti-addiction treatment may be an anti-addiction drug or may be a non-pharmacologic therapy such as counseling, psychotherapy or hypnosis therapy. The relapse use may be triggered by stress.

In certain embodiments, the subject is no longer exposed to an effective amount of an anti-addiction agent because the subject has become tolerant to the agent, such that the blood plasma concentration of the anti-addiction agent that was previously effective in treating the addiction is no longer effective. In other embodiments, the subject is no longer exposed to an effective amount of an anti-addiction agent because the subject is now exposed to a lower blood plasma concentration of the anti-addiction agent, and this lower blood plasma concentration is not effective.
In certain embodiments of the methods of the present invention, the subject has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour. This period of abstinence or limited or reduced use may be, e.g., at least 24 hours, at least 48 hours, at least 3 days, at least 5 days, at least one week, at least 2 weeks, at least 1 month, at least 2 months, at least 4 months, at least 6 months, at least 9 months, at least one year, at least 2 years, or at least 5 years.

In another embodiment, the present invention includes a method of treating or preventing relapse use of an addictive agent, comprising providing a PPARγ agonist and an opioid antagonist to a subject who has undergone physiological withdrawal from the addictive agent.

In a further embodiment, the present invention includes a method of treating or preventing relapse use of an addictive agent, comprising providing a PPARγ agonist and a CB1 antagonist, e.g., disulfiram, topiramate, levetiracetam, SSRIs, or ondansetron, to a subject who has undergone physiological withdrawal from the addictive agent.

In particular embodiments, the relapse use is triggered by stress, an environmental conditioning factor, or both. Examples of suitable PPARγ agonists are TDZs, such as pioglitazone, etc. One example of a suitable opioid receptor antagonist is naltrexone.

While the methods of the present invention may be practiced in subjects addicted to a single addictive agent, they may also be used in subjects addicted to two or more addictive agents. Similarly, while these methods may be used to prevent relapse use of the addictive agent from which the subject has undergone withdrawal, they may also be adapted to prevent relapse use or the commencement of use of an addictive agent different than the one from which the subject has undergone physiological withdrawal.
D. Methods of Reducing Withdrawal Symptoms and Treating Depression/Anxiety

Withdrawal, also known as withdrawal/abstinence syndrome, refers to the characteristic signs and symptoms that appear when a drug or addictive agent that causes physical dependence is regularly used for a long time and then suddenly discontinued or decreased in dosage. Withdrawal symptoms can vary significantly among individuals, but there are some commonalities. Brain dysfunction associated with withdrawal is often characterized by depression, anxiety and craving, and, if extreme, can help drive the individual to continue the drug despite significant harm - the definition of addiction - or even to suicide.

Increased heart rate and/or blood pressure, sweating, and tremors are common signs of withdrawal. More serious symptoms such as confusion, seizures, and visual hallucinations indicate a serious emergency and the need for immediate medical care. Alcohol, opiates, benzodiazepines, and barbiturates are the only commonly abused substances that can be fatal in withdrawal. Abrupt withdrawal from other drugs, such as nicotine or psychostimulants, can exaggerate mild to moderate neurotoxic side effects due to hyperthermia and generation of free radicals, but life-threatening complications are very rare.

As demonstrated in the accompanying Examples, PPARγ agonists reduce withdrawal symptoms (Example 21). In addition, they decreased anxiety and depression, which is also associated with withdrawal (Example 22). These data demonstrate that PPARγ agonists may be successfully used to reduce withdrawal symptoms, including depression and anxiety, thus making withdrawal easier for subjects and encouraging them to complete the withdrawal process.

The present invention includes a method of reducing one or more withdrawal symptoms associated with reduced or discontinued use of an addictive agent, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist to a subject undergoing physiological withdrawal from an addictive agent.

The PPARγ agonist may be provided to the subject before the subject begins withdrawal and/or during the withdrawal process. In a related method, a
subject is provided with a PPAR\(\gamma\) agonist over a period of time during which the subject uses a reduced amount of an addictive agent. In one embodiment, the subject uses a step-wise reduced amount of an addictive agent at the same time as a PPAR\(\gamma\) agonist, until physical withdrawal is completed. The subject may then discontinue use of the PPAR\(\gamma\) agonist or continue use of the PPAR\(\gamma\) agonist to prevent relapse. In one particular embodiment, the addictive agent is nicotine, and the subject reduces use of nicotine over a period of time during which the subject is provided with a PPAR\(\gamma\) agonist, such as a TZD, e.g., pioglitazone, alone or in combination with another therapeutic agent.

E. Pharmaceutical Compositions, Routes of Administration, Unit Dosage Forms, Kits,

The present invention has established the efficacy of using combinations of a PPAR\(\gamma\) agonist, e.g., a TZD such as pioglitazone, in combination with one or more additional therapeutic agents, such as opioid antagonists, antidepressents, antiepileptics, antiemetics, and CB1 receptor antagonists. Thus, the present invention further includes compositions comprising one or more PPAR\(\gamma\) agonists and one or more additional therapeutic agents, such as opioid antagonists, mixed opioid agonists/antagonists, antidepressents, antiepileptics, antiemetics, CRF1 receptor antagonists and CB1 receptor antagonists.

In particular embodiments, the composition comprises one PPAR\(\gamma\) agonist and one additional therapeutic agent. In one particular embodiment, a composition comprises a TZD and one additional therapeutic agent. In certain embodiments, the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist. In one embodiment, the opioid antagonist is naltrexone. In another embodiment, the mixed opioid partial agonist/antagonist is buprenorphine. In certain embodiments, the additional therapeutic agent is an antidepressant. In a particular embodiment, the antidepressant is bupropion. In certain embodiments, the additional therapeutic agent is an antiepileptic, an antiemetic, or an opioid antagonist or a mixed opioid partial agonist/antagonist.
In various embodiments, the composition comprises: pioglitazone and naltrexone; ciglitazone and naltrexone; rosiglitazone and naltrexone; englitazone and naltrexone; rivoglitazone and naltrexone; darglidazone and naltrexone; pioglitazone and fluoxentine; ciglitazone and fluoxentine; rosiglitazone and fluoxentine; englitazone and fluoxentine; rivoglitazone and fluoxentine; darglidazone and fluoxentine; pioglitazone and mirtazapine; ciglitazone and mirtazapine; rosiglitazone and mirtazapine; englitazone and mirtazapine; rivoglitazone and mirtazapine; darglidazone and mirtazapine; pioglitazone and topiramate; ciglitazone and topiramate; rosiglitazone and topiramate; englitazone and topiramate; rivoglitazone and topiramate; darglidazone and topiramate; pioglitazone and levetiracetam; ciglitazone and levetiracetam; rosiglitazone and levetiracetam; englitazone and levetiracetam; rivoglitazone and levetiracetam; darglidazone and levetiracetam; pioglitazone and gabapentin; ciglitazone and gabapentin; rosiglitazone and gabapentin; englitazone and gabapentin; rivoglitazone and gabapentin; darglidazone and gabapentin; piolitazone and ondansetron; ciglitazone and ondansetron; rosiglitazone and ondansetron; englitazone and ondansetron; rivoglitazone and ondansetron; darglidazone and ondansetron; pioglitazone and antalarmin; ciglitazone and antalarmin; rosiglitazone and antalarmin; englitazone and antalarmin; rivoglitazone and antalarmin; darglidazone and antalarmin.

The compositions of the present invention may be administered to a subject as a pharmaceutical composition or formulation. In particular embodiments, pharmaceutical compositions of the present invention may be in any form which allows for the composition to be administered to a subject. For example, the composition may be in the form of a solid, liquid or gas (aerosol). Typical routes of administration include, without limitation, oral, topical, parenteral, sublingual, rectal, vaginal, and intranasal. The term parenteral as used herein includes subcutaneous injections, intravenous, intramuscular, epidural, intrasternal injection or infusion techniques.

Pharmaceutical compositions used according to the present invention comprise a PPARγ agonist, another therapeutic agent, and a pharmaceutically
acceptable diluent, excipient, or carrier. "Pharmaceutically acceptable carriers" for therapeutic use are well known in the pharmaceutical art, and are described, for example, in Remington's Pharmaceutical Sciences, Mack Publishing Co. (A.R. Gennaro edit. 1985). For example, sterile saline and phosphate-buffered saline at physiological pH may be used. Preservatives, stabilizers, dyes and even flavoring agents may be provided in the pharmaceutical composition. For example, sodium benzoate, sorbic acid and esters of p-hydroxybenzoic acid may be added as preservatives. 1449. In addition, antioxidants and suspending agents may be used.

Pharmaceutical compositions of the invention are generally formulated so as to allow the active ingredients contained therein to be bioavailable upon administration of the composition to a subject. Compositions that will be administered to a subject may take the form of one or more dosage units, where for example, a tablet, capsule or cachet may be a single dosage unit, and a container comprising a combination of agents according to the present invention in aerosol form may hold a plurality of dosage units.

In particular embodiments, the composition comprising a PPARγ agonist and another therapeutic agent is administered in one or more doses of a tablet formulation, typically for oral administration. The tablet formulation may be, e.g., an immediate release formulation, a controlled release formulation, or an extended release formulation. In one embodiment, a tablet formulation comprises an effective amount of a composition comprising a PPARγ agonist and another therapeutic agent. In particular embodiments, a tablet comprises about 1, 5, 10, 20, 30, 50 100, 150, 200, 250, or 300 mg of a PPARγ agonist, such as pioglitazone, and about 1, 5, 10, 20, 30, 50 100, 150, 200, 250, or 300 mg of another therapeutic agent.

The present invention further includes unit dosage forms of pharmaceutical compositions comprising a PPARγ agonist and another therapeutic agent. Each unit dosage form comprises a therapeutically effective amount of a pharmaceutical composition of the present invention, when used in the
recommended amount. For example, a unit dosage form may include a therapeutically effective amount in a single tablet, or a unit dosage form may include a therapeutically effective amount in two or more tablets, such that the prescribed amount comprises a therapeutically effective amount.

A number of the PPARγ agonists and other therapeutic agents described herein are approved for human use at particular dosages. The present invention contemplates using these agents at their approved dosages or at other effective dosages. Since the combination of a PPARγ agonist and another therapeutic agent has been demonstrated to have synergistic efficacy, it is understood that effective amounts of one or both agents may be reduced when provided together, as compare to the effective amount of each when provided alone. In particular embodiments, a PPARγ agonist is provided to a subject in an amount in the range of 0.1-1000 mg/day, 1-1000 mg/day, 10-100 mg/day, or 25-50 mg/day. In one embodiment, pioglitazone is provided to a patient at about 30 mg/day.

Table 1 lists representative agents used in the present invention and provides the daily dosages at which these agents are conventionally administered to adults for other indications, which dosages are believed to be useful for administration in accordance with the methods of the present invention in the treatment or prevention of addiction and relapse use or practice. Dosages listed are oral unless otherwise indicated. It is believed that the dosages of these agents may be reduced when delivered in combinations of a PPARγ agonist and an additional therapeutic agent in accordance with the present invention for the treatment or prevention of addiction or for the treatment or prevention of relapse use. These reductions may be up to 10% of conventional dosages, or up to 20% of conventional dosages, or up to one third of conventional dosages, up to one half of conventional dosages or up to two thirds of conventional dosages. For example, pioglitazone is most commonly dosed at 30 mg per day for treatment of diabetes, which dosage was found to be effective for the treatment of alcoholism (Example 22). When combined with 50 mg/day naltrexone in accordance with the present invention for
treatment of addiction, it is believed therapeutic effect may be seen at 10-15 mg per day of pioglitazone.

Table 1

<table>
<thead>
<tr>
<th>Therapeutic Agent</th>
<th>Exemplary Dosage Applied as Single Agent</th>
</tr>
</thead>
<tbody>
<tr>
<td>pioglitazone</td>
<td>15-45 mg</td>
</tr>
<tr>
<td>rosiglitazone</td>
<td>2-8 mg</td>
</tr>
<tr>
<td>troglitazone</td>
<td>200-600 mg</td>
</tr>
<tr>
<td>rimonabant</td>
<td>10-20 mg</td>
</tr>
<tr>
<td>buprenorphine</td>
<td>0.3 mg (IV or IM)</td>
</tr>
<tr>
<td></td>
<td>12-16 mg (sublingual)</td>
</tr>
<tr>
<td>naltrexone</td>
<td>25-50 mg</td>
</tr>
<tr>
<td>fluoxetine</td>
<td>20-80 mg</td>
</tr>
<tr>
<td>mirtazipine</td>
<td>15-45 mg</td>
</tr>
<tr>
<td>topiramate</td>
<td>400 mg</td>
</tr>
<tr>
<td>levetiracetam</td>
<td>1,000-6,000 mg</td>
</tr>
<tr>
<td>gabapentin</td>
<td>900-1,800 mg</td>
</tr>
<tr>
<td>ondansetron</td>
<td>8-24 mg</td>
</tr>
<tr>
<td>bupropion</td>
<td>200-400 mg</td>
</tr>
</tbody>
</table>

In one particular embodiment, a unit dosage form of a pharmaceutical composition of the present invention comprises about 30 mg of pioglitazone and about 50 mg of naltrexone. This unit dosage form may consist of one or more tablets.

Certain combinations of PPARγ agonists and other therapeutic agents may not be readily adaptable to coformulation. For example, one of the agents may be more amenable to intravenous administration, while another of the agents may be more amenable to oral administration. Or, the serum half life of the two agents may be such that one must be administered more frequently than the other. Accordingly, the present invention contemplates kits comprising one or more unit dosage forms of
a PPARγ agonist and one or more unit dosage forms of another therapeutic agent, such that the two unit dosage forms may be provided to a subject in a therapeutically effective manner. In particular embodiments, a kit comprises unit dosage forms of pioglitazone and naltrexone; ciglitazone and naltrexone; rosiglitazone and naltrexone; englitazone and naltrexone; rivoglitazone and naltrexone; darglidazone and naltrexone; pioglitazone and fluoxetine; ciglitazone and fluoxetine; rosiglitazone and fluoxetine; englitazone and fluoxetine; rivoglitazone and fluoxetine; darglidazone and fluoxetine; pioglitazone and mirtazapine; ciglitazone and mirtazapine; rosiglitazone and mirtazapine; englitazone and mirtazapine; rivoglitazone and mirtazapine; darglidazone and mirtazapine; pioglitazone and topiramate; ciglitazone and topiramate; rosiglitazone and topiramate; englitazone and topiramate; rivoglitazone and topiramate; darglidazone and topiramate; pioglitazone and levetiracetam; ciglitazone and levetiracetam; rosiglitazone and levetiracetam; englitazone and levetiracetam; rivoglitazone and levetiracetam; darglidazone and levetiracetam; pioglitazone and gabapentin; ciglitazone and gabapentin; rosiglitazone and gabapentin; englitazone and gabapentin; rivoglitazone and gabapentin; darglidazone and gabapentin; piolitazone and ondansetron; ciglitazone and ondansetron; rosiglitazone and ondansetron; englitazone and ondansetron; rivoglitazone and ondansetron; darglidazone and ondansetron; pioglitazone and antalarmin; ciglitazone and antalarmin; rosiglitazone and antalarmin; englitazone and antalarmin; rivoglitazone and antalarmin; darglidazone and antalarmin.

In one embodiment, the present invention includes a kit comprising unit dosage forms of a PPARγ agonist and unit dosage forms of nicotine. In one embodiment, the unit dosage forms of nicotine comprise a plurality of different unit dosage forms of nicotine, wherein the different dosage forms of nicotine represent decreasing amount that may be taken one after the other over a period of time, so as to overcome addiction and effectuate withdrawal from the nicotine. The unit dosage forms of nicotine may be present, e.g., in the form of a skin patch, gum, or a lozenge.
EXAMPLES

The following examples describe a number of studies performed to demonstrate the effect of various PPARγ agonists for treating addiction and preventing relapse for a variety of addictive agents. Certain examples describe studies demonstrating the effect of PPARγ agonists used in combination with other therapeutic agents to treat alcohol addiction. These studies were performed using well-validated laboratory animal models for alcohol abuse and cocaine abuse.

Most of the studies described in Examples 1-21 were conducted using male, genetically selected alcohol-preferring rats, referred to as Marchigian Sardinian alcohol-preferring (msP) rats. These animals were bred at the Department of Pharmacological Sciences and Experimental Medicine of the University of Camerino (Marche, Italy) for 60 generations from Sardinian alcohol-preferring rats of the 13th generation, provided by the Department of Neurosciences of the University of Cagliari, Italy. At the time of the experiments, their body weight ranged between 300 and 350 g. They were housed in a room on a reverse 12-hourlight/dark cycle (lights off at 9:00 a.m.), temperature of 20-22°C and humidity of 45-55%. The rats were offered free access to tap water and food pellets (4RF18, Mucedola, Settimo Milanese, Italy). In the operant self-administration experiments, male heterogeneous Wistar rats (Charles River, Germany) were used.

Experiments were performed at 9:30 a.m., which is the beginning of the dark phase of the light/dark cycle. Separate groups of animals were used in each experiment. All procedures were conducted in adherence to the European Community Council Directive for Care and Use of Laboratory Animals and the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Pioglitazone, rosiglitazone, fluoxetine, mirtazapine, topiramate, gabapentine, ondansetron, and levetiracetam was purchased from commercial sources. Yohimbine and ciglitazone were purchased from SIGMA SRL(Mi, Italy). Naltrexone and GW9662 were obtained from TOCRIS (U.K).
Prior to administration, pioglitazone was suspended in distilled water, and the resulting suspension was maintained under constant agitation until administration. The drug was given orally (OS) via gavage procedure in a volume of 1.0 ml/kg. Yohimbine was dissolved in distilled water and was administered intraperitoneally (IP) in a volume of 1.0 ml/kg. Naltrexone hydrochloride was dissolved in distilled water and administered IP in a volume of 1.0 ml/kg. Rosiglitazone, fluoxetine, mirtazapine, topiramate, gabapentin and levetiracetam were suspended in distilled water, and resulting suspensions were maintained under constant agitation until administration. These drugs were given orally (OS) via gavage procedure in a volume of 1.0 ml/kg. Yohimbina was dissolved in distilled water and was administered intraperitoneally (IP) in a volume of 1.0 ml/kg. GW9662 was prepared in 5% DMSO and 5% TWIN 80 and was given either IP (1 ml/kg) or intacerebroventricularly (ICV, 1 µl/rat). Antalarmin was prepared in 10 % TWIN 80 and was given IP (1 ml/kg). Ondansetron was prepared in acqueous solution and was given IP (1 ml/kg).

At the beginning of the experiments, msP rats were allowed free choice between water and 10% (v/v) alcohol 24 h/day for at least 15 days. The fluids were offered in graduated drinking tubes equipped with metallic drinking spouts. The position (to the right or left) of alcohol and water drinking tubes was changed daily to avoid the development of side preference. Water and food were available ad libitum, while alcohol access was either restricted to 2 hours/day (Examples 1 and 2) or was available 24 hours/day (Examples 3 and 4). Alcohol, water and food intakes were measured.

Training and testing were conducted in standard operant chambers (Med Associate) located in sound-attenuating, ventilated environmental cubicles. Each chamber was equipped with a drinking reservoir (volume capacity: 0.30 ml) positioned 4 cm above the grid floor in the centre of the front panel of the chamber, and two retractable levers located 3 cm to the right or to the left of the drinking receptacle. Auditory and visual stimuli were presented via a speaker and a light
located on the front panel. A microcomputer controlled the delivery of fluids, presentation of auditory and visual stimuli, and recording of the behavioural data.

Rats were trained to self-administer 10% alcohol (v/v) in 30-min. daily sessions on a fixed-ratio 1 schedule of reinforcement, in which each response resulted in delivery of 0.1 ml of fluid as previously described (Economidou et al. 2006). For the first 3 days, rats were allowed to lever-press for a 0.2% (w/v) saccharin solution, and then trained to self-administer 10% alcohol by fading the saccharine (Weiss et al. 1993). During the first 6 days of training, rats were allowed to lever-press for a 5.0% (v/v) alcohol solution containing 0.2% (w/v) saccharin. Starting on day 7, the concentration of alcohol was gradually increased from 5.0% to 8.0% and finally to 10.0% (w/v), while the concentration of saccharin was correspondingly decreased to 0%.

Cue-induced reinstatement of alcohol-seeking behaviour experimental procedures consisted of three phases: (1) conditioning phase; (2) extinction phase; and (3) reinstatement phase, as described below.

For the conditioning phase, at the completion of the fading procedure (see above), in 30 min daily sessions, animals were trained to discriminate between 10% alcohol and water. Beginning with self-administration training at the 10% alcohol concentration, discriminative stimuli (SD) predictive of alcohol versus water availability were presented during the alcohol and water self-administration sessions, respectively. The discriminative stimulus for alcohol consisted of the odour of an orange extract (S⁺) whereas water availability (i.e. no reward) was signalled by an anise extract (S⁻). The olfactory stimuli were generated by depositing six to eight drops of the respective extract into the bedding of the operant chamber. In addition, each lever-press resulting in delivery of alcohol was paired with illumination of the chamber’s house light for 5 sec (CS⁺). The corresponding cue during water sessions was a 5 second tone (70 dB) (CS⁻). Concurrently with the presentation of these stimuli, a 5 sec. time-out period was in effect, during which responses were recorded but not reinforced. The olfactory stimuli serving as S⁺ or S⁻ for alcohol availability were introduced one minute before extension of the levers and remained present
throughout the 30-min. sessions. The bedding of the chamber was changed and bedding trays were cleaned between sessions. During the first three days of the conditioning phase, the rats were given alcohol sessions only. Subsequently, alcohol and water sessions were conducted in random order across training days, with the constraint that all rats received a total of 10 alcohol and 10 water sessions.

For the extinction phase, after the last conditioning day, rats were subjected to 30-min extinction sessions for 15 consecutive days. During this phase, sessions began by extension of the levers without presentation of the SD. Responses at the lever activated the delivery mechanism but did not result in the delivery of liquids or the presentation of the response-contingent cues (house light or tone).

The reinstatement testing phase began the day after the last extinction session. This test lasted 30-min under conditions identical to those during the conditioning phase, except that alcohol and water were not made available. Sessions were initiated by the extension of both levers and presentation of either the alcohol S+ or water S− paired stimuli. The respective SD remained present during the entire session and responses at the previously active lever were followed by activation of delivery mechanism and a 5-sec presentation of CS+ in the S+ condition or the CS− (tone) in the S− condition. Rats were tested under the S7CS− condition on day 1 and under S+/CS+ condition on day 2.

Stress-induced reinstatement of alcohol-seeking experimental procedures consisted of three phases: (1) training phase; (2) extinction phase; and (3) reinstatement phase, as described below.

For the training phase, after completion of the fading procedure, msP rats were trained to self-administer 10% (v/v) alcohol for 15 days in 30-min daily sessions under a FR1 schedule of reinforcement. During the infusion, a stimulus house light was turned on for 5 s (time out; TO). Lever presses during the TO period were counted, but did not lead to further infusions.

For the extinction phase, after the last alcohol self-administration session, animals were subjected to 30-min extinction sessions for 15 consecutive
days. Responses at the lever activated the delivery mechanism but did not result in the delivery of alcohol.

For the reinstatement phase, the day after the last extinction session, rats were injected with yohimbine (1.25 mg/kg) and after 30 minutes were placed in the operant chamber and lever presses was monitored for 30 min. It is known that administration of the α-2 adrenoreceptor antagonist yohimbine, increasing brain noradrenaline cell firing and release, acts as a pharmacological stressor and facilitates relapse to alcohol seeking (Le et al. 2005).

Analysis of variance (ANOVA) of data was used to evaluate the results. When appropriate, ANOVA was followed by post-hoc tests. In particular, the effect of acute administration of pioglitazone, naltrexone or their combination on alcohol intake (Examples 1 and 2) was evaluated by mean of a two-way ANOVA with two within factors (time and treatment). The effect of chronic administration of pioglitazone, naltrexone or their combination on alcohol intake (Examples 3 and 4) was evaluated by mean of a three-way ANOVA with one between factor (treatment) and two between factors (days and hours). The effect of pioglitazone on reinstatement of alcohol seeking (Examples 5 and 6) was evaluated by mean of a one-way ANOVA with repeated measures using drug dose as a within subject factor. Alcohol self-administration (Example 7) in Wistar rats was studied by one-way ANOVA with one within factor (dose). Post hoc analysis was carried out using the Newman-Keuls test.

EXAMPLE 1

EFFECT OF ACUTE PIOGLITAZONE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE

The effect of acute pioglitazone administration on voluntary ethanol intake was demonstrated by first training rats to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). After acquisition of a stable baseline of ethanol intake (6-8 g/kg bw; daily), alcohol access was restricted to 2 hours a day at the beginning of the dark phase. Water and food were freely available.
Once stable ethanol drinking baseline was reached (also under limited access conditions), rats (n=7) were tested for the effect of pioglitazone (0.0, 10.0, 30.0 mg/kg) using a within subject counterbalanced Latin square design where each animal received all drug doses. Before starting the treatment, rats were trained to gavage administration procedures for three days, during which they received vehicle (distilled water).

Treatments were carried out at intervals of at least three days. Before each ethanol drinking experiment, msP rats received two doses of pioglitazone or vehicle at 12 hours and at 1 hour before access to ethanol. Drinking experiments were conducted right at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 30, 60, 90 and 120 minutes after ethanol was made available.

Analysis of variance revealed the absence of a significant treatment effect on ethanol intake F(2,6)= 1.22 NS]. However, a significant treatment time interaction was detected [F(6,18)= 6.87 p<0.01]. As shown in Figure 1, post-hoc tests revealed that acute treatment with 30 mg/kg of pioglitazone significantly reduced ethanol consumption at 2 hours but not at 30, 60, or 90 minutes. The selectivity effect was demonstrated by the lack of significant effects on water and food consumption (data not shown).

**EXAMPLE 2**

**EFFECT OF ACUTE PIOGLITAZONE PLUS NALTREXONE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

In this experiment, the effect of the co-administration of pioglitazone and naltrexone on alcohol consumption was examined to demonstrate that PPARγ agonists could enhance the inhibitory action of opioid antagonists on ethanol intake. The dose of naltrexone used in these studies (0.25 mg/kg) was previously shown to be marginally effective in reducing ethanol intake in msP rats under the same experimental conditions (Ciccocioppo et al. 2007).
The msP rats (n=8) were prepared for the study as described in Example 1. After acquisition of a stable baseline of ethanol intake, alcohol access was restricted to 2 hours a day at the beginning of the dark phase. Water and food were freely available. Animals were tested for the effect of the combination between pioglitazone (0.0, 10.0, 30.0 mg/kg) given at 12 hours and at 1 hour before access to ethanol and naltrexone (0.0 and 0.25 mg/kg) injected 2 minutes after the second pioglitazone administration. The experiment was conducted using a within subject counterbalanced Latin square design where each animal received all drug doses.

These experiments were conducted at the beginning of the dark cycle and alcohol, and water and food intakes were monitored at 30, 60, 90 and 120 minutes after ethanol was made available. Water and food intakes were not significantly modified by the various treatments.

Analysis of variance revealed a significant overall effect of treatment \([F(3,7)= 5.95 \ p<0.01]\) on alcohol intake. As shown in Figure 2, post-hoc tests demonstrated that both naltrexone alone and naltrexone + pioglitazone significantly reduced ethanol intake at 30, 60, and 90 minutes. At 120 minutes, the treatments with naltrexone alone and naltrexone + pioglitazone (10 mg/kg) did not show significant effects. In contrast, compared to controls, the co-administration of naltrexone + pioglitazone (30 mg/kg) showed a significant effect also at 120 minutes (p<0.05). This data suggests that co-administration of the two drugs results in an enhancement of their effects, or could result in an increased duration of naltrexone effect.

**EXAMPLE 3**

**EFFECT OF SUBCHRONIC PIOGLITAZONE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of subchronic pioglitazone administration was demonstrated using rats trained to drink 10% (v/v) alcohol for 24 hours a day (free choice between water and ethanol) until a stable baseline of ethanol intake was reached. At this point, msP rats (N=9/group) were tested for the effect of pioglitazone (0.0, 10.0, or 30.0 mg/kg) on ethanol intake using a between subject design, in which each group
of animals received a different dose of drug. Before starting the treatment, rats were trained to gavage administration procedures for three days, during which they received vehicle (distilled water).

Pioglitazone treatment was continued for seven consecutive days, and drug (or vehicle) was administered twice a day at 12 hour and at 1 hour before the beginning of dark period of the light/dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours. Fluids and food intakes were monitored for three additional days after the end of the drug treatment period.

Sub-chronic (7 days) pioglitazone administration significantly reduced voluntary ethanol intake in msP rats. Analysis of variance revealed a significant overall effect of treatment \([F (2,33)= 9.51 ; p<0.01]\). As shown by post-hoc tests, the effect appeared from the first day of treatment at the highest drug dose (Figures 3A, 3B and 3C). The effect progressively increased during treatment, and starting from the 4th day of treatment, both drug doses (10 and 30 mg/kg) significantly reduced ethanol intake.

During treatment, water consumption was rather low and was not significantly affected by drug treatment. Conversely, food intake (Figure 3D) was significantly increased by pioglitazone \([F (2,33) = 7.34 \ p < 0.01 ]\). The effect was higher after administration of the lowest dose (10 mg/kg) of drug. At the end of the treatment, rats gradually recovered from the effect of the drug and ethanol intake and progressively returned at pre-treatment levels (data not shown).

**EXAMPLE 4**

**EFFECT OF SUBCHRONIC PIOGLITAZONE PLUS NALTREXONE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of chronic co-administration of pioglitazone and naltrexone on alcohol consumption was studied to evaluate if PPARγ agonists could also enhance the inhibitory action of opioid antagonists on ethanol intake after repeated treatments. As in the studies described in Example 2, a naltrexone dose (0.25 mg/kg) previously shown to be marginally effective in reducing ethanol intake in msP
rats was used (Ciccocioppo et al. 2007). According to a between-subject design, four groups of msP rats (N=9/group) were prepared as described in Example 3. Specifically, once a stable baseline of daily ethanol consumption was reached, different groups of msP rats were tested for the effect of pioglitazone in combination with naltrexone. For seven consecutive days, msP rats received pioglitazone treatments (0.0, 10.0, or 30.0 mg/kg) at 12 hour and at 1 hour before the beginning of the dark of the light/dark cycle, while naltrexone (0.0 and 0.25 mg/kg) was injected 2 minutes after the second pioglitazone administration. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours. Fluids and food intakes were monitored for three additional days after the end of the drug treatment period.

Sub-chronic (7 days) administration of naltrexone or naltrexone + pioglitazone significantly reduced voluntary ethanol intake in msP rats. Analysis of variance revealed a significant overall effect of treatment $[F(3,32)=9.59 \ p<0.01]$. As shown by post-hoc tests (Figures 4A, 4B and 4C), naltrexone significantly reduced ethanol intake at 2 hours ($p<0.05$) but not at 8 and 24 hours. In addition, the effect progressively decayed during treatment days. Animals treated with pioglitazone plus naltrexone, instead, significantly reduced their drinking at all time points tested (2, 8 and 24 hours). This effect remained significant for the entire period of treatment. These results indicate that pioglidazone and naltrexone co-administration may result in additive or synergistic effects.

During treatment, water consumption was rather low and was not significantly affected by drug treatment. Conversely, food intake was significantly increased by pioglitazone $[F(3,32)= 5.34 \ p < 0.05](Figure \ 4D)$. The effect was higher after administration of the lowest dose (10 mg/kg) of drug. At the end of the treatment, rats gradually recovered from the effect of the drug and ethanol intake progressively returned at pre-treatment levels.
EXAMPLE 5
EFFECT OF ACUTE PIOGLITAZONE ADMINISTRATION ON YOHIMBINE-INDUCED
REINSTATEMENT OF ALCOHOL SEEKING

To demonstrate the effect of TZDs on stress-induced relapse to alcohol seeking, following acquisition of a stable baseline of 10% ethanol, responding msP rats (n=10) were subjected to an extinction period (14 days) during which ethanol responding progressively decreased. The day after the last extinction session, rats were subjected to the reinstatement test. The animals were treated OS with pioglitazone (0.0, 10.0, or 30.0 mg/kg) at 12 hours and 1 hour before the reinstatement test. Yohimbine (1.25 mg/kg, IP) was given 30 min after the last pioglitazone administration.

Animals received all drug treatments according to a counterbalance Latin square design. A 3-day interval, during which animals were subjected to extinction sessions, was allowed between drug tests. In the reinstatement test, active and inactive lever responses were recorded.

A stable baseline of responding for 10% (v/v) alcohol was established in 15 days. Following this alcohol self-administration phase, extinction training began. During the extinction phase, responding progressively decreased, and the last extinction day values were 16.1 ±3.9. The intraperitoneal administration of the alpha-2 adrenoceptor antagonist yohimbine at the dose of 1.25 mg/kg significantly reinstated the operant response for alcohol F(1,18)=22.78 p<0.01]. As shown by the analysis of variance, pre-treatment with pioglitazone significantly reduced the effect of yohimbine [F(2,9)=12.21 , p<0.01] (Figure 5). Post-hoc analysis demonstrated a significant inhibition of reinstatement following administration of 30 mg/kg of pioglitazone (p<0.01).

At the lowest dose (10 mg/kg), pioglitazone showed a clear trend (p=0.07) to an inhibition of yohimbine effect. Analysis of inactive lever responding revealed absence of treatment effects at this lever. This indicated the selectivity of the effect of yohimbine in eliciting reinstatement of alcohol seeking.
EXAMPLE 6
EFFECT OF ACUTE PIOGLITAZONE ADMINISTRATION ON CUE-INDUCED REINSTATEMENT OF ALCOHOL SEEKING

In this experiment to demonstrate the effect of TZDs on cue-induced relapse to alcohol use, msP rats (n=14) were trained to operantly self-administer 10% ethanol or water in 30 min daily session on an FR-1 schedule of reinforcement, where each response resulted in delivery of 0.1 mL of fluid. Ethanol availability was signalled by the odor of an orange extract, which served as a discriminative stimulus. In addition, each lever press resulting in delivery of ethanol was paired with illumination of the house light for 5 s (S+/CS+). For water, anise odor and a 5 sec white noise were employed as discriminative and contiguous cues (SVCS−), respectively. Rats were than subjected to daily extinction sessions, during which lever presses progressively decreased.

The reinstatement test was conducted by re-exposing them to the conditioned stimuli predictive of ethanol or water availability but in the absence of the fluids. Pioglitazone (0.0, 10.0, 30.0 mg/kg) was given 12 hours and 1 hour before the reinstatement test. Experiments were conducted at the beginning of the dark phase of the light/dark cycle. Animals received all drug treatments according to a counterbalance Latin square design, and a 3-day interval was allowed between reinstatement sessions. In the reinstatement test, active and inactive lever responses were recorded.

Throughout the conditioning phase, in which animals discriminated between alcohol or water availability, rats responded at a higher level for alcohol. ANOVA showed a significant overall effect of conditioning [F(1 .28)= 41.89, p<0.01]. On the last day of the discrimination period, animals reached a lever pressing response of about 120 in 30 min., while the response for water was 20. During extinction, lever pressing progressively decreased to 5.87±1.07 of the last extinction day. In the reinstatement test, the ANOVA showed that cues had a significant overall effect on alcohol-seeking [F(1 .28)= 30.4, p<0.01]. A more detailed analysis showed a robust reinstatement of responding under the S+/CS+ (p<0.01 ) but not under the
S\textsuperscript{−}/CS\textsuperscript{−} compared with the last day of extinction. As shown in Figure 6, conditioned reinstatement of alcohol-seeking was not significantly modified by pre-treatment with pioglitazone. Responses at the inactive lever were not influenced by the treatment (data not shown).

**EXAMPLE 7**

**EFFECT OF CIGLITAZONE ADMINISTRATION ON ETHANOL SELF-ADMINISTRATION IN WISTRA RATS**

This study was performed to demonstrate the effect of pioglitazone on ethanol intake extends also to other PPAR\textgammag agonists. The effect of ciglitazone, a structurally different TDZ, on ethanol-self-administration was determined. In addition, to verify that the effect observed with pioglitazone extends to other experimental alcohol intake models, these studies were performed in heterogeneous Wistar rats under operant self-administration conditions.

Wistar rats (n=7) were trained to self-administer ethanol 30 min/day under FR1 schedule of reinforcement. Once a stable level of responding was reached, in a within subject counterbalance order (Latin square design), rats were treated with ciglitazone (0.0, 5.0 or 20.0 mg/kg) given IP 30 minutes before the beginning of the self-administration session. The number of responses to the active and inactive levers were recorded. A 3-day interval was allowed between self-administration sessions.

During ethanol self-administration, Wistar rats acquired robust operant alcohol responding. At the end of this phase, rats pressed the alcohol lever an average of 30-35 times in 30-min. At this point, animals were treated with ciglitazone IP. Results showed that ciglitazone treatment significantly reduced ethanol self-administration \[F(2,6)=5.87 \quad p<0.05\]. Responding at the inactive lever was very low and was not affected by drug treatment \[F(2,6)=1.52 \quad \text{NS}\]. Post hoc tests showed that ethanol self-administration was significantly reduced following administration of the highest dose of drug (Figure 7).
EXAMPLE 8
EFFECT OF ACUTE ROSIGLITAZONE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE

The ability of another TZD, rosiglitazone, to reduce ethanol intake was demonstrated. MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), rats (n= 28) were tested for the effect of rosiglitazone (0.0, 7.5 and 15 mg/kg) using a between subject design. Before starting the treatment, rats were trained to gavage administration procedures for three days, during which they received vehicle (distilled water). Rosiglitazone was given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant treatment effect on ethanol intake [F(2,18)= 0.4 p<0.05]. As shown in Figure 8, post-hoc Newman-Keuls tests revealed that acute treatment with 15 mg/kg of rosiglitazone significantly reduced ethanol consumption at 2 hours (p<0.05). Inhibition of ethanol drinking was highly significant at 24 hours (p<0.01). The selectivity effect was demonstrated by the lack of significant effects on water and food consumption (data not shown).

EXAMPLE 9
EFFECT OF IP ADMINISTRATION OF THE PPARγ ANTAGONIST GW9662 ON PIOGLITAZONE-INDUCED REDUCTION OF ETHANOL INTAKE

This experiment demonstrated that the effect of pioglitazone on ethanol intake was mediated by activation of PPARγ receptors. After acquisition of a stable baseline of ethanol intake, msP rats (n=22) were tested for the effect of GW9662 on pioglitazone-induced reduction of ethanol intake. Rats were treated with 30 mg/kg of pioglitazone given OS 1 hour before access to ethanol. GW9662 was administered IP 30 min after pioglitazone administration, and additional 30 min were awaited before giving ethanol access to rats. Before starting the treatment, rats were trained to gavage and IP administration procedures for three days. Experiment was
conducted in a between subject design (n = 22). Another group of msP rats (n=22) received GW9662 alone to demonstrate the effect of PPARγ blockade on ethanol consumption. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

As shown in Figure 9A, analysis of variance revealed that blockade of PPARγ receptors by GW9662 did not modify ethanol drinking in msP rats [F(2,18)= 0.40 NS]. However, analysis of variance revealed a significant treatment effect on ethanol intake F(3,24)= 18.64 p<0.01] following administration of pioglitazone (Figure 9B). Newman-Keuls tests revealed that treatment with 30 mg/kg of pioglitazone significantly reduced ethanol consumption at 8 and 24 hours (p<0.01 ). Pretreatment with GW9662 blocked the effect of pioglitazone in a dose related manner. Water and food consumption were not affected by drug treatments (data not shown).

EXAMPLE 10
EFFECT OF ICV ADMINISTRATION OF THE PPARγ ANTAGONIST GW9662 ON PI OGLITAZONE-INDUCED REDUCTION OF ETHANOL INTAKE

This experiment demonstrated that the effect of pioglitazone on ethanol intake is mediated by activation of brain PPARγ receptors. For this purpose, msP rats (n=6) were treated with GW9662 (5 µg/rat) ICV to selectively block brain PPARγ receptors, while pioglitazone (30 mg/kg) was given OS. The experiment was conducted using a within subject counterbalanced Latin square design, where each animal received all drug doses.

The drinking experiments were conducted at the beginning of the dark cycle, and alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant treatment effect on ethanol intake F(3,5)= 12.93 p<0.001]. As shown in Figure 10, post-hoc Newman-Keuls tests revealed that treatment with 30 mg/kg of pioglitazone significantly reduced ethanol consumption at 2 hours (p<0.05) 8 hours (p<0.05) and at 24 hours (p<0.01 ).
ICV administration of GW9662 did not significantly affect ethanol intake *per se*. However it completely prevented the effect of pioglitazone. Water and food consumption were not affected by drug treatments (data not shown).

**EXAMPLE 11**

**EFFECT OF ACUTE NALTREXONE ADMINISTRATION ON YOHIMBINE-INDUCED REINSTATEMENT OF ALCOHOL SEEKING**

The inability of naltrexone to reduce yohimbine-induced reinstatement of alcohol use was demonstrated. Following acquisition of a stable baseline of 10% ethanol, responding msP rats (n=10) were subjected to an extinction period (14 days) during which ethanol responding progressively decreased. The day after the last extinction session, rats were subjected to the reinstatement test.

To determine whether naltrexone was able to prevent the effect of the pharmacological stressor yohimbine, animals (n=7) were treated IP with the opioid antagonist (0.0, 0.25 and 1.0 mg/kg) 1 hour before the reinstatement test. Yohimbine (1.25 mg/kg, IP) was given 30 min after naltrexone administration. Animals received all drug treatments according to a counterbalance Latin square design. A 3-day interval, during which animals were subjected to extinction sessions, was allowed between drug tests. In the reinstatement test, active and inactive lever responses were recorded.

A stable baseline of responding for 10% (v/v) alcohol was established in 15 days. Following this alcohol self-administration phase, extinction training began. During the extinction phase, responding progressively decreased. The intraperitoneal administration of the alpha-2 adrenoceptor antagonist yohimbine at the dose of 1.25 mg/kg significantly reinstated the operant response for alcohol $F(1,8)=19.99 \ p<0.01$. As shown by the analysis of variance, pre-treatment with naltrexone did not significantly reduce the effect of yohimbine [$F(2,8)=0.46, \ NS$] (Figure 11). Analysis of inactive lever responding revealed absence of treatment effects at this lever (data not shown). This indicates the selectivity of the effect of yohimbine in eliciting reinstatement of alcohol seeking.
EXAMPLE 12
EFFECT OF ACUTE NALTREXONE ADMINISTRATION ON CUE-INDUCED REINSTATEMENT OF ALCOHOL SEEKING

The ability of naltrexone to reduce cue-induced reinstatement of alcohol seeking was demonstrated. In this experiment, msP rats (n=9) were trained to operantly self-administer 10% ethanol or water in 30 min daily session on an FR-1 schedule of reinforcement, where each response resulted in delivery of 0.1 ml of fluid. Ethanol availability was signalled by the odor of an orange extract, which served as a discriminative stimulus. In addition, each lever press resulting in delivery of ethanol was paired with illumination of the house light for 5 s (S+/CS+). For water, anise odor and a 5 sec white noise were employed as discriminative and contiguous cues (SVCS−), respectively. Rats were than subjected to a daily extinction sessions during which lever presses progressively decreased.

Reinstatement tests were conducted by re-exposing them to the conditioned stimuli predictive of ethanol or water availability, but in the absence of the fluids. Naltrexone (0.0, 0.25 and 1.0 mg/kg) was given one hour before the reinstatement test. Experiments were conducted at the beginning of the dark phase of the light/dark cycle. Animals received all drug treatments according to a counterbalance Latin square design, and a 3-day interval was allowed between reinstatement sessions. In the reinstatement test, active and inactive lever responses were recorded.

Throughout the conditioning phase, in which animals discriminated between alcohol or water availability, rats responded at a higher level for alcohol. During extinction, lever pressing progressively decreased. In the reinstatement test, the ANOVA showed that cues had a significant overall effect on alcohol-seeking [F(1,8)= 36.31, p<0.01]. A more detailed analysis showed a robust reinstatement of responding under the S7CS+ (p<0.01) but not under the S7CS− compared with the last day of extinction. As shown in Figure 12, conditioned reinstatement of alcohol-seeking was significantly reduced by naltrexone [F(2,8)= 15.90; p<0.01].
analysis revealed that both doses (0.25 and 1.0 mg/kg) of the opioid antagonist tested significantly reduced reinstatement of ethanol seeking (p<0.01). Responses at the inactive lever were not influenced by the treatment (data not shown).

**EXAMPLE 13**

**EFFECT OF CO-ADMINISTRATION OF PIOGLITAZONE AND NALTREXONE ON YOHIMBINE- AND CUE-INDUCED REINSTATEMENT OF ALCOHOL SEEKING**

The combined effect of a PPARγ agonist, pioglitazone, and an opioid antagonist, naltrexone, on various inducers of reinstated alcohol seeking was determined.

For yohimbine-induced reinstatement of ethanol seeking, after acquisition of a stable baseline of 10% ethanol responding, msP rats (n=9) were subjected to an extinction period (14 days) during which ethanol responding progressively decreased. The day after the last extinction session, rats were subjected to the reinstatement test.

To evaluate whether combination of naltrexone plus pioglitazone was able to prevent the effect of the pharmacological stressor yohimbine, animals were treated IP with the opioid antagonist (1.0 mg/kg) and OS with the TDZ (30 mg/kg) 1 hour before the reinstatement test. Yohimbine (1.25 mg/kg, IP) was given 30 min after naltrexone/pioglitazone administration. Animals received all drug treatments according to a counterbalance Latin square design. A 3-day interval, during which animals were subjected to extinction sessions, was allowed between drug tests. In the reinstatement test, active and inactive lever responses were recorded.

For cue-induced reinstatement of alcohol seeking, another group of msP rats (n=10) were trained to operantly self-administer 10% ethanol or water in 30 min daily session on an FR-1 schedule of reinforcement, where each response resulted in delivery of 0.1 ml of fluid. Ethanol availability was signalled by the odor of an orange extract, which served as a discriminative stimulus. In addition, each lever press resulting in delivery of ethanol was paired with illumination of the house light for 5 s (S+/CS+). For water, anise odor and a 5 sec white noise were employed as
discriminative and contiguous cues (SVCS’), respectively. Rats were then subjected to a daily extinction sessions, during which lever presses progressively decreased.

A reinstatement test was conducted by re-exposing them to the conditioned stimuli predictive of ethanol or water availability, but in the absence of the fluids. Naltrexone (1.0 mg/kg) and pioglitazone were co-administered 1 hour before the reinstatement test. Experiments were conducted at the beginning of the dark phase of the light/dark cycle. Animals received all drug treatments according to a counterbalance Latin square design and a 3-day interval was allowed between reinstatement sessions. In the reinstatement test, active and inactive lever responses were recorded.

For yohimbine-induced reinstatement of alcohol seeking, rats reached a stable baseline of responding for 10% (v/v) alcohol in 15 days. Following this time period, alcohol self-administration phase extinction training began. During the extinction phase, responding progressively decreased. The intraperitoneal administration of the alpha-2 adrenoceptor antagonist yohimbine at the dose of 1.25 mg/kg significantly reinstated the operant response for alcohol F(1,8)=12.86 p<0.01. As shown by the analysis of variance, pre-treatment with naltrexone plus pioglitazone significantly reduced the effect of yohimbine [F(2,8)=5.71, p<0.01] (Figure 13A). Analysis of inactive lever responding revealed absence of treatment effects at this lever.

For cue-induced reinstatement of alcohol seeking, msP rats rapidly learned to discriminate between alcohol or water availability; rats responded at a higher level for alcohol. During extinction, lever pressing progressively decreased. In the reinstatement test, the ANOVA showed that cues had a significant overall effect on alcohol-seeking [F(1,9)=31.83, p<0.01]. A more detailed analysis showed a robust reinstatement of responding under the S+/CS+ (p<0.01) but not under the S7CS− compared with the last day of extinction. As shown in Figure 13B, conditioned reinstatement of alcohol-seeking was significantly reduced by co-administration of naltrexone and pioglitazone [F(2,9)=16.58; p<0.01]. Responses at the inactive lever were not influenced by the treatment (data not shown).
EXAMPLE 14
EFFECT OF ACUTE PIOGLITAZONE PLUS FLUOXETINE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE

In this experiment, the effect of the co-administration of pioglitazone and fluoxetine on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists, e.g., TZDs, and antidepressants, e.g., selective serotonin uptake inhibitors, has synergistic effects on ethanol intake inhibition. For this purpose, a low dose of fluoxetine (3.0 mg/kg, OS) that did not reduce ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen that does not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once a stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n= 34) were tested for the effect of pioglitazone, fluoxetine or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and fluoxetine were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water, and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment \[F(3,30)= 5.37 \text{ p}<0.01\] on alcohol intake. As shown in Figure 14, post-hoc tests demonstrated that a low dose of pioglitazone alone or fluoxetine alone did not significantly modify ethanol intake in msP rats. However, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 2 and 8 hours (\text{p}<0.01 ), as well as at 24 hours (\text{p}<0.05). These data suggest that co-administration of the two drugs exert synergistic inhibitory actions on ethanol drinking.
A modest, nonsignificant trend to an increase of food intake was observed following drug treatment (data not shown). Water consumption was very low and was not modified by drug administration (data not shown).

**EXAMPLE 15**

**EFFECT OF ACUTE PIOGLITAZONE PLUS MIRTAZAPINE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of the co-administration of pioglitazone and mirtazapine on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this antidepressant had synergistic effects on ethanol intake inhibition. For this purpose, a low dose of mirtazapine (5.0 mg/kg, OS) that did not reduce ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen such that it did not significantly affect alcohol intake *per se*.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n= 34) were tested for the effect of pioglitazone, mirtazapine or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and mirtazapine were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment \[F(3,30)= 12.50 \ p<0.01\] on alcohol intake. As shown in Figure 15, post-hoc tests demonstrated that a low dose of pioglitazone alone or mirtazapine alone did not significantly modify ethanol intake in msP rats. However, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 2 and 8 hours (\(p<0.05\)); a significant reduction of ethanol intake at 2 hours was also reported for
pioglitazone alone (p<0.05). These data suggest that co-administration of the two drugs exert synergistic inhibitory actions on ethanol drinking.

A nonsignificant trend to an increase of food intake was observed following drug treatments (data not shown). Water consumption was very low and was not modified by drug administration (data not shown).

EXAMPLE 16
EFFECT OF ACUTE PIOGLITAZONE PLUS TOPIRAMATE ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE

In this experiment, the effect of the co-administration of pioglitazone and topiramate on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this antiepileptic has synergistic effects on ethanol intake inhibition. For this purpose, a low dose of topiramate (30.0 mg/kg, OS) that did not reduce ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen such that it did not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n= 34) were tested for the effect of pioglitazone, topiramate or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and topiramate were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment \[ F(3,30)= 4.35 \text{ p}<0.01 \] on alcohol intake. As shown in Figure 16, post-hoc tests demonstrated that a low dose of pioglitazone alone or topiramate alone did not significantly modify ethanol intake in msP rats. However, co-administration of the
two agents resulted in a marked inhibition of ethanol consumption at 2, 8 and 24 hours (p<0.05); a significant reduction of ethanol intake at 2 hours was also reported for topiramate alone (p<0.05). These data suggest that co-administration of the two drugs exert synergistic inhibitory actions on ethanol drinking.

A nonsignificant trend to an increase of food intake was observed following drug treatments (data not shown). Water consumption was very low and was not modified by drug administration (data not shown).

**EXAMPLE 17**

**EFFECT OF ACUTE PIOGLITAZONE PLUS LEVETIRACETAM ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of the co-administration of pioglitazone and levetiracetam on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this antiepileptic has synergistic effects on ethanol intake inhibition. For this purpose, a low dose of levetiracetam (100.0 mg/kg, OS) that did not reduce ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen, such that it did not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n=33) were tested for the effect of pioglitazone, levetiracetam or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and levetiracetam were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment \[ F(3,29)=3.76 \text{ p}<0.05 \] on alcohol intake. As shown in Figure 17, post-hoc tests demonstrated that a low dose of pioglitazone alone or levetiracetam alone did not
significantly modify ethanol intake in msP rats. Conversely, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 2 hours (p<0.01), as well as at 8 and 24 hours (p<0.05). These data suggest that co-administration of the two drugs exerts synergistic inhibitory actions on ethanol drinking.

Food and water consumption was not modified by drug administration (data not shown).

EXAMPLE 18
EFFECT OF ACUTE PIOGLITAZONE PLUS GABAPENTIN ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE

The effect of the co-administration of pioglitazone and gabapentin on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this antiepileptic has synergistic effects on ethanol intake inhibition. For this purpose, a low dose of gabapentin (60.0 mg/kg, OS) that did not reduce ethanol intake in msP rats in a pilot study was used. Also a pioglitazone dose (10 mg/kg, OS) was chosen, such that it did not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n= 36) were tested for the effect of pioglitazone, gabapentin or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and topiraate were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment [F(3,7)= 3.31 p<0.05] on alcohol intake. As shown in Figure 18, post-hoc tests demonstrated that a low dose of pioglitazone alone or gabapentin alone did not
significantly modify ethanol intake in msP rats. Conversely, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 2 and 8 hours (p<0.05). These data suggest that co-administration of the two drugs exerts synergistic inhibitory actions on ethanol drinking.

A nonsignificant trend to an increase of food intake was observed following drug treatments (data not shown). Water consumption was very low and was not modified by drug administration (data not shown).

**EXAMPLE 19**

**EFFECT OF ACUTE PIOGLITAZONE PLUS ONDANSETRON ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of the co-administration of pioglitazone and ondansetron on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this serotonin-3 (5-HT3) receptor selective antagonist have synergistic effects on ethanol intake inhibition. For this purpose, a low dose of ondansetron (1.0 mg/kg, IP) that did not reduce ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen, such that it did not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats (n= 36) were tested for the effect of pioglitazone, ondansetron, or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days, during which they received drugs vehicle (distilled water). Pioglitazone and ondansetron were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a nonsignificant overall effect of treatment [F(3,32)= 2.73 p<0.05], but a significant treatment time interaction on
alcohol intake was observed $[F(6,64)= 2.29 \ p<0.05]$. As shown in Figure 19, post-hoc tests demonstrated that a low dose of pioglitazone alone or ondansetron alone did not significantly modify ethanol intake in msP rats. However, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 24 hours ($p<0.05$). Water and food consumption was very low and was not modified by drug administration (data not shown). These data suggest that co-administration of the two drugs exerts synergistic inhibitory actions on ethanol drinking.

**EXAMPLE 20**

**EFFECT OF ACUTE PIOGLITAZONE PLUS ANTALARMIN ADMINISTRATION ON VOLUNTARY ETHANOL INTAKE**

The effect of the co-administration of pioglitazone and antalarmin on alcohol consumption was studied to demonstrate that co-treatment with PPARγ agonists and this corticotrophin releasing factor CRF1 receptor selective antagonist has synergistic effects on ethanol intake inhibition. For this purpose, a low dose of antalarmin (15.0 mg/kg, IP) that modestly reduced ethanol intake in msP rats in a pilot study was used. Also, a pioglitazone dose (10 mg/kg, OS) was chosen, such that it did not significantly affect alcohol intake per se.

MsP rats were first trained to drink 10% (w/v) alcohol for 24 hours a day (free choice between water and ethanol). Once stable ethanol drinking baseline was reached (6-8 g/kg/day), in a between subject design, msP rats ($n= 32$) were tested for the effect of pioglitazone, antalarmin or their combination. Rats treated with drug vehicles served as a control. Before starting the treatment, rats were trained to gavage administration for three days during which they received drugs vehicle (distilled water). Pioglitazone and antalarmin were given twice, at 12 hours and 1 hour before access to ethanol. Drinking experiments started at the beginning of the dark cycle. Alcohol, water and food intakes were monitored at 2, 8 and 24 hours after ethanol was made available.

Analysis of variance revealed a significant overall effect of treatment $[F(3,28)= 3.29 \ p<0.05]$ on alcohol intake. As shown in Figure 20, post-hoc tests
demonstrated that a low dose of pioglitazone alone or antalarmin alone did not significantly modify ethanol intake in msP rats. However, co-administration of the two agents resulted in a marked inhibition of ethanol consumption at 8 (p<0.01) and 24 hours (p<0.05); a significant reduction of ethanol intake at 8 hours was also reported for antalarmin alone (p<0.05). These data suggest that co-administration of the two drugs exerts synergistic inhibitory actions on ethanol drinking.

Water and food consumption was not modified by drug administration (data not shown).

**EXAMPLE 21**

**EFFECT OF PIOGLITAZONE ADMINISTRATION ON ALCOHOL WITHDRAWAL**

The effect of pioglitazone administration on alcohol withdrawal was determined in rats. Male Wistar rats were subjected to a six days of intermittent alcohol intoxication. During the dark phase, rats received 4 oral administration of 2.5-3.0 g/kg of 20% ethanol. The first ethanol dose was given at the beginning of the dark phase. The other 3 daily doses were administered at intervals of 3 hours. Rats were not injected during the light phase of the light/dark cycle. Targeted blood alcohol levels were 250-300 mg/dl. After 6 days of this treatment, rats undergo to spontaneous withdrawal, which generally appears between 8 and 14 hours after the last ethanol injection. Pioglitazone (0.0, 10 and 30 mg/kg) was administered twice, 12 hours and 1 hour before rating withdrawal symptoms. Behavioural signs of withdrawal included: (1) presence of the ventromedial distal flexion response; (2) tail stiffness/rigidity; and (3) tremors (Schulteis et al., 1995). Each sign was rated during a 3-5 min observation period on a scale of 0-2 (Macey et al., 1996; Schulteis, et al., 1995). All signs were cumulated to yield an overall withdrawal severity score.

Twelve hours after the last ethanol administration, animals treated with pioglitazone vehicle showed marked withdrawal symptoms. The analysis of variance showed an overall effect of pioglitazone treatment that reduced tail rigidity [F(4,25)=11.98 p<0.001] (Figure 21). Post hoc tests revealed that alcohol withdrawal signs were significantly reduced after administration of both 10 mg/kg and 30 mg/kg of
pioglitazone, with a highly significant effect of tail rigidity (p<0.01), tremors (p<0.01), and ventromedial limb retraction (p<0.01). Interestingly, while measuring withdrawal score, two out of the 7 animals of vehicle treated groups showed convulsion. Conversely, none of the 12 rats treated with pioglitazone showed seizures. These data suggest that pioglitazone not only helps to reduce ethanol drinking (see previous experiments), but it also possesses the ability to reduce or control alcohol withdrawal syndrome and related symptoms, including seizures.

**EXAMPLE 22**

**EFFECT OF PIOGLITAZONE ON ALCOHOL ABUSE IN HUMANS**

An observational study of human patients using pioglitazone (Actos®) for the treatment of diabetes was performed to demonstrate that PPARγ agonists, alone or in combination with opioid antagonists, are effective in reducing ethanol abuse.

A total of 12 patients were enrolled in the study. 4 patients (2 male and 2 female) received only psychotherapy (Control; CRT); 4 patients (male) received naltrexone 50 mg/day (NTX) + psychotherapy; and 4 patients (3 male and 1 female) received pioglitazone 30 mg/day (Actos®; ACT) + psychotherapy. The patients' ages ranged from 25 to 45 years old. All patients had previous unsuccessful experiences of alcohol detoxification. No major psychiatric co-morbidity was identified. Patient treated with Actos® were all diagnosed with diabetes.

Patients were scored at Timepoint 0 (immediately before beginning the treatment) for various psychological indicators. Anxiety was determined using the S.T.A.I. Y-1 Questioner; obsession for alcohol was determined using the Obsessive Compulsive Drinking Scale questioner; and depression was determined using the M.A.D.R.S 10 Item questioner. Daily and weekly ethanol consumption was also scored. Patients were also tested one a week for anxiety, depression, and obsession to drink.

In addition, patients' blood samples were taken at Timepoint 0 and after four weeks (T1) and 12 weeks (T2) of treatment. Hematological parameters
measured included: mean corpuscular volume (MCV); gamma-GT; aspartate aminotransferase (AST); alanine aminotransferase (ALT); and carbohydrate-deficient transferring (CDT). MCV and CDT are biomarkers for ethanol consumption, and GGT, ALT and AST are biomarkers for hepatic functionality.

Data were analyzed by analysis of variance followed by Newman-Keuls post hoc tests when appropriate.

Results demonstrated a rapid normalization of all blood parameters in patients treated with pioglitazone (ACT), as shown in Table 2.

Table 2: Mean value of blood tests

<table>
<thead>
<tr>
<th></th>
<th>T0 ACT</th>
<th>T0 NTX</th>
<th>T0 CTR</th>
<th>T1 ACT</th>
<th>T1 NTX</th>
<th>T1 CTR</th>
<th>T2 ACT</th>
<th>T2 NTX</th>
<th>T2 CTR</th>
</tr>
</thead>
<tbody>
<tr>
<td>MCV</td>
<td>102.26</td>
<td>101.96</td>
<td>102.78</td>
<td>99.82</td>
<td>101.96</td>
<td>103.26</td>
<td>91.37</td>
<td>97.96</td>
<td>104.87</td>
</tr>
<tr>
<td>GGT</td>
<td>192.21</td>
<td>167.38</td>
<td>173.58</td>
<td>86.7</td>
<td>91.35</td>
<td>181.67</td>
<td>38.26</td>
<td>42.9</td>
<td>179.47</td>
</tr>
<tr>
<td>ALT</td>
<td>51.6</td>
<td>62.7</td>
<td>58.7</td>
<td>45.6</td>
<td>52.8</td>
<td>52</td>
<td>24.9</td>
<td>41.8</td>
<td>55.9</td>
</tr>
<tr>
<td>AST</td>
<td>69.2</td>
<td>49.3</td>
<td>82.1</td>
<td>51.9</td>
<td>41.5</td>
<td>78.4</td>
<td>29.3</td>
<td>38.3</td>
<td>77</td>
</tr>
<tr>
<td>CDT</td>
<td>3.2</td>
<td>3.6</td>
<td>3.8</td>
<td>2.9</td>
<td>3.0</td>
<td>3.7</td>
<td>2.1</td>
<td>2.5</td>
<td>3.1</td>
</tr>
</tbody>
</table>

The decrease of MCV and CDT indicated that patients' ethanol drinking progressively decreased over the two-months of drug treatment. The decrease in GGT, ALT and AST reflected normalisation of hepatic function. In naltrexone treated patients (NTX), a decrease in MCV and CDT was also observed, but to a lesser extent compared to the pioglitazone group. Hepatic parameters were also ameliorated by naltrexone, but again the effect of pioglitazone was more pronounced. The Control group that received only psychotherapy did not show any improvement during the 2-month treatment.

Statistical analysis revealed an overall effect of treatment for all blood parameters measured (MCV, [F(2,9)= 89.7 PO.0001]; GGT; [F(2,9)= 5328 PO.0001]; ALT [F(2,9)= 52.57 PO.0001]; AST [F(2,9)= 771 PO.0001]; CDT [F(2,9)= 26.54 PO.0001]). Post hoc tests revealed that for all the five biomarkers a statistical difference (P<0.001) exists between controls (psychotherapy alone) and patients treated with naltrexone (PO. 001) or with pioglitazone (PO. 001).
Pioglitazone was more effective than naltrexone in reducing the values of MCV (P<0.001), GGT (P<0.001) and ALT (p<0.001). No significant difference between naltrexone and pioglitazone were detected for CDT and AST.

Results also showed a progressive decrease in anxiety score during treatment. Pioglitazone showed the highest effect, as shown in Table 3. In control patients, (psychotherapy alone) anxiety did not diminish during treatment.

<table>
<thead>
<tr>
<th>T</th>
<th>ACT</th>
<th>NIX</th>
<th>CTR</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>59</td>
<td>61</td>
<td>63</td>
</tr>
<tr>
<td>1.1</td>
<td>58</td>
<td>61</td>
<td>64</td>
</tr>
<tr>
<td>1.2</td>
<td>55</td>
<td>59</td>
<td>62</td>
</tr>
<tr>
<td>1.3</td>
<td>54</td>
<td>53</td>
<td>69</td>
</tr>
<tr>
<td>1.4</td>
<td>56</td>
<td>52</td>
<td>65</td>
</tr>
</tbody>
</table>

The analysis of variance revealed an overall effect of treatment ([F(2,9)= 142.86 PO.0001]). Post hoc tests revealed statistically significant difference between controls and patients treated with naltrexone (P<0.001) or with pioglitazone (P<0.001). Pioglitazone was more effective than naltrexone, and a significant difference between pioglitazone and naltrexone was also observed (p<0.001).

Results also showed a progressive decrease in obsessive compulsive score for alcohol. The effect was extremely robust for pioglitazone. As shown in Table 4. in control patients, OCDS remained at pre-treatment level.
Table 4: Obsessive compulsive drinking scale (OCDS) (mean score values)

<table>
<thead>
<tr>
<th></th>
<th>ACT</th>
<th>NTX</th>
<th>CTR</th>
</tr>
</thead>
<tbody>
<tr>
<td>T=0</td>
<td>50</td>
<td>49</td>
<td>52</td>
</tr>
<tr>
<td>T=1.1</td>
<td>45</td>
<td>47</td>
<td>49</td>
</tr>
<tr>
<td>T=1.2</td>
<td>37</td>
<td>45</td>
<td>48</td>
</tr>
<tr>
<td>T=1.3</td>
<td>36</td>
<td>41</td>
<td>49</td>
</tr>
<tr>
<td>T=1.4</td>
<td>34</td>
<td>40</td>
<td>47</td>
</tr>
<tr>
<td>T=2.1</td>
<td>31</td>
<td>41</td>
<td>47</td>
</tr>
<tr>
<td>T=2.2</td>
<td>29</td>
<td>43</td>
<td>49</td>
</tr>
<tr>
<td>T=2.3</td>
<td>28</td>
<td>42</td>
<td>50</td>
</tr>
<tr>
<td>T=2.4</td>
<td>28</td>
<td>44</td>
<td>51</td>
</tr>
</tbody>
</table>

T=0 corresponds to the beginning of the treatment; T=1.1 corresponds to month 1, week 1; T=1.2 corresponds to month 1, week 2, etc.

The analysis of variance revealed an overall effect of treatment ([F(2,9)= 329.27 PO.0001]). Post hoc tests revealed statistically significant difference between controls and patients treated with naltrexone (P<0.001) or with pioglitazone (P<0.001). Pioglitazone was more effective than naltrexone, and a significant difference between pioglitazone and naltrexone was also observed (p<0.001).

The initial Score in the MADRS scale indicated that this patient population did not have severe co-morbid depression. During treatment with pioglitazone, the depression score decreased starting from the second week of treatment, as shown in Table 5. At week 3, it reached the plateau. However, a floor effect might have contributed to rapid plateau.
Table 5: Depression Scale M.A.D.R.S (mean score values)

<table>
<thead>
<tr>
<th>T=0</th>
<th>ACT</th>
<th>NIX</th>
<th>CTR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>21</td>
<td>19</td>
<td>20</td>
</tr>
<tr>
<td>T=1.1</td>
<td>15</td>
<td>16</td>
<td>18</td>
</tr>
<tr>
<td>T=1.2</td>
<td>13</td>
<td>18</td>
<td>19</td>
</tr>
<tr>
<td>T=1.3</td>
<td>10</td>
<td>16</td>
<td>17</td>
</tr>
<tr>
<td>T=1.4</td>
<td>11</td>
<td>17</td>
<td>19</td>
</tr>
</tbody>
</table>

The analysis of variance revealed an overall effect of treatment ([F(2,9)= 42.12 PO.0001]). Post hoc tests revealed statistically significant difference between controls and patients treated with pioglitazone (P<0.001) but not naltrexone. Pioglitazone was significantly different also from naltrexone (p<0.001)

In summary, the blood parameters determined during the course of this study indicated normalization of different alcohol drinking related markers in patients treated with pioglitazone or naltrexone. The effect was more robust with pioglitazone. Patients under psychotherapy alone did not show improvements during treatment. This indicates that the difference between Controls and Drug treated patients depended upon the pharmacological intervention.

High comorbidity exists between alcohol abuse, anxiety and depression. The symptoms of these mood-related disorders tend to exacerbate during early alcohol detoxification phase, thus contributing to reduced patients compliance. In this respect, it is highly relevant that pioglitazone reduces anxiety and depressive symptoms in alcoholic patients. This could also explain why after two
months of drug administration, all 4 patients under pioglitazone were still in treatment, whereas 2 patients of the control group and 1 of the naltrexone the group dropped out. It is also highly relevant that pioglitazone consistently reduced OCDS score. Obsession for alcohol and the urge to drink (which are measured by OCDS scale) are the major predictors of relapse. These data indicate, therefore, that pioglitazone has anti-relapse properties.

The absence of a placebo treatment in the control (psychotherapy alone) group may have contributed to the high efficacy of drug treatments, since the effect of naltrexone was higher that that normally reported in controlled randomized clinical trials. However, placebo effect cannot account for the difference between pioglitazone and naltrexone efficacy. In fact, in this case, both groups of patients received pharmacological medications in association with psychotherapy. Based on this consideration, while it cannot be ruled out that the effect of pioglitazone could have been over estimated to some extent in these studies, it is evident that this drug has a significant efficacy in controlling alcohol abuse, and its effect may be superior to naltrexone.

EXAMPLE 23

EFFECT OF PIOGLITAZONE ON COCAINE SELF-ADMINISTRATION

The ability of pioglitazone to reduce cocaine use was demonstrated in a rat model of cocaine addiction. Cocaine hydrochloride (obtained from the National Institute on Drug Abuse, Bethesda, MD) was dissolved in sterile physiological saline at a concentration of 0.25 mg/0.1 ml. Drug or vehicle solution was infused at a volume of 0.1 ml over 4 s. Pioglitazone obtained from a commercial source was suspendend in distilled water, and the resulting suspension was maintained under constant agitation until administration. Pioglitazone was given orally (OS) via gavage procedure 12 hours and 1 hour before the beginning of cocaine self-administration.

Male Wistar rats weighing between 180 and 200 g at the time of arrival in the lab were used. The rats were housed in groups of three in a humidity- and
temperature-controlled (22°C) vivarium on a 12 h:12 h reverse light/dark cycle (on, 17:00; off, 05:00) with ad libitum access to food and water. One week after arrival, rats were subjected to surgery, and a silastic catheter was implanted into the right jugular vein.

Rats (n=6) were trained to self-administer cocaine in 2-h daily sessions on a fixed-ratio 5 schedule of reinforcement, in which each response resulted in delivery of 0.25 mg/0.1 ml of fluid cocaine solution. Cocaine self-administration training continued until a stable baseline of responding was reached (less than 10% variation for 3 consecutive days calculated for each single rat). At this point, drug testing begun.

In a within subject counterbalance order (Latin square design), rats were treated with pioglitazone (0.0, 10.0 or 30.0 mg/kg) given OS 12 hours and 1 hour before the beginning of the self-administration session. The number of responses to the active and inactive levers was recorded. A 3-day interval was allowed between drug testing. During these intervals, cocaine self-administration was continued to re-establish baseline lever responses.

The effect of pioglitazone administration on cocaine self-administration was evaluated by mean of a one-way within factor ANOVA followed by Newman-Keuls post hoc test.

Treatment with pioglitazone significantly reduced cocaine self-administration [F(2,5)=13.189 p<0.01]. Post hoc tests revealed a significant (p<0.01) reduction of cocaine self-administration at both 10.0 and 30.0 mg/kg of pioglitazone (Figure 22A). Responses at the left inactive lever were very low and were not modified by pioglitazone treatment (Figure 22B).

EXAMPLE 24
EFFECT OF PIOGLITAZONE ON NICOTINE USE

The ability of PPARγ agonists and antidepressent to reduce nicotine use was demonstrated in an animal model of nicotine addiction.
Bupropion hydrochloride (Sigma, Milan, Italy) was dissolved in saline. Nicotine tartrate (Sigma, Milan, Italy) was dissolved in isotonic saline at a concentration of 0.03 mg/0.1 ml free base. The pH of the nicotine solution was adjusted to 7 with dilute NaOH. Drug or vehicle solution was infused at a volume of 0.1 ml over 4 s. Pioglitazone was obtained from commercial source; it was suspendend in distilled water, and the resulting suspension was maintained under constant agitation until administration. Pioglitazone was given orally (OS) via gavage procedure at 12 hours and 1 hour before the beginning of nicotine self-administration.

Male Wistar rats weighing between 180 and 200 g at the time of arrival in the lab were used. The rats were housed in groups of three in a humidity- and temperature-controlled (22° C) vivarium on a 12 h:12 h reverse light/dark cycle (on, 17:00; off, 05:00) with ad libitum access to food and water. One week after arrival, the rats were subjected to surgery, and a silastic catheter was implanted into the right jugular vein.

Rats (n=9) were trained for one week to self-administer cocaine in 2-h daily sessions on a fixed-ratio 5 schedule of reinforcement, in which each five response resulted in delivery of 0.25 mg/0.1 ml of fluid cocaine solution. After the successful completion of cocaine training, rats were allowed to self-administer nicotine at the 0.03 mg/kg/infusion dose by switching the delivery of cocaine for the delivery of a nicotine infusion. Nicotine self-administration training continued until stable baseline of responding was established (less than 20% variation for 3 consecutive days calculated for each single rat). At this point, drug testing began.

In a within subject counterbalance order (Latin square design), rats were treated with pioglitazone (0.0 and 30.0 mg/kg) given OS 12 hours and 1 hour before the beginning of the self-administration session. The number of responses to the active and inactive levers was recorded. A 3-day interval was allowed between drug testing. During these intervals, nicotine self-administration was continued to re-establish lever responses baseline.
The effect of pioglitazone administration on nicotine self-administration was evaluated by mean of a paired t-test. Statistical significance was set at $P<0.05$

After a few training days, Wistar rats acquired robust operant responding for nicotine. As shown in Fig 23A, treatment with 30 mg/kg pioglitazone significantly reduced nicotine self-administration ($t_{df8}=-2.70$, $p<0.05$). Responses at the left inactive lever was very low and were not modified by pioglitazone treatment (Fig.23B). These results demonstrate that PPARγ agonists are effective in reducing nicotine use.

**EXAMPLE 25**

**EFFECT OF PIOGLITAZONE AND SELECTED THERAPEUTIC AGENTS ON NICOTINE USE**

The ability of PPARγ agonists in combination with other therapeutic agents, such as bupropion, nicotine replacement formulations, naltrexone, varenicline, and CB1 receptor antagonist/inverse agonists, e.g., rimonabant, rosonabant, taranabant, and CP-945598, to synergistically reduce nicotine use is determined in a rat model of nicotine addiction.

Experiments are conducted using operant self-administration paradigms, essentially as described in Example 23 (see also Bruijnzeel and Markou, 2003; Rauhut et al 2003). Briefly, male Wistar rats are implanted with a permanent silastic catheter into the right jugular vein for intravenous nicotine self-administration (0.03 mg/infusion). Using operant self-administration chambers, rats are trained to self-infuse nicotine under a fixed ratio 5 schedule of reinforcement (five lever presses to obtain one nicotine infusion). Nicotine self-administration training is continued until stable baseline of responding is established. At this point, drug testing is begun.

In a within subject counterbalance order (Latin square design), rats are treated with pioglitazone (predicted dose range 5-30.0 mg/kg) or with other PPARγ agonists in combination with bupropion, nicotine (replacement formulations; i.e., nicotine patches), naltrexone, varenicline, or rimonabant. To evaluate synergism between PPARγ agonists and these latter drugs, the lowest effective dose for each
of the compounds is tested in association with the PPARγ agonist. A dose range for bupropion is 10-100 mg/given OS; a dose range for naltrexone is 0.25-2.5 mg/kg given IP; a dose range for varenicline is 0.25-2.5 mg/kg given IP; and a dose range for rimonabant is (0.1-3.0 mg/kg given IP) (Bruijnzeel and Markou, 2003; Rauhut et al. 2003; Steensland P et al. 2007; Cohen et al. 2005). The number of responses to the active and inactive levers is recorded. A 3-day interval is allowed between drug testing. During these intervals, nicotine self-administration is continued to re-establish lever responses baseline.

Data is analyzed by analysis of variance followed by post-hoc tests (Newman-Keuls or Dunnets) where appropriate. Statistical significance is set at $P < 0.05$. It is expected that these experiments will demonstrate that the combination of a PPARγ agonist and any of the listed drugs will act synergistically in reducing nicotine self-administration, thereby demonstrating the efficacy of using PPARγ and any of these drugs to treat addiction.

**EXAMPLE 26**

**EFFECT OF PIOGLITAZONE AND ANTIDEPRESSANTS OR OPIOID AGONIST/ANTAGONIST PARTIALAGONISTS ON COCAINE USE**

The ability of PPARγ agonists in combination with antidepressants, bupropion, fluoxetine, or the opioid parial agonist agonist/antagonist, buprenorphine, to synergistically reduce cocaine use is determined in a rat model of cocaine addiction.

Experiments are conducted using operant self-administration paradigms as described in Example 23 (see also Glatz et al. 2002; Peltier et al. 1993). Briefly, male Wistar rats are implanted with a permanent silastic catheter into the right jugular vein for intravenous cocaine self-administration (0.25 mg/infusion). Using operant self-administration chambers, rats are trained to self-infuse cocaine under a fixed ratio 5 schedule of reinforcement (five lever presses to obtain one cocaine infusion). Cocaine self-administration training is continued until stable baseline of responding is established. At this point, drug testing is begun.
In a within subject counterbalance order (Latin square design), rats are treated with pioglitazone (predicted dose range 5-30.0 mg/kg) or with another PPARγ agonist in combination with bupropion, fluoxetine, or buprenorphine. To evaluate synergism between PPARγ agonists and these latter drugs, the lowest effective dose for each of the compound is tested in association. A dose range for bupropion is 10.0-100.0 mg/kg given OS; a dose range for fluoxetine is 3.0-15.0 mg/kg given OS; and a dose range for buprenorphine is 0.1-5.0 mg/kg given IP (Glatz et al. 2002; Peltier et al. 1993; Sorge et al. 2005). The number of responses to the active and inactive levers are recorded. A 3-day interval is allowed between drug testing. During these intervals, nicotine self-administration is continued to re-establish lever responses baseline.

Data is analyzed by analysis of variance followed by post-hoc tests (Newman-Keuls or Dunnets) where appropriate. Statistical significance is set at $P < 0.05$. It is expected that these experiments will demonstrate that the combination of a PPARγ agonist and any of the listed drugs will act synergistically in reducing cocaine self-administration, thereby demonstrating the efficacy of using PPARγ and any of these drugs to treat addiction.

**EXAMPLE 27**

**EFFECT OF PIOGLITAZONE ON DEVELOPMENT OF OPIATE ADDICTION**

The ability of PPARγ agonists to reduce opiate use and prevent opiate addiction is determined in a rat model of opiate addiction.

Experiments are conducted using a conditioned place preference apparatus and a well established procedure to study morphine induced conditioned place preference (Ciccocioppo et al. 2000). Briefly, using a two-chamber place conditioning apparatus, male Wistar rats are trained to associate morphine effects to one side of the box and saline to the other side. Multiple groups of rats are used, and the experiment is conducted in a between subject design. Animals are pretreated with pioglitazone vehicle before the injection of morphine vehicle. Control group receive morphine and pioglitazone vehicles in both compartments.
The rats are conditioned during a conditioning phase of six days. Every other day, for three times, rats receive subcutaneous injections of 3 mg/kg of morphine or its vehicle. Pioglitazone (5.0-30.0 mg/kg) is injected one hour before morphine. During conditioning, the guillotine door remains closed, and the rats are confined for 1 h in one compartment of the box. The day following the last conditioning session, rats are allowed to explore the entire box for 15 min, and the time spent in each compartment is measured.

Place preference score (referred to as Δ time) for each rat is obtained by subtracting the time spent in the compartment associated with morphine vehicle to the time spent in the compartment associated to morphine injections. The Δ time values are submitted to statistical analysis. Data is analyzed by analysis of variance followed by post-hoc tests (Newman-Keuls or Dunnets) where appropriate. Statistical significance is set at $P < 0.05$.

It is predicted that morphine will elicit a marked conditioned place preference, and treatment with pioglitazone will reduce the acquisition of morphine-induced place conditioning (see for review; Sanchis-Segura and Spanagel 2006). These results will demonstrate the ability of PPARγ agonists to prevent the development of addiction to opioids and more specifically to morphine.

The various embodiments described above can be combined to provide further embodiments. All of the U.S. patents, U.S. patent application publications, U.S. patent applications, foreign patents, foreign patent applications and non-patent publications referred to in this specification and/or listed in the Application Data Sheet, are incorporated herein by reference, in their entirety. Aspects of the embodiments can be modified, if necessary to employ concepts of the various patents, applications and publications to provide yet further embodiments.

These and other changes can be made to the embodiments in light of the above-detailed description. In general, in the following claims, the terms used should not be construed to limit the claims to the specific embodiments disclosed in the specification and the claims, but should be construed to include all possible
embodiments along with the full scope of equivalents to which such claims are entitled. Accordingly, the claims are not limited by the disclosure.
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CLAIMS

1. A method of treating or preventing an addiction, comprising:
determining that a subject has or is at risk of developing an addiction;
and
providing to the subject an amount of an agonist of a peroxisome
proliferator-activated receptor gamma (PPARγ agonist) effective for the treatment or
prevention of the addiction.

2. The method of claim 1, wherein the subject is addicted to an
addictive agent.

3. The method of claim 2, wherein the subject is addicted to an
addictive agent selected from the group consisting of: alcohol, nicotine, marijuana, a
marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, and a
psychostimulant.

4. The method of claim 3, wherein the addictive agent is alcohol.

5. The method of claim 3, wherein the addictive agent is nicotine.

6. The method of claim 3, wherein the opioid agonist is selected
from the group consisting of: morphine, methadone, fentanyl, sufentanil and heroin.

7. The method of claim 3, wherein the psychostimulant is cocaine,
amphetamine or an amphetamine derivative.

8. The method of claim 2, wherein the subject is addicted to more
than one addictive agent.
9. The method of claim 1, wherein the subject is addicted to an addictive or compulsive behavior.

10. The method of claim 9, wherein the addictive or compulsive behavior is selected from the group consisting of: pathological gambling, pathological overeating, pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking.

11. The method of claim 1, wherein the PPARγ agonist is a thiazolidinedione (TZD).

12. The method of claim 11, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

13. The method of claim 12, wherein the TZD is pioglitazone.

14. A method of treating or preventing an addiction, comprising providing to a subject having an addiction, a peroxisome proliferator-activated receptor gamma (PPARγ agonist) and an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of the addiction.

15. The method of claim 14, wherein said additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a
corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

16. The method of claim 15, wherein the opioid antagonist is naltrexone or nalmefene.

17. The method of claim 15, wherein the antidepressant is fluoxetine, mirtazapine, or bupropion.

18. The method of claim 15, wherein the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin.

19. The method of claim 15, wherein the CRF-1 receptor antagonist is antalarmin.

20. The method of claim 15, wherein the selective serotonin-3 (5-HT3) antagonist is ondansetron.

21. The method of claim 15, wherein the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant.

22. The method of claim 15, wherein the mixed opioid agonist/antagonist is buprenorphine.

23. The method of claim 14, wherein the subject is addicted to an addictive agent.

24. The method of claim 23, wherein the subject is addicted to an addictive agent selected from the group consisting of: alcohol, nicotine, marijuana, a
marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, and a psychostimulant.

25. The method of claim 24, wherein the addictive agent is alcohol.

26. The method of claim 24, wherein the addictive agent is nicotine.

27. The method of claim 24, wherein the opioid agonist is selected from the group consisting of: morphine, methadone, fentanyl, sufentanil and heroin.

28. The method of claim 24, wherein the psychostimulant is cocaine, amphetamine or an amphetamine derivative.

29. The method of claim 24, wherein the subject is addicted to more than one addictive agent.

30. The method of claim 24, wherein the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist.

31. The method of claim 30, wherein the opioid antagonist is naltrexone.

32. The method of claim 30, wherein the mixed opioid partial agonist/antagonist is buprenorphine.

33. The method of claim 23, wherein the addictive agent is nicotine and the additional therapeutic agent is an antidepressant.
34. The method of claim 33, wherein the antidepressant is bupropion.

35. The method of claim 14, wherein the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant.

36. The method of claim 35, wherein the antidepressant is bupropion.

37. The method of claim 14, wherein the subject is addicted to two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist.

38. The method of claim 37, wherein the opioid antagonist is naltrexone or nalmefene.

39. The method of claim 37, wherein the mixed opioid partial agonist/antagonist is buprenorphine.

40. The method of claim 14, wherein the subject is addicted to an addictive or compulsive behavior.

41. The method of claim 40, wherein the addictive or compulsive behavior is selected from the group consisting of: pathological gambling, pathological overeating, pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking.
42. A method of preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour.

43. The method of claim 42, further comprising providing to the subject an additional therapeutic agent wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective prevention of the relapse use or practice.

44. A method of treating relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist) to a subject who has undergone a period of abstinence from, or limited or reduced use of, the addictive agent or practice of the addictive or compulsive behaviour.

45. The method of claim 44, further comprising providing to the subject an additional therapeutic agent wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment of the relapse use or practice.

46. A method of preventing relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisone proliferator-activated receptor gamma (PPARγ agonist) to a subject, wherein the subject previously reduced or eliminated use of the addictive agent or practice of the addictive or compulsive behaviour in response to treatment with an effective amount of an anti-addiction treatment, and wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment.
47. The method of claim 46, further comprising providing to the subject an additional therapeutic agent, wherein each of the PPAR\( \gamma \) agonist and the additional therapeutic agent contribute to the effective prevention of the relapse use.

48. A method of treating relapse use of an addictive agent or practice of an addictive or compulsive behaviour, comprising providing an effective amount of a peroxisone proliferator-activated receptor gamma (PPAR\( \gamma \) agonist) to a subject, wherein the subject previously reduced or eliminated use of the addictive agent or practice of the addictive or compulsive behaviour in response to treatment with an effective amount of an anti-addiction treatment, and wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment.

49. The method of claim 46, further comprising providing to the subject an additional therapeutic agent, wherein each of the PPAR\( \gamma \) agonist and the additional therapeutic agent contribute to the effective treatment of the relapse use or practice.

50. The method of any one of claims 42-49, wherein the method treats or prevents relapse use of an addictive agent.

51. The method of claim 50, wherein the addictive agent is selected from the group addictive agent is selected from the group consisting of: alcohol, nicotine, marijuana, a marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, and a psychostimulant.

52. The method of claim 51, wherein the addictive agent is alcohol.

53. The method of claim 51, wherein the addictive agent is nicotine.
54. The method of claim 51, wherein the opioid agonist is selected from the group consisting of: morphine, methadone, fentanyl, sufentanil and heroin.

55. The method of claim 51, wherein the psychostimulant is cocaine, amphetamine or an amphetamine derivative.

56. The method of any one of claims 42-49, wherein the methods treats or prevents relapse practice of an addictive behaviour or compulsion.

57. The method of claim 56, wherein the addictive or compulsive behavior is selected from the group consisting of: pathological gambling, pathological overeating, pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking.

58. The method of any one of claims 42-29, wherein the PPARγ agonist is a thiazolidione (TZD).

59. The method of claim 58, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

60. The method of claim 59, wherein the TZD is pioglitazone.

61. The method of any one of claims 43, 45, 47, and 49, wherein the additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an
antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, a 5-HT2A/2C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

62. The method of claim 61, wherein the opioid antagonist is naltrexone or nalmefene.

63. The method of claim 61, wherein the antidepressant is fluoxetine, mirtazapine, or bupropion.

64. The method of claim 61, wherein the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin.

65. The method of claim 61, wherein the CRF-1 receptor antagonist is antalarmin.

66. The method of claim 61, wherein the selective serotonin-3 (5-HT3) antagonist is ondansetron.

67. The method of claim 61, wherein the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant.

68. The method of claim 61, wherein the mixed opioid agonist/antagonist is buprenorphine.

69. The method of any one of claims 43, 45, 47, and 49, wherein the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

70. The method of any one of claims 42-49, wherein the relapse use or relapse practice is stress-induced.
71. The method of claim 42-47, wherein the subject has undergone physiological withdrawal from the addictive agent during the period of abstinence from, or limited or reduced use of, the addictive agent.

72. The method of any one of claims 46-49, wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment because the subject has become conditioned to the anti-addiction treatment.

73. The method of any one of claim 46-49, wherein the subject is no longer exposed to an effective amount of the anti-addiction treatment because the subject has reduced or eliminated exposure to the anti-addiction treatment.

74. A method of reducing one or more symptoms associated with physiological withdrawal from an addictive agent, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist to a subject undergoing physiological withdrawal from an addictive agent.

75. A method of reducing one or more symptoms associated with physiological withdrawal from an addictive agent, comprising providing an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent to a subject undergoing physiological withdrawal from an addictive agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to reducing one or more symptoms associated with physical withdrawal from the addictive agent.

76. The method of claim 74 or claim 75, wherein the PPARγ agonist is a thiazolidinedione (TZD).
77. The method of claim 76, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

78. The method of claim 56, wherein the TZD is pioglitazone.

79. The method of claim 75, wherein the additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT₂A/₂C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

80. A pharmaceutical composition, comprising a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of an addiction.

81. The pharmaceutical composition of claim 80, wherein the PPARγ agonist is a thiazolidinedione (TZD).

82. The pharmaceutical composition of claim 81, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

83. The pharmaceutical composition of claim 82, wherein the TZD is pioglitazone.

84. The pharmaceutical composition of claim 80, wherein the addiction is an addiction to an addictive agent.
85. The pharmaceutical composition of claim 84, wherein the addictive agent is selected from the group consisting of: alcohol, nicotine, marijuana, a marijuana derivative, an opioid agonist, a benzodiazepine, a barbiturate, and a psychostimulant.

86. The pharmaceutical composition of claim 80, wherein the addiction is an addiction to an addictive or compulsive behaviour.

87. The pharmaceutical composition of claim 86, wherein the addictive or compulsive behavior is selected from the group consisting of: pathological gambling, pathological overeating, pathological use of electronic devices, pathological use of electronic video games, pathological use of electronic communication devices, pathological use of cellular telephones, addiction to pornography, sex addiction, obsessive compulsive disorder, compulsive spending, anorexia, bulimia, intermittent explosive disorder, kleptomania, pyromania, trichotillomania, compulsive overexercising, and compulsive overworking.

88. The pharmaceutical composition of claim 80, wherein the additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

89. The pharmaceutical composition of claim 88, wherein the opioid antagonist is naltrexone or nalmefene.

90. The pharmaceutical composition of claim 88, wherein the antidepressant is fluoxetine, mirtazapine, or bupropion.
91. The pharmaceutical composition of claim 88, wherein the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin.

92. The pharmaceutical composition of claim 88, wherein the CRF-1 receptor antagonist is antalarmin.

93. The pharmaceutical composition of claim 88, wherein the selective serotonin-3 (5-HT3) antagonist is ondansetron.

94. The pharmaceutical composition of claim 88, wherein the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant.

95. The pharmaceutical composition of claim 88, wherein the mixed opioid agonist/antagonist is buprenorphine.

96. The pharmaceutical composition of claim 80, wherein the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist.

97. The pharmaceutical composition of claim 96, wherein the opioid antagonist is naltrexone.

98. The pharmaceutical composition of claim 96, wherein the mixed opioid partial agonist/antagonist is buprenorphine.

99. The pharmaceutical composition of claim 80, wherein the addictive agent is nicotine and the additional therapeutic agent is an antidepressant.
100. The pharmaceutical composition of claim 99, wherein the antidepressant is bupropion.

101. The pharmaceutical composition of claim 80, wherein the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant.

102. The pharmaceutical composition of claim 101, wherein the antidepressant is bupropion.

103. The pharmaceutical composition of claim 80, wherein the addictive agent comprises two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist.

104. The pharmaceutical composition of claim 103, wherein the opioid antagonist is naltrexone or nalmefene.

105. The pharmaceutical composition of claim 103, wherein the mixed opioid partial agonist/antagonist is buprenorphine.

106. The pharmaceutical composition of claim 80, wherein the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

107. A unit dosage form of a pharmaceutical composition adapted for the treatment of alcohol addiction, wherein said unit dosage form comprises a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and an additional therapeutic agent, wherein said unit dosage form comprises the PPARγ agonist and the additional therapeutic agent in a combined amount effective in the treatment of an addiction, and wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of the addiction.
108. The unit dosage form of claim 107, wherein the PPARγ agonist is a thiazolidinedione (TZD).

109. The unit dosage form of claim 108, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

110. The unit dosage form of claim 109, wherein the TZD is pioglitazone.

111. The unit dosage form of claim 107, wherein the additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT2A/2C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

112. The unit dosage form of claim 111, wherein the opioid antagonist is naltrexone or nalmefene.

113. The unit dosage form of claim 111, wherein the antidepressant is fluoxetine, mirtazapine, or bupropion.

114. The unit dosage form of claim 111, wherein the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin.

115. The unit dosage form of claim 111, wherein the CRF-1 receptor antagonist is antalarmin.
116. The unit dosage form of claim 111, wherein the selective serotonin-3 (5-HT3) antagonist is ondansetron.

117. The unit dosage form of claim 111, wherein the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant.

118. The unit dosage form of claim 111, wherein the mixed opioid agonist/antagonist is buprenorphine.

119. The unit dosage form of claim 107, wherein the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.

120. A kit useful for the treatment or prevention of an addiction, comprising: a first container comprising a peroxisome proliferator-activated receptor gamma (PPARγ) agonist; and a second container comprising an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to the effective treatment or prevention of an addiction.

121. The kit of claim 120, wherein the PPARγ agonist is a thiazolidinedione (TZD).

122. The kit of claim 121, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

123. The kit of claim 122, wherein the TZD is pioglitazone.

124. The kit of claim 120, wherein the additional therapeutic agent is selected from the group consisting of: an opioid antagonist, a mixed opioid partial agonist/antagonist, an antidepressant, an antiepileptic, an antiemetic, a
corticotrophin-releasing factor-1 (CRF-1) receptor antagonist, a selective serotonin-3 (5-HT3) antagonist, a 5-HT₂A/²C antagonist, and a cannabinoid-1 (CB1) receptor antagonist.

125. The kit of claim 124, wherein the opioid antagonist is naltrexone or nalmefene.

126. The kit of claim 124, wherein the antidepressant is fluoxetine, mirtazapine, or bupropion.

127. The kit of claim 124, wherein the antiepileptic is selected from the group consisting of: topiramate, levetiracetam, and gabapentin.

128. The kit of claim 124, wherein the CRF-1 receptor antagonist is antalarmin.

129. The kit of claim 124, wherein the selective serotonin-3 (5-HT3) antagonist is ondansetron.

130. The kit of claim 124, wherein the cannabinoid-1 (CB1) receptor antagonist is rimonabant or tanarabant.

131. The kit of claim 124, wherein the mixed opioid agonist/antagonist is buprenorphine.

132. The kit of claim 120, wherein the PPARγ agonist is pioglitazone and the additional therapeutic agent is naltrexone.
133. The kit of claim 120, wherein the addictive agent is alcohol and the additional therapeutic agent is an opioid antagonist or a mixed opioid antagonist/partial agonist.

134. The kit of claim 133, wherein the opioid antagonist is naltrexone.

135. The kit of claim 133, wherein the mixed opioid partial agonist/antagonist is buprenorphine.

136. The kit of claim 120, wherein the addictive agent is nicotine and the additional therapeutic agent is an antidepressant.

137. The kit of claim 136, wherein the antidepressant is bupropion.

138. The kit of claim 120, wherein the addictive agent is a psychostimulant and the additional therapeutic agent is an antidepressant.

139. The kit of claim 138, wherein the antidepressant is bupropion.

140. The kit of claim 120, wherein the addictive agent comprises two or more addictive agents and the additional therapeutic agent is an opioid antagonist or a mixed opioid partial agonist/antagonist.

141. The kit of claim 140, wherein the opioid antagonist is naltrexone or nalmefene.

142. The kit of claim 140, wherein the mixed opioid partial agonist/antagonist is buprenorphine.
143. A kit comprising one or more unit dosage forms of a peroxisome proliferator-activated receptor gamma (PPARγ) agonist and one or more unit dosage forms of nicotine.

144. The kit of claim 143, wherein said one or more unit dosage forms of nicotine comprise two or more different amounts of nicotine.

145. The kit of claim 143, wherein said PPARγ agonist is a thiazolidinedione (TZD).

146. The kit of claim 145, wherein the TZD is selected from the group consisting of: pioglitazone, rosiglitazone, ciglitazone, troglitazone, englitazone, rivoglitazone and darglidazone.

147. A method of preventing a subject from becoming addicted, or reducing the likelihood that a subject will become addicted, to an addictive therapeutic agent, comprising providing to a subject in need thereof an addictive therapeutic agent, and an effective amount of a peroxisome proliferator-activated receptor gamma (PPARγ agonist), wherein the effective amount of the PPARγ agonist is an amount effective in preventing the subject from becoming addicted, or reducing the likelihood that the subject will become addicted, to the addictive therapeutic agent.

148. The method of claim 147, further comprising providing to the subject an additional therapeutic agent, wherein each of the PPARγ agonist and the additional therapeutic agent contribute to preventing the subject from becoming addicted, or reducing the likelihood that the subject will become addicted, to the addictive therapeutic agent.
149. The method of claim 147 or claim 148, wherein the addictive therapeutic agent is an opioid agonist.
Effect of Pioglitazone on Ethanol Intake

**FIG. 1**
Effect of Pioglitazone + Naltrexone on Ethanol Intake

![Graph showing the effect of Pioglitazone and Naltrexone on ethanol intake over time.](graph)

**FIG. 2**
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Effect of Pioglitazone on Ethanol Intake at 2-h

![Graph a showing the effect of Pioglitazone on ethanol intake at 2 hours.]

**Fig. 3A**

Effect of Pioglitazone on Ethanol Intake at 8-h

![Graph b showing the effect of Pioglitazone on ethanol intake at 8 hours.]

**Fig. 3B**
Effect of Pioglitazone on Ethanol Intake at 24-h

FIG. 3C

Effect of Pioglitazone on Food Intake at 24-h

FIG. 3D
**Effect of Pioglitazone + Naltrexone on Ethanol Intake at 2-h**

*FIG. 4A*

**Effect of Pioglitazone + Naltrexone on Ethanol Intake at 8-h**

*FIG. 4B*
FIG. 4C

Effect of Pioglitazone + Naltrexone on Ethanol Intake at 24-h

FIG. 4D

Effect of Pioglitazone + Naltrexone on Food Intake at 24h
Effect of Pioglitazone on Yohimbine-Induced Reinstatement of Ethanol-Seeking Behavior

Mean (±SEM) Number of Rewards (30 min)

EtOH 10%  Ext  0.0  10  30  Pio

Yohimbine 0.125 mg/kg

**

FIG. 5
Effect of Pioglitazone in Cue-Induced Reinstatement of Ethanol Seeking Behavior

FIG. 6
Effect of Rosiglitazone on Ethanol Intake

- Veh
- Ros 7.5 mg/kg
- Ros 15 mg/kg

24h | 8h | 2h

Ethanol Intake (g/kg)
Effect of the PPARγ antagonist GW9662 (i.p) on Ethanol intake

**FIG. 9A**

Effect of GW9662 (i.p) + Pioglitazone on Ethanol Intake

**FIG. 9B**
Effect of GW9662 (ICV) plus Pioglitazone OS on Ethanol Intake

**FIG. 10**
Effect of Naltrexone on Yohimbine-Induced Reinstatement of Ethanol-Seeking Behavior

**FIG. 11**
Effect of Naltrexone + Pioglitazone on Yohimbine-Induced Reinstatement of Ethanol-Seeking Behavior

**FIG. 13A**

Effect of Naltrexone + Pioglitazone on Cue-Induced Reinstatement of Ethanol-Seeking Behavior

**FIG. 13B**
Effect of Pioglitazone + Topiramate on Ethanol Intake

FIG. 16

Ethanol Intake (g/kg)

24h Time (hours)

mg/kg

Veh + Veh
Pio 10
Top 30
Pio 10 + Top 30

*
Score Withdrawal and Pioglitazone 10-30 mg/kg

FIG. 21
Effect of Pioglitazone on Cocaine Self-Administration

**FIG. 22A**

**FIG. 22B**
Effect of Pioglitazone on Nicotine Self-Administration

**FIG. 23A**

- **A**
  - Mean ± (SEM) Number of Rewards
  - Active lever
  - veh
  - pio 30 mg/kg
  - 

- **B**
  - Mean ± (SEM) Number of Responses
  - Inactive lever
  - veh
  - pio 30 mg/kg
  - 

**FIG. 23B**
### INTERNATIONAL SEARCH REPORT

**International application No**
PCT/US 08/60146

**A CLASSIFICATION OF SUBJECT MATTER**

- **IPC(8) - A01 N 43/04; A61 K 31/70 (2008.04)**
- **USPC - 514/44**

According to International Patent Classification (IPC) or to both national classification and IPC

**B FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

USPC  514/44

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

USPC  514/44, 183, 210 2, 297, 369, 381, 394 559 (text search-see search terms below)

Electronic data base consulted during the international search (name of database and, where practicable, search terms used)

PUBWEST(PGPB,USPT,USOC,EPAB,JPAIB), Google, PubMed

Search terms addiction, PPARgamma, glitazone, thiazolidinedione, naltrexone, gabapentin, nmonabant

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- **A** special category of cited documents
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**Date of the actual completion of the international search**

8 July 2008 (08 07 2008)

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<td>Y</td>
<td>CHU et al  Dependence-induced increases in ethanol self-administration in mice are blocked by the CRF1 receptor antagonist antalarmin and by CRF1 receptor knockout Pharmacol Biochem Behav , 3 April 2007, Vol 86, No 4, pp 813-821, abstract</td>
<td>19, 65, 91, 114, 127</td>
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<td>L</td>
<td>CHU et al  Dependence-induced increases in ethanol self-administration in mice are blocked by the CRF1 receptor antagonist antalarmin and by CRF1 receptor knockout Pharmacol Biochem Behav , 3 April 2007, Vol 86, No 4, pp 813-821, ABSTRACT only [downloaded from <a href="http://www.ncbi.nlm.nih.gov/pubmed/17482248?ordinalpos=3&amp;tool=EntrezSystem2&amp;EntrezPubmed/Pubmed_ResultsPanel/Pubmed_RVDocSum">http://www.ncbi.nlm.nih.gov/pubmed/17482248?ordinalpos=3&amp;tool=EntrezSystem2&amp;EntrezPubmed/Pubmed_ResultsPanel/Pubmed_RVDocSum</a>] <em>cited to show publication date of Chu et al (above)</em></td>
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