A systematic review of (pre)clinical studies on the therapeutic potential and safety profile of kratom in humans

Elisabeth Prevete1 | Kim Paula Colette Kuypers1 | Eef Lien Theunissen1 | Ornella Corazza2,3 | Giuseppe Bersani3 | Johannes Gerardus Ramaekers1

1Department of Neuropsychology and Psychopharmacology, Faculty of Psychology and Neuroscience, Maastricht University, Maastricht, the Netherlands
2Department of Clinical, Pharmacological and Biological Sciences, College Lane, University of Hertfordshire, Hatfield, United Kingdom
3Department of Medico-Surgical Sciences and Biotechnologies, Faculty of Pharmacy and Medicine, Sapienza University of Rome, Latina, Italy

Correspondence
Johannes G. Ramaekers and Elisabeth Prevete, Department of Neuropsychology and Psychopharmacology, Faculty of Psychology and Neuroscience, Maastricht University, 616, 6200 MD Maastricht, the Netherlands. Email: j.ramaekers@maastrichtuniversity.nl and e.prevete@maastrichtuniversity.nl

Present address
Elisabeth Prevete, Department of Human Neurosciences, Sapienza University of Rome, Rome, Italy. Email: elisabeth.prevete@uniroma1.it

Abstract

Introduction: Kratom (Mitragyna speciosa) is a tropical plant traditionally used as an ethnomedicinal remedy for several conditions in South East Asia. Despite the increased interest in its therapeutical benefits in Western countries, little scientific evidence is available to support such claims, and existing data remain limited to kratom’s chronic consumption.

Objective: Our study aims to investigate (pre)clinical evidence on the efficacy of kratom as a therapeutic aid and its safety profile in humans.

Methods: A systematic literature search using PubMed and the Medline database was conducted between April and November 2020.

Results: Both preclinical (N = 57) and clinical (N = 18) studies emerged from our search. Preclinical data indicated a therapeutic value in terms of acute/chronic pain (N = 23), morphine/ethanol withdrawal, and dependence (N = 14), among other medical conditions (N = 26). Clinical data included interventional studies (N = 2) reporting reduced pain sensitivity, and observational studies (N = 9) describing the association between kratom’s chronic (daily/frequent) use and safety issues, in terms of health consequences (e.g., learning impairment, high cholesterol level, dependence/withdrawal).

Conclusions: Although the initial (pre)clinical evidence on kratom’s therapeutic potential and its safety profile in humans is encouraging, further validation in large, controlled clinical trials is required.

KEYWORDS
adverse effects, kratom, mitragynine, opioid withdrawal, pain, therapeutic benefits

1 INTRODUCTION

Kratom (Mitragyna speciosa, Rubiaceae family) is an indigenous tropical tree from Southern East Asia (e.g., Malaysia, Thailand, Laos, Cambodia), which also grows in East-West Africa and Papua New Guinea (Hassan et al., 2013; Kruegel and Grundmann, 2018). This evergreen non-seasonal plant is also known locally with other names, such as Biak-Biak, Ketum, Kakuam, Ithang, Thom, and Mambog (Hassan et al., 2013; Velti and Grundmann, 2019). It exerts stimulant cocaine-like effects in doses smaller than 5 g and sedative-like effects...
Kratom leaves are generally smoked, chewed, or brewed as an herbal decoction (Hassan et al., 2013; Kruegel and Grundmann, 2018). It has been used traditionally for centuries to treat several medical conditions such as diarrhea and pain, to mitigate opioid and alcohol withdrawal symptoms, to detoxify from other substances, like cannabis or methamphetamine, to improve sexual desire, and to combat fatigue (Grewal, 1932a; Hassan et al., 2013; Saref et al., 2019a; Singh et al., 2017; Vicknasingam et al., 2010).

Kratom has recently gained popularity as an ethnomedicinal remedy in Western countries, especially in the United States (US), where it is sold online and elsewhere (e.g., gas station, specialty shops) in different formulations, such as tablets, supplements, capsules, or powder (Prozialeck et al., 2012; Tavakoli et al., 2016). Several user-based surveys revealed use to self-treat acute/chronic pain, among other psychiatric conditions, including opioid and substance use disorders (Bath et al., 2020; Coe et al., 2019; Garcia-Romeu et al., 2020; Grundmann, 2017). A case report also referred to its successful use in alleviating COVID-19 related pain (Metastasio et al., 2020).

However, despite this increased scientific interest in kratom, the evidence supporting such self-reported claims is still lacking. It is known that its psychoactive effects are mainly dependent on its major metabolite 7-OH-mitragynine (7HMG) and mitragynine (MG), which together account for 68% of all the alkaloids present in the plant (Hassan et al., 2013; Kruegel and Grundmann, 2018; Sheldard, 1974; Takayama, 2004).

Mitragynine (IUPAC name (E)-2-[2S,3S,12bS]-3-ethyl-8-methoxy-1,2,3,4,6,7,12b-octahydroindolo[2,3-a]quinolizin-2-yl]-3-methoxyprop-2-enone) is an indole alkaloid Corynanthe-type having a monoterpenic portion similarly to yohimbine and the psychedelic substance voacangine (Han et al., 2020; Hassan et al., 2013; Kong et al., 2017a; Ramanathan et al., 2015). It is insoluble in both basic and aqueous solutions but possesses a high solubility in typical organic solvents (e.g., acetone, acetic acid, alcohol, chloroform, and diethyl-ether) (Han et al., 2020; Kong et al., 2017a; Ramanathan et al., 2015). It has intermediate lipophilicity and a high capacity to cross the blood-brain barrier (Yusof et al., 2019).

The compound has been described as a G-protein biased atypical opioid (Fauzi et al., 2020; Gutridge et al., 2020; Raffa et al., 2018) that acts as mu- and delta-opioid receptor agonist (Foss et al., 2020; Matsumoto et al., 1996b, 2006), and kappa-opioid receptor antagonist-like, without β-arrrestin recruitment (Kruegel et al., 2016; Todd et al., 2020; Váradi et al., 2016). Mitragynine also possesses a non-opioid action through (a2) adrenergic receptors, adenosine (A2A), dopamine (D2), and serotonin (5-HT2A, 5-HT2C, and 5-HT7) receptors (Harun et al., 2015; Hiranita et al., 2019; Matsumoto et al., 1996a, 1996b, 1997).

The contribution of these receptors in the (acclaimed) effects of kratom has yet to be determined. A drawback is that most of the available data has been collected in users. It derives from online surveys, drug fora, and case reports. Additionally, Ramachandram et al. (2019) reported that the association between the pharmacodynamics and -kinetics of mitragynine in (pre)clinical models had not been studied yet.

Limited evidence has shown that the compound possesses a biphasic elimination pattern after both oral (p.o.) (half-life (T1/2):3–9 h) and intravenous (i.v.) (T1/2:13 h) administration in rodents (Kong et al., 2017b; Ya et al., 2019), and a large volume of distribution when it was administered (i.v.) in dogs (Maxwell et al., 2020). On the other side, mitragynine has been shown to follow a two compartmental model after oral intake in a small sample of kratom users, with a T1/2 of 23.24 ± 16.07 h (Trakulsrichai et al., 2015).

The metabolism of mitragynine has been described to be mainly hepatic in both human microsomes (Kamble et al., 2019) and preclinical models (Ya et al., 2019), and it would be mediated by cytochrome P450 (CYP450) (Basiliere and Kerrigan, 2020; Hanapi et al., 2013; Kong et al., 2011), which may also be involved in potential drug-drug interaction.

Serious adverse events, including fatalities (Corkery et al., 2019; Wong and Mun, 2020), have been reported only in Western countries, mainly when kratom is used in recreational settings. Suggested reasons are extreme high dose, and co-administration of benzodiazepines, amphetamines, or ethanol, or the presence of adulterants, like the synthetic O-desmethyl tramadol (Anwar et al., 2016; Corkery et al., 2019; Kronstrand et al., 2011; Olsen et al., 2019). Other serious events have been associated with chronic kratom use (Alsarraf et al., 2019; Anwar et al., 2016; Grundmann, 2017; Schimmel and Dart, 2020) and include the risk of addiction, dependence, and withdrawal (Singh et al., 2018c; Veltri and Grundmann, 2019).

The Food and Drug Administration (FDA) and the US Drug Enforcement Administration (DEA) considered these kratom-related reports as dangerous and consequently proposed to place the plant in Schedule I of the Controlled Substances Act (CSA) in 2016 (Eastlack et al., 2020; Grundmann, 2017; Henningfield et al., 2018). However, since a broad public opposition reversed this action, kratom is still legal at the federal level in the US, with many users claiming its therapeutic potential, in the absence of sufficient clinical evidence.

Given this background, the current systematic review aims to investigate whether kratom has potential medical benefits based on preclinical and clinical studies measuring acute and chronic effects on behavior and other clinical outcomes. The second aim was to investigate possible safety issues in humans. The medical applications of kratom reported by users in traditional and non-traditional settings were used to define this review’s search strings.

2 | MATERIALS AND METHODS

2.1 | Data sources and search strategy

A literature search was performed using the PubMed and the Medline database to identify the scientific publications related to kratom’s potential therapeutic utility and safety, as investigated in (pre) clinical research. The search, which was carried out between April
and August 2020, consisted of assessing titles and abstracts using both Medical Subject Headings, or subheadings (MeSH) and free-text terms. The choice of search terms was informed by recent high-quality reviews, papers, and online surveys that reported anecdotal data related to kratom’s benefits in treating pain, psychiatric symptoms and conditions, and several other medical applications (e.g., hypertension, inflammatory conditions, diabetes).

The query’s search strings included a combination of substance [1] and symptoms/condition [2] strings; both included the Boolean command ‘OR’, and they were combined with ‘AND’. The terms used in [1] were kratom, mitragynine, mitragyna, Mitragyna speciosa. The terms used in [2] were: ADD, addiction, ADHD, affective disorders, analgesia, analgesics, anorexia, antihelmintic, antidepressant, anti-inflammatory, antimalarial, antinociceptive, anxiety, anxiolytic, attention deficit disorder, attention deficit hyperactivity disorder, bipolar disorder, blood pressure, cough, dependence, depression, diabetes, diarrhea, diarrheal disease, fever, gastric infection, inflammation, mood disorders, "muscle AND relaxation", opioid use disorder, pain, psychosis, psychotic disorders, stress, stress disorders post traumatic, substance-related disorders, treatment-resistant depression, withdrawal. Terms in this string were combined with ‘OR’. No period restrictions were applied. This search led to 224 hits and was updated on November 2020, to identify records that could have potentially been published during the preparation of this paper for submission. This search gave 7 additional articles.

### 2.2 Inclusion/exclusion criteria

Taking into account the review method and the aim of this study, exclusion criteria were the following: (1) non-original research articles or publications not pertinent or not potentially related to the aims, including those mainly focused on methods of identification in biological samples or sold products, chemistry and physicochemical properties, pharmacology, including pharmacodynamic and pharmacokinetic properties, toxicology or other topics (fatalities, harm reduction, legal status); (2) review, commentaries, or other surveys of the literature; (3) case series and case reports because of their high potential of bias in the study designs; (4) data in humans derived from online surveys.

Studies were included if they met all of the following criteria: (1) preclinical study, in vitro or in vivo, investigating the pharmacology or toxicology potentially related to the review aim, and (2) any clinical outcome providing sufficient scientific evidence of kratom, mitragynine, mitragyna and related or derivative compounds, that would support the traditional medical uses or anecdotal benefits reported by users.

### 2.3 Study selection

All procedures were performed according to PRISMA guidelines (Moher et al., 2009). The selection was conducted in two stages: an initial screening of titles and abstracts against the inclusion criteria to identify potentially relevant papers, followed by screening the full papers assessed for eligibility. The selection was discussed in a small team of four (EP, ET, JR, KK).

### 2.4 Data extraction

When a record reported a combination of review-relevant and -irrelevant data, only the former was included. Based on the included articles’ content, the review was organized in the following categories: (pre)clinical evidence related to potential therapeutic use in pain, withdrawal and dependence, and other medical conditions, and therapeutic application or safety issues in humans.

### 3 RESULTS

#### 3.1 Studies description

In total, 63 studies met the eligibility criteria. After an initial screening, 17 were removed, as they focused on Mitragyna genus per se or on kratom pharmacology and toxicology data and thus not relevant for this review. Additional studies (29) were included in the analysis as a further assessment of relevant citations emerged. Overall, 75 records were deemed relevant to this systematic review (details of the selection process are shown in Figure 1). These included 18 studies performed in humans, and 57 preclinical studies, that were mainly in vivo studies with a brief observation period, with nine having a more extended observation period (Cheaha et al., 2015; Grewal, 1932b; Harun et al., 2020; Hassan et al., 2020; Khor et al., 2011; Kumarnsit et al., 2006, 2007a; Meepong and Sooksawate, 2019; Wilson et al., 2020), and other nine were in vitro (Abdul Aziz et al., 2012; Fakurazi et al., 2013; Ghazali et al., 2011; Goh et al., 2014; Grewal, 1932b; Jamil et al., 2013; Juanda et al., 2019; Parthasarathy et al., 2009; Yuniarti et al., 2020). Since six preclinical studies gave evidence for two potential therapeutic uses, the related content will be described in each specific section of the results.

#### 3.2 (Pre)clinical evidence of potential therapeutic use

##### 3.2.1 Pain

Twenty-three in vivo (mice, rats, or dogs) studies provided evidence for kratom’s potential therapeutic use in the treatment of acute pain (Carpenter et al., 2016; Criddle, 2015; Fakurazi et al., 2013; Hiranita et al., 2019; Idid et al., 1998; Macko et al., 1972; Matsumoto et al., 1996a, 1996b, 2004, 2005, 2006, 2008; Mossadeq et al., 2009; Reanmongkol et al., 2007; Sabeghdam et al., 2010, 2013; Shamima et al., 2012; Stolt et al., 2014; Takayama et al., 2002; Thongpradichote et al., 1998; Wilson et al., 2020) and chronic pain...
Foss et al., 2020; Matsumoto et al., 2014). The antinociceptive effects of the studied preparations were shown in the different models of acute thermal or mechanical stimulus-induced pain, and neuropathic pain, after administration via a range of routes (p.o., i.p., i.v., or i.c.v.).

The studied preparations were *Mitragyna speciosa* (MS) aqueous or methanol or alkaloid extracts (Carpenter et al., 2016; Criddle, 2015; Mossadeq et al., 2009; Reanmongkol et al., 2007; Sabetghadam et al., 2010, 2013), lyophilized kratom tea (LKT) (Wilson et al., 2020), mitragynine alone (Carpenter et al., 2016; Criddle, 2015; Fakurazi et al., 2013; Foss et al., 2020; Hiranita et al., 2019; Idid et al., 1998; Macko et al., 1972; Matsumoto et al., 1996a, 1996b; Shamima et al., 2012; Thongpradichote et al., 1998), or mitragynine + paynantheine (Stolt et al., 2014), and its synthetic derivatives MG Pseudoindoxyl (Takayama et al., 2002) and [(E)-methyl 2-(3-ethyl-7a,12a-(epoxyethanoxy)-9-fluoro-1,2,3,4,6,7,12b-octahydropyrido[2,3-a]quinolizin-2-yl)-3-methoxyacrylate] (MG M-9) (Matsumoto et al., 2008), or 7HMG (Matsumoto et al., 2004, 2005, 2006), and its derivatives (E)-methyl 2-((2S,3S,7aS,12aR,12bS)-3-ethyl-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindolo[2,3-a]quinolizin-2-yl)-3-methoxyacrylate (MGM-15) and (E)-methyl 2-((2S,3S,7aS,12aR,12bS)-3-ethyl-9-fluoro-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindolo[2,3-a]quinolizin-2-yl)-3-methoxyacrylate (MGM-16) (Matsumoto et al., 2014).

According to the evidence included in our analysis, mitragynine’s analgesic effect was similar to classical opioids oxycodone and morphine (MOR) (Carpenter et al., 2016; Criddle, 2015). When combined with MOR in long-term treatment, the analgesic effect was more pronounced (Fakurazi et al., 2013). Further, it was described as more potent and relatively safer than the MS alkaloid extract (Sabetghadam et al., 2013). Wilson et al. (2020) also found LKT’s analgesic effect similar to MOR, with relatively fewer negative effects (Wilson et al., 2020). 7HMG, a partial mu- and delta-
opioid receptors agonist, was described as more potent than MOR (Matsumoto et al., 2004, 2006), with a minor intestinal transit inhibition (Matsumoto et al., 2006). However, it was also found responsible for a locomotor activity increase in a dose-dependent manner (Matsumoto et al., 2008) and producing cross-tolerance to MOR (Matsumoto et al., 2005, 2008). Among its derivatives, authors found MGM-16 to have a superior potency as an opioid agonist in comparison to both MGM-15 and 7HMG (Foss et al., 2020; Matsumoto et al., 2014), and Matsumoto et al. (2008) reported MGM-9 to have higher potency, with lower adverse effects, whether compared to MOR and 7HMG (Matsumoto et al., 2008).

Further, four studies (Fakurazi et al., 2013; Macko et al., 1972; Mossadeq et al., 2009; Wilson et al., 2020) showed that kratom also exerts other therapeutic effects besides analgesic properties, including applications in opioid withdrawal, described in more detail below.

3.2.2 Withdrawal and dependence

Twelve in vivo studies (mice, rats or zebrafish) and two in vitro studies provided evidence for kratom so potential therapeutic use in the treatment of both opioid (Cheaha et al., 2017; Fakurazi et al., 2013; Harun et al., 2020; Hassan et al., 2020; Hemby et al., 2019; Jamil et al., 2013; Khor et al., 2011; Meepong and Sooksawate, 2019; Wilson et al., 2020; Yue et al., 2018) and alcohol use disorders (Cheaha et al., 2015; Gutridge et al., 2020; Kumarnsit et al., 2007a; Vijeeppallam et al., 2019), as shown by the effects of the studied preparations (kratom extracts, LKT, mitragynine and other alkaloids; p.o. or i.p. or i.v. or intragastrically) in models of induced withdrawal, drug consumption/replacement, and dependence.

Among the extracts, the MS alkaloid (Cheaha et al., 2015) and aqueous extract (Kumarnsit et al., 2007a) attenuated ethanol withdrawal. The methanol extract was found to reduce the ethanol-seeking behavior (Vijeepallam et al., 2019), and both extracts (with or without 7HMG) and alkaloids (e.g., paynantheine, speciogynine, mitragynine, 7HMG) diminished alcohol intake (Gutridge et al., 2020). LKT (Wilson et al., 2020) and mitragynine were reported to lessen morphine withdrawal (Cheaha et al., 2017; Harun et al., 2020; Khor et al., 2011), with Hassan et al. (2020) suggesting that this mitragynine effect may resemble that produced by methadone and buprenorphine (Hassan et al., 2020). Additionally, mitragynine attenuated morphine dependence as well (Hemby et al., 2019; Jamil et al., 2013; Meepong and Sooksawate, 2019), and Yue et al. (2018) demonstrated a reduction by the compound of response rates in the model of heroin-induced Conditioned Place Preference (CPP) (Yue et al., 2018). Further, Fakurazi et al. (2013) found that mitragynine possesses the potential to reduce morphine tolerance in a chronic morphine administration model, defined by transcription factor CAMP response element binding (CREB)’s activation and the consequent increase in cAMP level’s expression (Fakurazi et al., 2013).

3.2.3 Other medical conditions

Twenty-two (15 in vivo in mice, rats or frogs, rabbits and cats, 7 in vitro) studies, plus four previously described to report also effects in pain (Macko et al., 1972; Mossadeq et al., 2009) and withdrawal or dependence (Khor et al., 2011; Kumarnsit et al., 2007a), provided evidence for kratom’s potential therapeutic use in the treatment of some conditions.

Both mitragynine and MS extracts (p.o. or i.p.) were found to produce several effects including gastroprotective action (Chitrakarn et al., 2018), inhibition of acid gastric secretion (Tsuchiya et al., 2002), and anti-inflammatory (Aziddin et al., 2005; Chitrakarn et al., 2018; Macko et al., 1972; Mossadeq et al., 2009), stress mitigating (Hazim et al., 2011; Khor et al., 2011; Vázquez López et al., 2017), anxiolytic-like (Hazim et al., 2014; Khor et al., 2011; Moklas et al., 2013) and antidepressant-like effects (Idayu et al., 2011; Kumarnsit et al., 2007a, 2007b), anorectic action (Chitrakarn et al., 2008; Grewal, 1932b; Kumarnsit et al., 2006, 2007b), antimutagen/anticancer (Ghazali et al., 2011; Goh et al., 2014), antioxidant (Goh et al., 2014; Grewal, 1932b; Parthasarathy et al., 2009; Yuniarti et al., 2020), and muscle relaxant effect (Chitrakarn et al., 2010). The extract had a more significant action in terms of muscle relaxation when compared to mitragynine (Chitrakarn et al., 2010).

Further, only mitragynine was found to have also dose-dependent anthelmintic activity (Abdul Aziz et al., 2012), antitussive (Macko et al., 1972), paramoecia killing action, anti-hypertensive, and anesthetic effects (Grewal, 1932b), while MS extracts showed to exert antibacterial (Juanda et al., 2019; Parthasarathy et al., 2009), bodyweight decreasing and dose-dependent anti diarrheal (Chitrakarn et al., 2008), antipsychotic-like (Vijeepallam et al., 2016), antipyretic effects (Salleh et al., 2011) and facilitation of learning (Senik et al., 2012).

For a complete overview, see Table 1.

3.2.4 Therapeutic application and safety issues in humans

Among the 18 clinical studies, three were experimental studies, with respectively an interventional, a prospective, and a randomized placebo-controlled, double-blind design. 15 were observational with a cross-sectional (N = 13) and a retrospective (N = 2) design. All these clinical studies were performed in Southern East Asia, and participants were kratom users.

Among the observational studies (Leong Bin Abdullah et al., 2019a, 2019b, 2020a, 2020b; Saref et al., 2019a, 2019b; Singh et al., 2014, 2015, 2018a, 2018b, 2018c, 2018d, 2019a, 2019b, 2019c), none reported evidence of therapeutic application. Safety issues related to chronic kratom use were shown in nine of these studies. Issues reported were high cholesterol level (Leong Bin Abdullah et al., 2020a), a slight increase in both HDL and LDL (cholesterol) values (Singh et al., 2018a), visual episodic memory or
| Author                        | Research Question                                                                 | Studied Compound (Dose, Route)                                                                 | Positive Control (Dose, Route)                                                                 | Animal, Tissue Type, Groups (Sample Size) | Clinical Model | Test/Measures                                                                 |
|------------------------------|-----------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------|------------------------------------------|---------------|------------------------------------------------------------------------------|
| Carpenter et al. (2016)      | Comparison on thermal nociception between MS articles and opioid agonists          | MG (30 mg/kg, i.p.) MSE (300 mg/kg, i.p.) MS alkaloids fraction (75 mg/kg, i.p.)               | MOR (10 mg/kg, i.p.) Oxycodone (3 mg/kg, i.p.)                                             | Sprague Dawley rats, 6 (9–10)            | Acute thermal pain             | HPT/Increase in latencies to perform an antinociceptive response             |
|                              |                                                                                   | MG (100 mg/kg, p.o.) MSE (300 mg/kg, p.o.)                                                   | Oxycodone (6 mg/kg, p.o.) //                                                                | Sprague Dawley rats, 4 (8–9)            | //                          | //                                                                           |
| Criddle (2015)               | Comparison between MS articles and opioid agonists                                | MG (30 mg/kg, i.p.) MSE (300 mg/kg, i.p.) MS alkaloids fraction (75 mg/kg, i.p.)             | MOR (10 mg/kg, i.p.) Oxycodone (3 mg/kg, i.p.)                                             | Sprague-Dawley rats, 6 (10)             | Acute thermal pain             | HPT/Increase in latencies to perform an antinociceptive response             |
|                              |                                                                                   | MG (100 mg/kg, p.o.) MSE (300 mg/kg, p.o.)                                                   | Oxycodone (6 mg/kg, p.o.) //                                                                | Sprague Dawley rats, 4 (8–10)           | //                          | //                                                                           |
| Fakurazi et al. (2013)*      | Enhancement of MG's analgesic action in combination with MOR                        | MG (15, 25 mg/kg, i.p.) MOR + MG (5 mg/kg + 15, 25 mg/kg, i.p.)                              | MOR (5 mg/kg, i.p.) Y/N                                                                      | ICR mice, 6 (7)                         | Acute thermal pain             | HPT/Increase in latencies to perform an antinociceptive response             |
| Foss et al. (2020)           | Effect on neuropathic pain                                                         | MG (1, 5, 10 mg/kg, i.p.) ND (ND)                                                           | N/Y                                                                                         | Male Sprague-Dawley rats, 4 (7–8)        | Alodynia oxaliplatin (6 mg/kg i.p.) induced; locomotor activity             | Mechanical sensitivity test/Reduction of paw withdrawal threshold; % of ambulatory counts in the VEH (0) |
|                              |                                                                                   | MG (30 mg/kg, i.p) ND (ND)                                                                  | //                                                                                          | Male Sprague-Dawley rats, 2 (7)          | //                          | % of ambulatory counts in the VEH (−)                                       |
| Hiranita et al. (2019)       | Effect on schedule-controlled responding and antinociception                      | MG (32, 5.6, 10, 17.8, 32, 56 mg/kg, i.p.)                                                   | ND (ND)                                                                                     | Sprague-Dawley rats, 2 (16)             | Operant procedures for food reinforcement; acute thermal pain               | Multiple cycles fixed ratio 10 schedules of food delivery/Reduction of schedule-controlled responding; HPT/Increase in latencies to perform an antinociceptive response (like MOR) |
|                              |                                                                                   | MG + MOR (32, 5.6, 10, 17.8, 32, 56 mg/kg + 32, 5.6, 10, 17.8, 32, 56 mg/kg, i.p.)          | ND (ND)                                                                                     | //                                       | //                          | Multiple cycle fixed ratio 10 schedule of food delivery (0), HPT (0) (MG 17.8 mg) |
| Author                  | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                      | Test/Measures                                                                                           |
|-------------------------|----------------------------------------------------------------------------------|--------------------------------|--------------------------------|---------|------------------------------------------|-------------------------------------|-------------------------------------------------------------------------------------------------------|
| **Antinociceptive Effects** |                                                                                  |                                |                                |         |                                          |                                     |                                                                                                         |
| Idid et al. (1998)      | Comparison of antinociceptive effect between MG, paracetamol and MOR             | MG (200 mg/kg, p.o.)          | MOR (5 mg/kg, p.o.)            | N/Y     | Albino mice, 4 (6)                      | Pain; acute thermal pain            | Acetic acid-induced writhing test/Inhibition of writhing constrictions; HPT, cold TFT/Increase in latencies to perform an antinociceptive response |
| Macko et al. (1972)     | MG pharmacology                                                                  | MG (92 mg/kg, p.o.)           | ND (ND)                        | ND/ND   | Mice, ND                                | Acute thermal pain                  | HPT/Increase in latencies to perform an antinociceptive response                                        |
|                         |                                                                                  | MG (92 mg/kg, s.c.)           | ND                            |         | Rat, ND                                 | //                                  | TFT/Increase in latencies to perform an antinociceptive response                                        |
|                         |                                                                                  | MG (ND, s.c.)                 | ND                            |         | //                                      | //                                  | //                                                                                                      |
| Matsumoto et al. (1996a)| Roles of central monoaminergic systems in the antinociceptive action             | MG (1, 3, 10 mg, i.c.v.)      | MOR (0.3, 1, 3 mg/mouse, i.c.v.) | N/Y     | ddY mice, 2 (40)                       | Acute thermal pain                  | TPT, HPT/Increase in latencies to perform an antinociceptive response                                  |
| Matsumoto et al. (1996b)| Antinociceptive effect                                                            | MG (3, 10, 30 mg/kg, i.p.)    | ND (ND)                        | N/Y     | ddY mice, 4 (10)                       | Acute thermal pain                  | TFT, HPT/Increase in latencies to perform an antinociceptive response                                  |
|                         |                                                                                  | MG (1, 3, 10 mg/mouse, i.c.v.)| ND                            |         | //                                      | //                                  | //                                                                                                      |
| Matsumoto et al. (2004)| Opioid effects                                                                    | 7HMG (2.5, 5, 10 mg/kg, s.c. or p.o.) | MOR (2.5, 5, 10 mg/kg, s.c.) or MOR (20 mg/kg, p.o.) | N/Y     | ddY mice, 3 (6)                       | Acute thermal pain                  | TFT/Increase in latencies to perform an antinociceptive response                                        |
|                         |                                                                                  | 7HMG (5, 10, 20 mg/kg, s.c. or p.o.) | MOR (5, 10, 20 mg/kg, s.c.) or MOR (20 mg/kg, p.o.) |         | //                                      | //                                  | HPT/Increase in latencies to perform an antinociceptive response                                        |
| Matsumoto et al. (2005)| Antinociceptive and opioid effects                                                | 7HMG (2.5, 5, 10 mg/kg, s.c.) | ND                             | N/Y     | ddY mice, 4 (6)                       | Acute thermal pain                  | TFT/Increase in latencies to perform an antinociceptive response                                        |
TABLE 1 (Continued)

| Author                  | Research Question                              | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model          | Test/Measures                                                                 |
|-------------------------|------------------------------------------------|--------------------------------|--------------------------------|---------|------------------------------------------|-------------------------|-------------------------------------------------------------------------------|
| **Antinociceptive Effects** |                                                |                                |                                |         |                                          |                         |                                                                                |
| Matsumoto et al. (2006) | Mechanism of antinociception and comparison with MOR | 7HMG (0.25, 0.5, 1.0, 2.0 mg/kg, s.c.) or MOR (1.25, 2.5, 5, 8 mg/kg, s.c.) | ND (ND)                      | Y/Y     | ddY-strain mice, 5 (7–8)                | Acute thermal pain       | TFT, HPT/Increase in latencies to perform an antinociceptive response          |
| Matsumoto et al. (2008) | MG derivative compounds’ effects               | MGM-9 (0.25, 0.5, 1, 2 mg/kg, s.c.) | MOR (1.25, 2.5, 5, 8 mg/ kg, s.c.) 7HMG (0.25, 0.5, 1.2 mg/kg, s.c.) | Y/Y     | ddY-strain mice, 5 (7–9)                | Acute thermal pain       | TFT, HPT/Increase in latencies to perform an antinociceptive response          |
|                         |                                                | MGM-9 (1, 2, 4, 8 mg/kg, p.o.) | MOR (25, 50, 100 mg/kg, p.o.) 7HMG (1, 2, 4 mg/kg, p.o.) | //       | //                                        |                          |                                                                                |
|                         |                                                | MGM-9 (0.025, 0.05, 0.1, 0.2 mg/kg, s.c.) | MOR (0.25, 0.5, 1 mg/kg, s.c.) 7HMG (0.05, 0.1 0.2, 0.4 mg/kg, s.c.) | //       | //                                        |                          |                                                                                |
|                         |                                                | MGM-9 (0.25, 0.5, 1, 2 mg/kg, s.c.) | MOR (25, 5, 10 mg/kg, p.o.) 7HMG (0.5, 1, 2, 4 mg/kg, p.o.) | //       | //                                        |                          |                                                                                |
| Matsumoto et al. (2014) | 7HMG derivatives’ potential effect on acute/chronic pain | MGM-15 (0.125, 0.25, 0.5, 1 mg/kg, s.c.) or (0.5, 1, 2, 4 mg/kg, p.o.) | ND (ND)                      | N/Y     | ddY-strain mice, 5 (8)                  | Acute thermal pain       | TFT/Increase in latencies to perform an antinociceptive response               |
|                         |                                                | MGM-16 (0.025, 0.05, 0.1, 0.2 mg/kg, s.c.) or (0.125, 0.25, 0.5, 1 mg/kg, p.o.) | ND (ND)                      | //       | //                                        |                          |                                                                                |
|                         |                                                | MGM-16 (0.1, 0.2, 0.4 mg/kg, s.c.) | ND (ND)                      | Y/Y     | ddY-strain mice, 5 (6–7)                | Neuropathic pain         | Sciatic nerve ligation induced thermal/mechanical hyperalgesia/Increase in paw withdrawal threshold |
|                         |                                                | MGM-16 (0.5, 1, 2 mg/kg, p.o.) | Gabapentin (100 mg/kg, p.o.) | //       | ddY-strain mice, 6 (6–7)                | //                      | –                                                                              |
| Author               | Research Question                      | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                  | Test/Measures                                                                  |
|----------------------|----------------------------------------|-------------------------------|-------------------------------|------------------------------------------|--------------------------------|--------------------------------------------------------------------------------|
| Mossadeq et al. (2009)* | Antinociceptive activity                | MS ME (50, 100, 200 mg/kg, i.p.) | ASA (100 mg/kg, i.p.) MOR (5 mg/kg, i.p.) | Y/N Sprague-Dawley rats, 6 (10)          | Pain                           | Formalin test/Inhibition of time spent in antinociceptive response              |
|                      |                                        | MS ME (50, 100, 200 mg/kg, i.p.) | ASA (100 mg/kg, i.p.) MOR (5 mg/kg, i.p.) | // Balb C mice, 6 (10)                  | Acute thermal and mechanical pain | HPT/Increase in latencies to perform an antinociceptive response; acetic acid-induced writhing test/Inhibition of writhing constrictions |
| Reanmongkol et al. (2007) | Effects on analgesic and behavioral activities | MS ME (50, 100, 200 mg/kg, p.o.) or MS Alk-E (5, 10, 20 mg/kg, p.o.) | MOR (10 mg/kg, p.o.) | N/Y Swiss mice, 5 (10) | Acute thermal pain | HPT/Increase in latencies to perform an antinociceptive response |
|                      |                                        | MS ME (50, 100, 200 mg/kg, p.o.) or MS Alk-E (5, 10, 20 mg/kg, p.o.) | MOR (10 mg/kg, p.o.) | // Wistar rats, 5 (6) |                          | TFT (0)                                                                          |
|                      |                                        | MS ME (50, 100, 200 mg/kg, p.o.) or MS Alk-E (5, 10, 20 mg/kg, p.o.) | Methamphetamne (1 mg/kg, i.p.) | // Swiss mice, 5 (10) | ND                           | Locomotor activity (0)                                                           |
|                      |                                        | MS ME (50, 100, 200 mg/kg, p.o.) or MS Alk-E (5, 10, 20 mg/kg, p.o.) | ND (ND) | // Swiss mice, 4 (10) | Pentobarbital-induced sleep | Sleeping time (0)                                                              |
| Sabetghadam et al. (2010) | Antinociceptive activity | MS Alk-E (5, 10, 20 mg/kg, p.o.) | MOR (5 mg/kg, s.c.) Aspirin (300 mg/kg, p.o.) | Y/Y Sprague-Dawley rats, 6 (5) | Acute thermal pain | HPT, TFT/Increase in latencies to perform an antinociceptive response            |
| Shemida et al. (2012) | Investigation on antinociceptive effect | MG (3, 10, 15, 30, 35 mg/kg, i.p.) | MOR (3 mg/kg, i.p.) | Y/N ICR mice, 7 (8) | Acute thermal pain | HPT/Increase in latencies to perform an antinociceptive response               |

(Continues)
| Author               | Research Question                                      | Studied Compound (Dose, Route)                                                                 | Positive Control (Dose, Route) | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                  | Test/Measures                                      |
|----------------------|-------------------------------------------------------|-----------------------------------------------------------------------------------------------|---------------------------------|----------------------------------------|----------------------------------|---------------------------------------------------|
| Stolt et al. (2014)  | Effects on analgesia and behavior                      | MG + paynantheine (0.5 + 0.025 mg/kg, p.o.) MG + paynantheine (2 + 0.1 mg/kg, p.o.) MG + paynantheine (4 mg/kg + 0.2 mg/kg, p.o.) | ND (ND)                         | WT mice and mu-opioid receptor KO mice, 8 (8–15) | Locomotor activity                | Locomotor activity recording/ Reduced time spent in (horizontal + vertical) activity |
|                      |                                                       | MG + paynantheine (2 + 0.1 mg/kg, i.p.) MG + paynantheine (4 mg/kg + 0.2 mg/kg, i.p.)              | //                              | WT mice and mu-opioid receptor KO mice, 6 (8–15) | Locomotor activity; anxiety; acute thermal pain | Locomotor activity recording/ Reduced time spent in (horizontal + vertical) activity; EPM test/Reduced time spent on open arms; HPT (0) |
|                      |                                                       | MG + paynantheine (10 mg/mouse + 0.5 mg/mouse, i.c.v.) MG + paynantheine (20 mg/mouse + 1 mg/mouse, i.c.v) | //                              | //                                      | Anxiety; acute thermal pain       | Locomotor activity recording/ Reduced time spent in (horizontal + vertical) activity; EPM test (ND); HPT/Increase in latencies to perform an antinociceptive response |
| Takayama et al. (2002)| Synthesis and opioid agonistic activities            | MG (ND, 75 nmol/mouse, i.c.v.) MG Pseudoindoxyl (ND, 12 nmol/mouse, i.c.v.)                     | MOR (ND, 9 nmol/mouse, i.c.v.)   | Mice, 4 (9–12)                          | Acute thermal pain               | TFT/Increase in latencies to perform an antinociceptive response |
| Thongpradichote et al. (1998) | Opioid receptor subtypes involved in the antinociceptive action | MG + antagonists (10 mg, i.c.v.)                                                                | MOR (3 mg, i.c.v.)               | ddY mice, 3 (7–9)                       | Acute thermal pain               | TPT, HPT/Increase in latencies to perform an antinociceptive response |
| Wilson et al. (2020) | LKT’s antinociception and liabilities                | LKT (45, 200, 1000, 2000, 4000 mg/kg, p.o.)                                                    | MOR (1, 3, 10, 60 mg/kg, i.p. or p.o.) | C57BL/6J mice, 5 (8)                    | Acute thermal pain               | 55°C warm-water tail-withdrawal assay/Increase in latency to remove the tail |
| Author                  | Research Question                                                                 | Studied Compound (Dose, Route)                                                                 | Positive Control (Dose, Route)                                                                 | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                                                                 | Test/Measures                                                                                       |
|------------------------|------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------|-------------------------------------------|---------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------|
| Cheaha et al. (2015)   | Effect on EW-induced physical and cortical hyperexcitabilities                     | MS Alk-E (60 mg/kg, intragastrically)                                                       | Fluoxetine (10 mg/kg, intragastrically)                                                       | Wistar rats, 3 (8–9)                   | ND                                                                              | EEG and EMG signal acquisition (0)                                                                 |
|                        |                                                                                    | MS Alk-E (60 mg/kg, intragastrically)                                                       | Fluoxetine (10 mg/kg, intragastrically)                                                       | (ED) Wistar rats, 3 (6-7);             | EW                                                                               | (Withdrawal induced) gamma activity in frontal and parietal cortex and locomotor activity/Attenuation |
|                        |                                                                                    |                                                                                               |                                                                                                 | (not EW) Wistar rats, 1 (6-7);          |                                                                                  |                                                                                                  |
|                        |                                                                                    |                                                                                               |                                                                                                 | (not ED) Wistar rats, 1 (6-7)          |                                                                                  |                                                                                                  |
| Cheaha et al. (2017)   | Effect on NAL-precipitated MOR withdrawal and neural signalling in the nucleus accumbens | MG (30, 90, 120 mg/kg, p.o.)                                                               | MS Alk-E (20, 40, 60, 80, 100 mg/kg, p.o.)                                                     | (MOR dependent) Swiss albino ICR mice, 9 (10-12) | NAL-precipitated MOR withdrawal                                                      | Jumping behavior and dry-wet fecal excretions/Reduction only with MS Alk-E                        |
|                        |                                                                                    | MS Alk-E (80 mg/kg, p.o.)                                                                   | MOR (15 mg/kg, i.p.)                                                                            | (MOR dependent) Swiss albino ICR mice, 3 (6-8) | ND; locomotor activity                                                             | LFP in nucleus accumbens (0); Spontaneous motor activity (0)                                |
| Fakurazi et al. (2013)*| Effect of the combination on MOR tolerance                                         | MG (15, 25 mg/kg, i.p.)                                                                   | MOR (5 mg/kg, i.p.) MOR + MG (5 mg/kg +15, 25 mg/kg, i.p.)                                   | ICR mice, 6 (7)                        | Tolerance and dependence                                                           | cAMP/CREB level in thalamus and cortex immunoblotting analysis/Down-regulation with MG + MOR     |
| Guttridge et al. (2020)| Potential reduction of alcohol intake                                               | MSE with 7HMG (10, 30 mg/kg−1, i.p.)                                                       | ND (ND)                                                                                         | C57BL/6 mice, 2 (8–12)                | Moderate- binge alcohol consumption                                                | Level of alcohol intake in two-bottle choice and intermittent, limited access paradigm/Decrease  |
|                        |                                                                                    | MSE without 7HMG (10, 30 mg/kg−1, i.p.) or paynantheine (10, 30 mg/kg−1, i.p.) or speciogynine (10, 30 mg/kg−1, i.p.) or 7HMG (1, 3, 10 mg/kg−1, i.p.) | ND (ND)                                                                                         | C57BL/6 mice, 2 (8–12)                | Locomotor activity                                                                | Locomotion assessment/Reduction with extracts, +/- with other compounds                          |

(Continues)
| Author            | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model | Test/Measures                                                                 |
|-------------------|-------------------------------------------------------------------------------------|--------------------------------|--------------------------------|---------|------------------------------------------|----------------|--------------------------------------------------------------------------------|
| Harun et al. (2020) | Potential therapeutic effect on MOR-dependence                                      | MG (1, 10, 30 mg/kg, i.p.)     | Buprenorphine (0.1, 0.3, 1.0 mg/kg, i.p.) | N/Y     | (MOR dependent) Sprague-Dawley rats, 3 (12) | NAL-precipitated MOR withdrawal | Food-maintained operant responding/ Attenuation of withdrawal, buprenorphine necessary |
| Hassan et al. (2020) | Effect on MOR withdrawal                                                           | MG (5, 10, 15, 30 mg/kg, i.p.) | ND (ND)                        | N/Y     | (MOR dependent) Sprague-Dawley rats, 5 (6); Sprague-Dawley rats, 1 (6) | Drug replacement treatment in MOR withdrawal | MOR withdrawal behaviors/Attenuation as methadone and buprenorphine |
| Hemby et al. (2019) | Abuse liability and therapeutic potential                                           | MG (25, 50, 100, 150 μg/inf, i.v.) | ND (ND)                        | Y/N     | Fischer 344 rats, 5 (8-9)                 | Human drug consumption | Substitution of MOR following MOR self-administration paradigm (0); Re-assessment of MOR self-administration/ Reduction |
|                    |                                                                                     | 7HMG (2.5, 5, 10, 20 μg/inf, i.v.) | ND (ND)                        | //      | Fischer 344 rats, 5 (8-9)                 |                             | Substitution of MOR following MOR self-administration paradigm/ Substitution; Re-assessment of MOR self-administration/ Increase |
| Jamil et al. (2013) | Effect on cAMP and mu-opioid receptor mRNA expression following chronic MOR treatment | MG (0.1, 1, 10, 50 μM, replacing the growth media in the cell culture) | MOR (0.1, 1, 10, 100 μM, replacing the growth media in the cell culture) | ND/ND   | Human neuroblastoma SK-N-SH cell, ND     | Tolerance and dependence | cAMP level upon forskolin stimulation/Reduction in the brief term, long term (−); mu-opioid receptor mRNA expression (0) |
|                    |                                                                                     | MG + MOR co- and pre-treatment (0.1, 1, 10, 50 μM + 50 μM, replacing the growth media in the cell culture) | NAL (50 μM, replacing the growth media in the cell culture) | //      | //                                        |                             | cAMP level upon forskolin stimulation in MOR co-treatment/ Reduction; mu-opioid receptor mRNA expression/Down-regulation reduction |
### TABLE 1 (Continued)

| Author | Research Question | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model | Test/Measures |
|--------|-------------------|--------------------------------|--------------------------------|---------|------------------------------------------|----------------|--------------|
| **Withdrawal and dependence** |
| Khor et al. (2011)* | Effect on anxiety behavior, cortisol level and expression of stress pathway-related genes in MOR withdrawal | MG (2 mg/L, exposure in water) | MOR (1.5 mg/L, exposure to water) | Y/N | WT zebrafish, 4 (20–25) | Stress-related anxiogenic behaviors in MOR withdrawal; MOR withdrawal-induced stress genes | Novel tank diving tests/Attenuation; Real-time PCR mRNA expression analysis/Reduction of genes expression |
| | | MG (1, 2 mg/L, exposure in water) | ND (ND) | // | WT zebrafish, 3 (20-21) | Anxiogenic behaviors in MOR withdrawal | Novel tank diving tests/Attenuation |
| | | MG (1, 2 mg/L, exposure in water) | MOR (1.5 mg/L, exposure in water) | ND (ND) | // | WT zebrafish, 5 (14–23) | MOR withdrawal-induced high cortisol level | Whole-body cortisol assay/Reduction |
| Kumarnsit et al. (2007a)* | Effect on EW | MS AE (100, 300, 500 mg/kg, p.o.) | ND (ND) | N/Y | Swiss albino mice, 4 (10) | Locomotor activity | Spontaneous motor activity (0) |
| | | MS AE (300 mg/kg, p.o.) | Not EW | // | (ED) Swiss albino mice, 3 (10) | EW | EW induced behaviors/Reduction |
| Meeppong and Sooksawate (2019) | Abuse liability and potential in the treatment of opioid addiction | MG + Sal (5, 10 mg/kg + ND, i.p.) Sal + VEH (ND, i.p.) | MG + MOR (5, 10 mg/kg + 5 mg/kg, i.p.) Sal + MOR (ND + 5 mg/kg, i.p.) | Y/N | Wistar rats, 5 (6–9) | Abuse liability | Acquisition of MOR-induced CPP/Suppression |
| | | MOR or Sal + MG (5 mg/kg or ND + 5, 10 mg/kg, i.p.) Sal + MG (ND + 5, 10 mg/kg, i.p.) | MOR + Sal (5 mg/kg + ND, i.p.) Sal + Sal (ND, i.p.) | // | Wistar rats, 4 (6–9) | // | Expression of MOR-induced CPP/Attenuation |
| | | (Chronic Sal) Sal + NAL (ND + 3 mg/kg, i.p.) | (Chronic Sal) Sal + Sal (ND, i.p.) | // | ICR mice, 4 (6–8) | NAL-induced chronic MOR withdrawal | Jumping behavior, Straub tail reaction/Reduction |
| Vijeepallam et al. (2019) | Effect on ethanol-seeking behavior | MS ME (50, 75, 100 mg/kg, p.o.) | Acamprosate (300 mg/kg, p.o.) Clozapine (1 mg/kg, p.o.) | Y/Y | Swiss albino mice, 7 (4) | Ethanol-induced CPP | Ethanol-place preference/Reversed effect |
| | | MS ME (50, 75, 100 mg/kg, p.o.) MS ME + Sal or VEH (100 mg/kg + ND, p.o.) | Acamprosate (300 mg/kg, p.o.) Clozapine (1 mg/kg, p.o.) Sal + VEH (ND, ND) | // | Swiss albino mice, 8 (10–11) | Drug self-administration | Difference between pre- and post-conditioning with ethanol/Reduction of the runtime to reach goal box, prolonged runtime in ethanol-conditioned |
| Author               | Research Question                          | Studied Compound (Dose, Route)                                                                 | Positive Control (Dose, Route)                                       | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                | Test/Measures                      |
|----------------------|--------------------------------------------|-----------------------------------------------------------------------------------------------|---------------------------------------------------------------------|------------------------------------------|-----------------------------------|------------------------------------|
| **Withdrawal and dependence** |                                            |                                                                                               |                                                                     |                                          |                                   |                                    |
| Wilson et al. (2020)* | LKT’s therapeutic potential to prevent NAL-precipitate MOR WD | LKT escalating doses (30, 35, 45, 60, 100, 125 mg/kg, p.o.) + LKT (25 mg/kg, p.o.) 7 days MOR (10, 15, 20, 30, 50, 60, 70, 75, 80 mg/kg, i.p.) + LKT (40 mg/kg, p.o.) 4 days MOR (10, 15, 20, 30, 50, 60, 70, 75, i.p.) + LKT continuing (100 mg/kg) + LKT (40 mg/kg, p.o.) 4 days MOR (10, 15, 20, 30, 50, 60, 70, 75, i.p.) + LKT taper (100, 80, 70, 60, 50 mg/kg, p.o.) + LKT (40 mg/kg, p.o.) | MOR escalating doses (10, 15, 20, 30, 50, 60, 70, 75 mg/kg, i.p.) + MOR (25 mg/kg, i.p.) | Mice, 6 (8–10)                       | NAL-precipitated MOR withdrawal | Opioid withdrawal behaviors/Attenuation |
| Yue et al. (2018)   | Abuse liability                            | MG (0.1, 0.3, 1, 3 mg/kg, i.v.)                                                               | Heroine (0.001, 0.003, 0.01, 0.03 mg/kg/inj, i.v.)                  | Sprague-Dawley rats (trained to self-administer methamphetamine), ND (6) | Methamphetamine induced CPP     | Response rates (0)                   |
| Abdul Aziz et al. (2012) | Anthelmintic properties                     | MG (0.2, 0.4, ND mg/mL, ND)                                                                  | ND (ND)                                                            | L3 stage larvae of strongyles, ND       | ND                               | Observation/Absence of motility - mortality |
| Aziddin et al. (2005) | Anti-inflammatory properties                | MS Alk-E (50 mg/kg, i.p.)                                                                    | ASA (20 mg/kg, i.p.)                                                | Sprague Dawley rats, 3(6)              | Acute inflammation               | Carrageenin-induced paw edema/Inhibition |
TABLE 1 (Continued)

| Author                      | Research Question               | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                                                                 | Test/Measures                                                                 |
|-----------------------------|---------------------------------|--------------------------------|--------------------------------|---------|-------------------------------------------|--------------------------------------------------------------------------------|--------------------------------------------------------------------------------|
| Chittrakarn et al. (2008)   | Effect on the gastrointestinal tract | MS ME (50, 100, 200, 400 mg/kg, p.o.) | Loperamide (6 mg/kg, p.o.) | N/Y     | Wistar rats, 6 (10)                      | Castor oil-induced diarrhea                                                   | Fecal matter and change in bodyweight/Reduction; intestinal transit and intestinal fluid/Inhibition |
|                             |                                 | MS ME (100, 200, 400 mg/kg, p.o.) | NAL (5 mg/kg, i.p.)         | //      | Wistar rats, 8 (10)                      | //                                                                             | Defecation frequency, total score, fecal weight (0); Change in bodyweight (0) |
|                             |                                 | MS ME (100, 200, 400 mg/kg, p.o.) | MOR (3 mg/kg, i.p.)         | //      | Wistar rats, 18 (10)                      | ND                                                                            | Bodyweight daily recording/Reduction; CCK blood levels from the orbital plexus (0); intestinal transit and intestinal fluid (0) |
|                             |                                 | MG (0.0156 mg/ml, addition to the preparation) | ND (ND)                         | N/N     | Wistar rats' isolated phrenic nerve and hemidiaphragm preparation, 5 (8) | ND                                                                            | Recording of neurally evoked twitch/Decrease; Recording of direct muscle twitch/Decrease |
|                             |                                 | MS ME (0.1, 0.25, 0.5, 1 mg/ml, addition to the preparation) + positive controls | Pancuronium (0.6 mmol, addition to the preparation) Succinylcholine (1.3 mmol, addition to the preparation) | //      | //                                        | //                                                                             | -                                                                                       |
|                             |                                 | MG (2 mg/ml, addition to the preparation) MS ME (10, 20, 40 mg/ml, addition to the preparation) | Xilocaine (0.10%, addition to the preparation) | N/N     | Wistar rat's isolated sciatic nerve preparation, 6 (6) | //                                                                            | Recording of compound action potential/Block                                         |

(Continues)
| Author                  | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                                                      | Test/Measures                                                                                                                                 |
|-------------------------|-----------------------------------------------------------------------------------|--------------------------------|--------------------------------|-------------------------------------------|----------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------|
| Chittrakarn et al. (2018) | Effect on the peptic ulcer and reflux esophagitis                                 | MS ME (200, 400 mg/kg, p.o.)  | MG (2 mg/kg, p.o.) Ranitidine (50 mg/kg, p.o.) | N/Y Wistar rats, 5(8)                      | WIR stress induced peptic ulcer                                      | Total gastric acid, gastric content pH, ulcer index, histopathological examination/Reduction of ulcer index, moderate regeneration of ulcerous lesions |
|                         |                                                                                   | MS ME (200, 400 mg/kg, p.o.)  | Omeprazole (20 mg/kg, p.o.)       | Wistar rats, 4 (8) ASA treatment or ET-induced peptic ulcer |                                                                         | //; only moderate regeneration of ulcerous lesions in ET model                                                                       |
|                         |                                                                                   | MS ME (200, 400 mg/kg, p.o.)  | Sucralfate (500 mg/kg, p.o.)      |                                                                         | Reflux esophagitis                                                    | //; only moderate regeneration of ulcerous lesions in ET model                                                                       |
| Ghazali et al. (2011)   | Potential (anti) mutagenic activity                                                | MS AE (3.125, 12.5, 50 mg/ml, addition to bacterial culture) | 2-NF or 2-AA (0.1 ml, addition to bacterial culture) | Y/Y Salmonella typhimurium strain TA 98 cell, 2.5 x 10^8 cells/ml | 2-NF, NaN3 and 2-AA induced mutagenicity                               | Ames (Antimutagenicity) test/ + 59 metabolic activator (+); without (0)                                                                   |
|                         |                                                                                   | MS AE (3.125, 12.5, 50 mg/ml, addition to bacterial culture) | NaN3 or 2-AA (0.1 ml, addition to bacterial culture) |                                                                         |                                                                         |                                                                                                                                           |
| Goh et al. (2014)       | Antioxidant value and anticancer functions                                         | MG (6.2, 12.5, 25, 50, 100, 200 mM, addition to the cell culture) | SRM (6.2, 12.5, 25, 50, 100, 200 mM, addition to the solution) BA (//) 5-FU (//) | N/Y Cancer normal cell lines, 96 well μtitre plates | ND                                                                      | MTT cell antiproliferation assay/Cytotoxicity                                                                                              |
|                         |                                                                                   | MG (0.032, 0.0625, 0.125, 0.25, 0.5, 1 mg/ml, addition to the cell culture) | SRM (0.032, 0.0625, 0.125, 0.25, 0.5, 1 mg/ml, addition to the solution) Quercetin (//) BHT (//) | N/N DPPH solution, ND | DPPH radicals scavenging assay/Reduction of DPPH free radicals |                                                                                                                                           |
|                         |                                                                                   | MG (0.5, 1, 2, 4 mM, addition to the cell culture) | SRM (0.5, 1, 2, 4 mM, addition to the solution) Quercetin (//) BHT (//) | ABTS solution + trolox, ND | ABTS antioxidant assay/High TEAC                                     |                                                                                                                                           |
|                         |                                                                                   | MG (0.0625, 0.125, 0.25, 0.5, 1 mg/ml, addition to the cell culture) | SRM (0.0625, 0.125, 0.25, 0.5, 1 mg/ml, addition to the solution) Quercetin (//) BHT (//) | FeCl3 solution, ND | FRAP (+)                                                              |                                                                                                                                           |
| Author        | Research Question       | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | Animal, Tissue Type, Groups (Sample Size) | Clinical Model | Test/Measures                              |
|--------------|-------------------------|--------------------------------|--------------------------------|------------------------------------------|----------------|-------------------------------------------|
| Grewal (1932b) | MG pharmacology         | MG fumarate (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | N/N | Paramecia, ND | ND/Death |
|              |                         | MG fumarate (1:250, 1:330, 1:500, 1:1000, 1:200000, dilution) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Staphylococcus Albus and B. coli, ND | ND/No growth at the highest dilution |
|              |                         | MG fumarate (1:20000-1:100000, dilution) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | ND/Decreased amplitude of movements and muscle tone |
|              |                         | MG fumarate (15–20 mgm, i,v.) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | Intestinal movement by balloon method/Decrease |
|              |                         | MG fumarate (ND, dilution) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | ND/Muscle relaxation |
|              |                         | MG fumarate (1:15,000, ND) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | – |
|              |                         | MG fumarate (1:10,000, dilution) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | Perfused heart trough hepatic vein/Reduction of contraction |
|              |                         | MG fumarate (2.5, 5 mg, i,v.) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | ECG, pulmonary artery pressure, blood pressure/ Fall of blood pressure with an increase in cardiac output |
|              |                         | MG fumarate (1.0%, 0.1%, 0.01%, instillation) | Quinine (1:2000, 1:4000, 1:7000, 1:10000, 1:200000, dilution) | ND (ND) | Ringer solution (ND) | ND/Local anesthetic effect |
| Author          | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                          | Test/Measures                                                                 |
|-----------------|------------------------------------------------------------------------------------|-------------------------------|-------------------------------|---------|------------------------------------------|------------------------------------------|--------------------------------------------------------------------------------|
| Hazim et al.    | Effects on exploratory behavior, short-term memory and motor coordination          | MG (20, 40, 80 mg/kg/p.o.) MS Alk-E (20, 40, 80 mg/kg/p.o.) | Amphetamine (10 mg/kg, i.p.) Scopolamine (1 mg/kg, i.p.) | N/Y     | Swiss Albino mice, 9 (9)                 | Spontaneous locomotor activity           | Y-maze test/Increased spontaneous locomotor activity and novelty-induced rearing behavior |
| Hazim et al.    | Effect on anxiety-related behaviors                                                | MG (10, 20, 40 mg/kg, p.o.)   | Diazepam (10 mg/kg, p.o.)     | N/Y     | Sprague–Dawley rats, 5 (8)               | Anxiety                                 | OF test/Increase in CZE and % CZT; EPM test/Increase in %OAE and %OAT           |
| Idayu et al.    | Antidepressant effect and effect towards the HPA neuroendocrine system            | MG (5, 10, 30 mg/kg, i.p.)    | Fluoxetine (20 mg/kg, i.p.) Amtriptyline (10 mg/kg, i.p.) | N/Y     | ICR mice, 6 (8)                          | Depression; FST or TST induced stress    | FST, TST/Reduction of immobility time (comparable to fluoxetine and amitriptyline); Corticosterone serum level/Reduction |
| Juanda et al.   | Potential antibacterial activity                                                   | MS ME (3%, 6%, 9%, 12%, 15%, 18%, 21%, 24%, 27%, 30% of 62.27 g, addition to the suspension) | ND (ND) | Aeromonas hydrophilla, ND                | ND                                        | Agar diffusion method using the NA medium/Growth inhibition and death |

Other effects

| Author          | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                          | Test/Measures                                                                 |
|-----------------|------------------------------------------------------------------------------------|-------------------------------|-------------------------------|---------|------------------------------------------|------------------------------------------|--------------------------------------------------------------------------------|
| MG (1: 10,000, ND) | ND (ND)                             | Frog’s sciatic nerve, ND       | ND                            | Excitability and conduction (0) |
| MG (5 mgm, Inj)     | Urethane (ND)                        | Rabbits, mice and frogs, ND   | ND                            | Faradic stimulation of the central end of right vagus/ Reduction of the threshold, vasodilatation |
| MG (2.5, 5 mg, Inj)   | ND (ND)                             | Injection into the jugular vein, ND | ND                            | Stimulaion of the chorda tympani for 10 s/ Autonomic effect |
| MG (15 mg/kg, ND)    | ND                                  | Rabbits, ND                   | ND                            | Observation/Weight loss, coat deterioration |

Hazim et al. (2011)

Hazim et al. (2014)

Idayu et al. (2011)

Juanda et al. (2019)
| Author                      | Research Question                                                                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | Clinical Model | Test/Measures                                    |
|-----------------------------|-------------------------------------------------------------------------------------|--------------------------------|-------------------------------|----------------|--------------------------------------------------|
| Other effects               |                                                                                     |                                |                               |                |                                                  |
| Khor et al. (2011)*         | See WD section                                                                      |                                |                               |                |                                                  |
| Kumarnsit et al. (2006)     | Effect on food, water intake and bodyweight                                         | MSE (15, 30, 45, 50 mg/kg, p.o.)| Imipramine (40 mg/kg, i.p.)   | Wistar rats, 6 (10) | ND  Food, water intake and bodyweight measurement/Decrease |
|                             |                                                                                     | MSE (40 mg/kg, i.p.)            | ND (ND)                       | Wistar rats, 2 (10)| //                                                  |
| Kumarnsit et al. (2007a)    | Effect on EW                                                                        | MS AE (100, 300, 500 mg/kg, i.p)| ND (ND)                       | ND             | TST/Reduced immobility duration                  |
| Kumarnsit et al. (2007b)    | Stimulant effect on the dorsal raphe nucleus and antidepressant-like activity        | MSE (60, 90 mg/kg, i.p)         | ND (ND)                       | Wistar rats, 3 (7-10) | Depression                       |
|                             |                                                                                     | MSE (40 mg/kg, i.p)             | //                            | Wistar rats, 3 (7)| ND                                                  |
| Macko et al. (1972)*        | MG pharmacology                                                                     | MG (ND, p.o.)                   | Codeine (ND, p.o.)            | Dogs, ND | Cough                                           |
|                             |                                                                                     | MG (ND, p.o.)                   | ND                            | Rat, ND        | Inflammation                                    |
|                             |                                                                                     |                                |                               |                | Electromagnetically evoked cough reflex/Inhibition |
| Moklas et al. (2013)        | Effect on cognitive and behavioral performances                                      | MS ME (10 mg/kg, 30 mg/kg, 100 mg/kg, p.o.) | MS AE (10 mg/kg, 30 mg/kg, 100 mg/kg, p.o.) | Sprague Dawley rats, 7 (ND) | Anxiety; locomotor activity; cognitive performance |
|                             |                                                                                     | MS ME (10 mg/kg, 30 mg/kg, 100 mg/kg, p.o.) | Scopolamine (ND, i.p.)        | ICR mice, 8 (ND) | Rota-rod performance                            |
|                             |                                                                                     |                                |                               |                | Time of falling from the revolving rotarod/Sedative effects |
| Mossadeq et al. (2009)*     | Anti-inflammatory activity                                                           | MS ME (50, 100, 200 mg/kg, i.p.)| ASA (100 mg/kg, i.p.)         | Sprague-Dawley rats, 5 (8) | Acute and chronic inflammation                    |
|                             |                                                                                     |                                |                               |                | Carrageenin-induced paw edema and cotton pellet-induced granuloma/Inhibition |
| Parthasarathy et al. (2009)  | Antioxidant and antibacterial activity                                               | MS AE or ME or Alk-E (0.6, 0.5, 0.4, 0.3, 0.2, 0.1, 0.05, 0.01 mg/ml, addition to the solution) | BHT (ND, addition to the solution) | DPPH solution, 2.5 ml | Antioxidant activity                              |
|                             |                                                                                     |                                |                               |                | DPPH radicals scavenging assay/Inhibition          |

(Continues)
| Author          | Research Question                  | Studied Compound (Dose, Route)                          | Positive Control (Dose, Route)          | SAL/VEH | Animal, Tissue Type, Groups (Sample Size) | Clinical Model                         | Test/ Measures                          |
|-----------------|------------------------------------|-------------------------------------------------------|----------------------------------------|---------|----------------------------------------|----------------------------------------|----------------------------------------|
| Salleh et al. (2011) | Potential antipyretic properties    | MS crude ME (50 mg/kg, 100 mg/kg, 200 mg/kg, i.p.)   | Ketoprofen (1 mg/kg, i.p.)             | N/Y     | BALB/c mice, 5 (6)                    | Brewer’s yeast-induced pyrexia          | Rectal temperature recording/Reduction |
| Senik et al. (2012) | Effect on cognitive function        | MS ME (100, 500, 1000 mg/kg, p.o.)                    | MOR (430 mg/kg, p.o.) Piracetam (500 mg/kg, p.o.) | N/Y     | (Albino) Sprague-Dawley rats, 6 (10)  | Learning; long term memory             | One-way PA test and two-way active avoidance test/ Learning acquisition (+) |
| Tsuchiya et al. (2002) | Effect on gastric acid secretion    | MG (30 mg, i.c.v.) MG (3, 10, 30 mg, i.c.v.)          | ND (ND)                                | N/Y     | Wistar rats, 2 (7-9)                  | ND                                     | Basal gastric acid secretion (0)       |
| Vázquez López et al. (2017) | Action on stress                  | MG + paynantheine solution (1 mg/kg + 0.05 mg/kg, i.p.) | Ethanol (3 v/v%, i.p.)                | N/N     | WT mice, 2 (9-15)                     | ND                                     | Vocalization threshold in response to electrical stimulation of the tail root/ Basal response current (mA) and AI (0) |
|                 |                                     | MG + paynantheine solution (1 mg/kg + 0.05 mg/kg, i.p.) | Ethanol (3 v/v%, i.p.)                |         | (Stressed and not stressed) WT mice, 2 (9-15) | Restraint–stress-induced analgesia     | Vocalization threshold in response to electrical stimulation of the tail root/Reduction of AI |
| Author                     | Research Question                 | Studied Compound (Dose, Route) | Positive Control (Dose, Route) | Animal, Tissue Type, Groups (Sample Size) | Clinical Model | Test/Measures                   |
|----------------------------|-----------------------------------|-------------------------------|--------------------------------|------------------------------------------|----------------|-------------------------------|
| Vijeepallam et al. (2016)  | Antipsychotic-like effect         | MS ME (50, 75, 100, 125, 250, 500 mg/kg, p.o.) | ND (ND) | Swiss albino mice, 8 (8) | (ApoMOR-induced) climbing behavior | Climbing behavior index/Attenuation |
|                           |                                   | MS ME (75, 100 mg/kg, p.o.)   | //                              | Swiss albino mice, 4 (8) | (Haloperidol-induced) catalepsy | Mouse bar test/Potentiation of catalepsy |
|                           |                                   | MS ME (50, 75, 100, 125, 250, 500 mg/kg, p.o.) | Clozapine (1 mg/kg, p.o.) | Swiss albino mice, 9 (8) | (Ketamine-induced) social withdrawal | Social behavior observation/Social interaction (+) and withdrawal reduction |
| Yuniarti et al. (2020)    | Antioxidant activity              | Kratom leaf ethanol extract (20, 40, 60, 80 and 100 μg/ml, ND) | Vitamin C | N/N | DPPH solution, ND | Antioxidant activity test | DPPH radicals scavenging assay, + or - Vitamin C/High reduction of DPPH free radicals |

Note: %OAE: open arm entries; %OAT: time spent on open arms; %CZT: time spent in central zone; //: same data as above; -: impairment; 0: no effect.

Abbreviations: 2-AA, 2-aminoanthracene; 2-NF, 2-Nitrofluorene; 5-FU, 5-Fluoruracil; 5-HETE, 5-hydroxy-6,11,14-eicosatetraenoic acid; 7HMG, 7-hydroxymitragynine; ABTS, 2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulphonic acid); AE, aqueous extract; AI, analgesic index; Alk-E, alkaloid extract; ASA, acetylsalicylic acid; BHT, butylated hydroxytoluene; CAMP, cyclic adenosine monophosphate; CCK, cholecystokinin; CLAMS, comprehensive lab animal monitoring system; CFU, colony forming units; CP, Conditioned place preference; CREB, cAMP response element binding; CZE, number of entries in central zone; DPPH, 1,1-diphenyl-2-picryl hydrazylfree; ED, ethanol dependent; EPM, elevated plus-maze; ET, ethanol treatment; EW, ethanol withdrawal; FR, fixed-ratio; FRAP, ferric reducing antioxidant power assay; FST, forced swim test; HPA, hypothalamic-pituitary-adrenal; HPT, hot plate test; i.c.v., intracerebroventricular; i.p., intraperitoneal; i.v., intravenous; Inj, injection; KO, knock-out; LKT, lyophilized kratom tea; LFP, local field potential; ME, methanol extract; MG, mitragynine; MGM-15, (E)-methyl 2-((2S,5S,7aS,12aR,12bS)-3-ethyl-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindol[2,3-a]quinolin-2-yl)-3-methoxycarlylate; MGM-16, (E)-methyl 2-((2S,5S,7aS,12aR,12bS)-3-ethyl-9-fluoro-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindol[2,3-a]quinolin-2-yl)-3-methoxycarlylate; MGM-9, (E)-methyl 2-((2S,5S,7aS,12aR,12bS)-3-ethyl-9-fluoro-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindol[2,3-a]quinolin-2-yl)-3-methoxycarlylate; MOR, morphine; MS, Mitragyna speciosa; MSE, Mitragyna speciosa extract; MTT, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide; NAL, naloxone; NaN3, sodium azide; OF, open field; p.o., per oral; PA, passive avoidance; RA, retinoid acid; s.c., subcutaneous; SAL, saline solution; SRM, silane reduced analogue; TEAC, high trolox equivalent antioxidant capacity; TFT, tail flick test; TPT, tail pinch test; TST, tail suspension test; VEH, vehicle; WD, withdrawal and dependence; WIR, water immersion restraint; WT, wild type.

*Record with more than 1 evidence, the content is reported in the specific section.
learning impairment (Singh et al., 2019c), severe-to-moderate dependence (Singh et al., 2014), with severe dependence negatively affecting physical well-being (Leong Bin Abdullah et al., 2019a). Concerns during kratom cessation were physical (e.g., muscle spasms, pain, watery eyes and nose, fever, diminished appetite, gastrointestinal effects-diarrhea, constipation-, and severe fatigue) (Singh et al. 2014, 2018d, 2019a), and psychological withdrawal symptoms, for example, sleep problems, restlessness/nervousness, anger (Singh et al., 2014, 2018d), and mild levels of anxiety and depression (Singh et al., 2018c). Some of these adverse effects were described as linked to a greater kratom tea/juice daily consumption (Singh et al., 2018a, 2018c, 2018d, 2019a) or a major daily use frequency (Leong Bin Abdullah et al., 2020a).

Further, evidence of severe psychosis (Leong Bin Abdullah et al., 2019b) and electrocardiogram (ECG) alterations (Leong Bin Abdullah et al., 2020b) was found to be not related to kratom use. Additionally, kratom was not found to have a negative impact on life quality (Leong Bin Abdullah et al., 2019a), nor causing any alterations of hepatic (Leong Bin Abdullah et al., 2020a) or other biochemical parameters (Singh et al., 2018a), hormones levels (testosterone, FSH and LH; Singh et al., 2018b), social functioning in a traditional setting (Singh et al., 2015), motor function, and cognitive profile concerning attention, working memory, and executive functions (Singh et al., 2019c).

Among the experimental studies, evidence of potential therapeutic applications was shown in two studies. In fact, in the interventional study performed by Grewal (1932a), participants (N = 5) were orally administered mitragynine acetate (0.05 g–50 mg) or MS powdered leaves (0.65, 1.3 g), and kratom was found to reduce heat sensitivity and electrical resistance of the skin, to improve muscular work, and to cause dilatation of the skin blood vessels (Grewal, 1932a). Furthermore, some safety issues were reported by Grewal (1932a), such as giddiness, slight sight’s alterations and nystagmus, muscle tenseness, pupils contraction, hand/tongue’s tremor, stomach irritation and nausea, sleepiness sensation, and distorted motor coordination at higher doses (Grewal, 1932a). However, the adverse effects reported by Grewal (1932a) came from a small sample (N = 5) and cannot be generalized.

Vicknasingam et al. (2020) did not report any safety issues in a recent Randomized Controlled Trial (RCT) with participants being administered three kratom decoction drinks (mitragynine dose is not described). They found that kratom increased tolerance to discomforts nor withdrawal symptoms for at least 20 h (Vicknasingam et al., 2020). Finally, Trakulsrichai et al (2015) performed a prospective study on 10 participants, which were administered one dose (range 6.25–11.5 mg) per day for one week and a final dose on the eighth day (range 6.25–23 mg). This study did not show any therapeutic application but, in the absence of any serious adverse effect, reported some minor safety issues after kratom administration, such as a temporary blood pressure/heart rate increase and tongue numbness. Other details are provided in Table 2.

### 4 DISCUSSION

To our knowledge, this is the first systematic review that provides an overview of (pre)clinical evidence of mitragynine/kratom therapeutic use and safety issues in humans. Among the records included in this analysis (N = 75), 24% provided data in humans, while 76% supported its potential therapeutic use in the treatment of either acute and chronic pain (41%), substance use disorders (25%), such as morphine withdrawal and dependence, ethanol withdrawal, seeking behavior and intake; and other medical conditions based on several kratom effects (46%). Two out of the 18 clinical studies reported evidence of potential therapeutic application in pain. In contrast, some issues chronic kratom use related, such as learning impairment, alterations of cholesterol level, dependence, and withdrawal symptoms, were reported in 50% of them.

Many plant-based medicines, including kratom, have historically been used in tropical regions to treat common health problems (Brown et al., 2017). However, over the years, its use has become diffuse also in Western countries for both recreational and self-medicating purposes. Among the latter, the most commonly reported by users is pain relief (Grundmann, 2017; Schimmel et al., 2020; Singh et al., 2020).

The antinoceptive effects of kratom preparations, such as mitragynine (Carpenter et al., 2016; Criddle, 2015; Fukurazi et al., 2013; Foss et al., 2020; Hirani et al., 2019; Ildid et al., 1998; Macko et al., 1972; Matsumoto et al., 1996a, 1996b; Shaima et al., 2012; Thongpradichote et al., 1998), LKT (Wilson et al., 2020), MG Pseudoindoxyl (Takeyama et al., 2002), MGM-9 (Matsumoto et al., 2008), MGM-15 and MGM-16 (Matsumoto et al., 2014), and 7HMG (Matsumoto et al., 2004, 2005, 2006) recently suggested to be the key mediator of mitragynine's analgesic effects (Kruegel et al., 2019), may be considered as preclinical evidence of kratom's potential therapeutic use in pain treatment and would explain why many users claim this benefit. Further, this therapeutic application is supported by two clinical studies that found kratom to reduce pain sensitivity (Grewal, 1932a; Vicknasingam et al., 2020). According to the evidence reviewed in this paper, kratom was reported to exert antinociceptive effects through a multimodal regulation. This is suggested to involve spinal and supraspinal delta-1, mu-1 (Matsumoto et al., 2014; Shaima et al., 2012; Thongpradichote et al., 1998), and potentially kappa-opioid receptors (Wilson et al., 2020) together with α-adrenergic receptors (Foss et al., 2020). Other suggested mechanisms underlying these analgesic properties would include descending noradrenergic and serotonergic systems (Matsumoto et al., 1996a), Fos expression in the raphe nucleus (Kumarnsit et al., 2007b), neuronal Ca2+ channels blockade (Matsumoto et al., 2005; Takeyama et al., 2002), and inhibition of some hyperalgesia mediators involved in anti-inflammatory processes (Mossadeq et al., 2009). In a double connection, it is also suggested that the inhibition of active pain substances release (Aziddin et al., 2005) and a decreased COX-2 mRNA/prostaglandin E2 production (Utar et al., 2011) would mediate kratom's anti-inflammatory effects, which we found in some studies (Aziddin et al., 2005; Chitrakarn et al., 2018; Macko et al., 1972; Mossadeq et al., 2009). We also found kratom to show some actions suggested to
| Author Place | Participants (N, Gender) | Control Group (N, Gender) | Design | Research Question | Studied Compound (Dose, Route) | Administration Time (n) | (Majority of Participants) kratom Daily Consumed (glasses)/times Daily | Average MG Content X day | Test/Measures | Safety issues (YES, NO) |
|--------------|--------------------------|---------------------------|--------|-------------------|-------------------------------|------------------------|---------------------------------------------------------------|------------------------|----------------|---------------------|
| **Interventional Studies** |
| Grewal (1932a) | ND Volunteers (5, male) | ND | ND | Interventional study | MG acetate (0.05 g, p.o.) | 1 (N = 3); 2 (N = 2) | (ND)/ND | ND | Produced symptoms (−) | YES |
| | | | | | MG acetate (50 mg, p.o.) | 1 (N = 3); 2 (N = 2) | (ND)/ND | ND | Choice reaction time (+); heat tolerance (+); weight lift test (+); steadiness (+); dotting test (+/−); electrical skin resistance (+/−); vision test (0) | YES |
| | | | | | MS powdered leaves (0.65, 1.3 g p.o.) | 1 (N = 3); 2 (N = 2) | (ND)/ND | ND | Produced symptoms (−) | YES |
| | | | | | MS powdered leaves (0.65, 1.3 g p.o.) | 1 (N = 3); 2 (N = 2) | (ND)/ND | ND | Choice reaction time (−); heat tolerance (change: −); weight lift test (+); dotting test (0); electrical skin resistance (−) | YES/NO |
| Trakulsrichai et al. (2015) | Thailand Chronic regular kratom users (10, male) | ≥6 months | ND | Prospective study | PK of MG, blood pressure, and pulse rate change after taking kratom | MG (6.25–11.5 mg, p.o. for 7 days + final dose range 6.25–23 mg, p.o.) | Daily (7 days) + 8th day | (ND)/1–9 | ND | Blood exams and urine samples (0); observations (−); tongue numbness; safety and vital signs (−) | YES |
| Vicknasingam et al. (2020) | Malaysia Regular kratom users (26, male) | ≥12 months | ND | Randomized placebo-controlled, double-blind study | Evaluation of changes in pain tolerance, physiologic responses, and in potential withdrawal signs or symptoms | Kratom decoction drink (approximating MG concentration levels found in field decoctions) | 3 during the day | (ND)/multiple | ND | CPT (+); blood samples and vital signs (0); COWS (0); subjective symptoms (0) | NO |
| Author | Place | Participants (N, Gender) | K-Use (Time) | Control Group (N, Gender) | Design | Research Question | Studied Compound (Dose, Route) | Administration Time (n) | (Majority of Participants) Kratom Daily Consumed (glasses)/times Daily | Average MG Content X day | Test/Measures | Safety issues (YES, NO) |
|--------|-------|--------------------------|--------------|---------------------------|--------|------------------|-----------------------------|------------------------|-------------------------------------------------|----------------------|----------------|------------------------|
| Leong Bin Abdullah et al. (2019a) | Malaysia | Regular kratom users (150, male) | ≥1 year | ND | Cross-sectional survey | Quality of life of kratom users and its associated factors | ND | ND | >3 glasses/≥4 times daily | ND | WHOQOL-BREF (0–100) (−) | YES |
| Leong Bin Abdullah et al. (2019b) | Malaysia | Regular kratom users (150, male) | ≥1 year | ND | Cross-sectional clinical survey | Prevalence/severity of psychosis/psychotic symptoms in users and the evaluation of associations between K-use and the occurrence of psychotic symptoms | ND | ND | ≥4 glasses/>3 | ND | BPRS, MINI, DSMV (−) | NO |
| Leong Bin Abdullah et al. (2020a) | Malaysia | Regular kratom users (100, male) | ≥1 year | Healthy subjects, no K-use (100, male) | Analytical cross-sectional study | Fasting lipid profile of users and its associations with K-use | ND | ND | ≥4 glasses/>3 | ND | Serum TG and HDL (0); serum TC and LDL (−77); liver parameters (0) | YES |
| Leong Bin Abdullah et al. (2020b) | Malaysia | Regular kratom users (100, male) | ≥1 year | Healthy subjects, no K-use (100, male) | Analytical cross-sectional study | Prevalence of ECG abnormalities and QTc intervals in regular kratom users | ND | ND | ≥4 glasses/ND | 434.28 mg | ECG sinus tachycardia and borderline QTc intervals (no link with K-use) | NO |
| Saref et al. (2019a) | Malaysia | Illicit drug users with current K-use (260, male) | ≥7 days | ND | Cross-sectional study | Self-report relationship between kratom initiation, illicit drugs use, and HIV risk behaviors | ND | ND | ≥2 glasses/≤2 | ND | Face-to-face interview drug use (+) | NO |
| Saref et al. (2019b) | Malaysia | Illicit opioid users with current K-use (163, male) | ≥7 days | ND | Cross-sectional study | Self-reported prevalence and severity of opioid-related adverse effects after kratom initiation | ND | ND | ≥3 glasses/ND | 214.29 mg | Face-to-face interview drug use (+) | NO |
| Singh et al. (2014) | Malaysia | Regular kratom users (293, male) | >1–3 years | ND | Cross-sectional study | Kratom dependence, withdrawal, and craving | ND | ND | ≥3.5 glasses/>3 | 276.5 mg | LDQ/severe (>50%)/moderate dependence (45%); MWC (-); MCQ short form/high craving (23%), low craving (77%) | YES |
| Singh et al. (2015) | Malaysia | Regular kratom users (293, male) | ≥6 months | ND | Cross-sectional survey | Kratom effects on social functioning | ND | ND | ND | ND | ASI (0) | NO |
| Author                | Place        | Participants (N, Gender) | K-Use (Time) | Control Group (N, Gender) | Design                        | Research Question                                                                 | Studied Compound (Dose, Route) | Administration Time (n) | (Majority of Participants) | Average MG Content X day | Test/ Measures                  | Safety issues (YES, NO) |
|---------------------|-------------|--------------------------|-------------|---------------------------|-------------------------------|---------------------------------------------------------------------------------|-------------------------------|------------------------|--------------------------|--------------------------|-----------------------------|--------------------------|
| Singh et al. (2018a) | Malaysia    | Regular kratom users (58, male) | ≥ 2 years   | Healthy subjects, no K-use (19, male) | Cross-sectional study         | Kratom effects on hematological and clinical-chemistry                           | ND                            | ND                     | ≥3.5/2                   | 76.3–114.8 mg            | Full-blood test/ Hematological (0) and biochemical parameters (0) | YES                      |
| Singh et al. (2018b) | Malaysia    | Regular kratom users (19, male) | ≥ 2 years   | ND                        | Cross-sectional study         | Kratom effects on testosterone levels after long-term K-use                      | ND                            | ND                     | 3.5/ND                   | 76.23–94.15 mg            | Full-blood test/ Hematological and biochemical parameters (0); full-blood test/ testosterone, FSH, LH levels (0) | NO                       |
| Singh et al. (2018c) | Malaysia    | Regular kratom users (150, male) | ≥ 1 year    | ND                        | Retrospective study           | Severity of kratom withdrawal symptoms (anxiety and depression)                  | ND                            | ND                     | ≥4/2                     | ND                       | BDI (−); BAI (−)            | YES                      |
| Singh et al. (2018d) | Malaysia    | Regular kratom users (170, male) | > 2 years   | ND                        | Cross-sectional study         | Severity of kratom withdrawal symptoms (pain, sleep problems)                    | ND                            | ND                     | ≥3.5/2                   | 76–115 mg                | BPI (−); PSQI (−)           | YES                      |
| Singh et al. (2019a) | Malaysia    | Regular kratom users (125, male) | ND          | ND                        | Retrospective study           | Constipation prevalence from K-use and fatigue severity during kratom cessation | ND                            | ND                     | ≥3/ND                    | ND                       | CAS (−); FSS (−)            | YES                      |
| Singh et al. (2019b) | Malaysia    | Regular kratom users (62 male, 1 female) | > 2 years   | No kratom users (25, male) | Cross-sectional study         | Kratom effects on cognitive functions                                            | ND                            | ND                     | ≤3 glasses/1–3           | 72.5–74.9 mg              | CANTAB: PAL (−); MOT (0); DMS (0); SWM (0); RTI (0); AST (0) | YES                      |

Note: (n): number; (N = ): subjects; −: impairment; +: improvement; 0: no effect.

Abbreviations: ASI, addiction severity index; AST, attention switching task; BAI, Beck anxiety inventory; B-BAES, brief-biphasic alcohol effects scale; BDI, Beck depression inventory; BPI, brief pain inventory; BPRS, brief psychiatric rating scale; CANTAB, Cambridge neuropsychological test automated battery; CAS, constipation assessment scale; COWS, clinical opioid withdrawal scale; CPT, cold pressor task; DMS, delayed matching to sample; DSMV, diagnostic and statistical manual of mental disorders V edition; ECG, electrocardiogram; FSH, follicle stimulating hormone; FSS, fatigue severity scale; K-use, kratom use (time is based on study’s inclusion criteria); KDS, kratom dependence scale; LDQ, leads dependence questionnaire; LH, luteinizing hormone; MCQ, marijuana craving questionnaire; MG, mitragynine; MINI: mini international neuropsychiatric interview; MOT, motor screening task; MS, Mitragyna speciosa; MWC, marijuana withdrawal checklist; PAL, paired associates learning; PK, pharmacokinetics; PSQI: Pittsburgh sleep quality index; RTI, reaction time; SWM, spatial working memory; TC, total cholesterol; TG, triglycerides; WHOQOL-BREF, World Health Organization quality of life- BREF.
be possibly involved in pain reduction with herbal remedies (Forouzanfar and Hosseinzadeh, 2018), such as muscle relaxant effects by acting on the neuromuscular junction (Chitrakarn et al., 2010), and antioxidant properties potentially related to phenolic content (Ghazali et al., 2011; Goh et al., 2014; Grewal, 1932b; Parthasarathy et al., 2009; Yuniarti et al., 2020).

In preclinical studies, kratom was found to reduce ethanol (Cheaha et al., 2015; Gutridge et al., 2020; Kumarnsit et al., 2007a; Vijeepallam et al., 2019) and morphine (Cheaha et al., 2017; Fukurazi et al., 2013; Harun et al., 2020; Hassan et al., 2020; Hemby et al., 2019; Jamil et al., 2013; Khor et al., 2011; Meepong and Sooksawate, 2019; Wilson et al., 2020; Yue et al., 2018) withdrawal and dependence as well. First, it has been suggested that kratom may reduce opioid dependence by acting on mu- and delta-opioid receptors (Harun et al., 2020; Hemby et al., 2019), inducing cAMP pathway down-regulation (with CREB would be the basis of tolerance and dependence), and reducing mRNA mu-opioid receptor expression (Fakurazi et al., 2013; Jamil et al., 2013), and/or avoiding the acquisition/expression of morphine-induced CPP (Meepong and Sooksawate, 2019). The mitigation of opioid withdrawal has been suggested to be dependent on mu-, delta- (Hazim et al., 2011), and kappa-opioid receptors, but also both kratom anxiolytic (Khor et al., 2011; Meepong and Sooksawate, 2019) and antidepressant activity through the serotonergic system (Cheaha et al., 2017) may be involved. The latter mechanism is presumed to be also involved in ethanol withdrawal (Cheaha et al., 2015; Kumarnsit et al., 2007a); the alcohol intake reduction was described to be mainly mediated by delta-opioid receptors (Gutridge et al., 2020).

These findings provide some initial evidence for the therapeutic use of kratom in the treatment of both opioid and alcohol withdrawal and dependence, and support the empirical use of kratom in self-treating of drugs/opioid detoxification and withdrawal as mainly reported by users (Bowe and Kerr, 2020; Boyer et al., 2008, 2007; Grundmann et al., 2020; Schimmel et al., 2020; Singh et al., 2020). Further, among studies conducted in users in our analysis, 12% showed an association between kratom initiation and reduction in the prevalence of adverse effects related to opiates (e.g., respiratory depression, constipation, physical pain) (Saref et al., 2019b), and either in regular drugs use (Saref et al., 2019a). According to Saref et al. (2019a), this evidence suggests that kratom may also be a useful agent, less risky than opioids, for harm-reduction purposes (Saref et al., 2019a). This data may be supported by the findings of a recent study that showed LKT to induce fewer side effects (e.g., physical dependence/respiratory depression) compared to MOR without affecting motor activity (Wilson et al., 2020). Similarly, some authors reported that mitragynine is a compound with a minor addictive potential (Meepong and Sooksawate, 2019; Thériault et al., 2020; Yue et al., 2018), when compared to MOR (Cheaha et al., 2017; Harun et al., 2015), neither it caused physiological dependence (Harun et al., 2020). Moreover, despite kratom and 7HMG were reported to have rewarding effects (Gutridge et al., 2020), with 7HMG having a higher abuse potential (Hassan et al., 2019; Sabetghadam et al., 2013; Yusoff et al., 2016), a recent study found that both mitragynine and 7HMG did not show rewarding actions in the intracranial self-stimulation (Behnood-Rod et al., 2020).

Furthermore, kratom has also been presumed to act on the serotonergic/adrenergic system and dorsal raphe nucleus (Kumarnsit et al., 2007a, 2007b), and to lessen both corticotrophin-releasing factor (CRF) and prodynorphin mRNA expression by acting on the hypothalamic-pituitary-adrenal axis (HPA) in the Central Nervous System (CNS) (Idayu et al., 2011; Khor et al., 2011). These mechanisms are reported to mediate stress mitigating (Hazim et al., 2011; Khor et al., 2011; Vázquez López et al., 2017), anxiolytic-like (Hazim et al., 2014; Khor et al., 2011; Moklas et al., 2013), and antidepressant-like effects (Idayu et al., 2011; Kumarnsit et al., 2007a, 2007b). These effects, together with kratom's antipsychotic-like effects through 5-HT2 and D2 receptors inhibition (Vijeepallam et al., 2016), may be considered as preclinical evidence of kratom's potential therapeutic use in psychiatric disorders as well since many users claim kratom's benefits to self-treat depression, anxiety and attention deficit hyperactivity disorder (ADHD) (Bath et al., 2020; Veltri and Grundmann, 2019). That is linear with the idea reported in the literature that some plants such as kratom, having an indole moiety like common antidepressant drugs, might be a potential alternative plant-based remedy for treating depression (Hamid et al., 2017) and psychological disorders (Johnson et al., 2020).

Then, we found kratom to exert therapeutic effects in additional medical domains. These included a peptic ulcer protective action (Chitrakarn et al., 2018) and acid gastric secretion inhibition (Tsuchiya et al., 2002), with a possible indirect anorectic action (Chitrakarn et al., 2008; Grewal, 1932b; Kumarnsit et al., 2006, 2007b), antidiarrheal effect (Chitrakarn et al., 2008), anhelmintic (Abdul Aziz et al., 2012), antibacterial effects (Juanda et al., 2019; Parthasarathy et al., 2009), antipyretic (Salleh et al., 2011), antimitagen/anticancer (Ghazali et al., 2011; Goh et al., 2014), antitussive (Macko et al., 1972), and antihypertensive effect (Grewal, 1932b), that appear in line with the traditional application of the plant for treating stomach ailments, diabetes, diarrhea, infections, fever, cough, hypertension (Brown et al., 2017; Eastlack et al., 2020; Hassan et al., 2013; Kruegel and Grundmann, 2018; Ramachandram et al., 2019; Saref et al., 2019a; Singh et al., 2017, 2020; Suhaimi et al., 2016; Vicknasingam et al., 2010). It was also found of potential benefit in treating COVID-19 symptoms (Mastasio et al., 2020).

Finally, the facilitation of learning through the modulation of memory consolidation (Senik et al., 2012) may provide preclinical evidence of kratom on nootropic effects. This data was also confirmed in other preclinical studies where kratom showed cognitive enhancing properties (Hazim et al., 2011; Ilmie et al., 2015). Further, kratom use did not seem to have long-term cognitive effects on users, but it was found to affect only visual episodic memory causing learning impairment in chronic users with at least two years of use (Singh et al., 2019c). This latest data is in strong contrast with preclinical evidence related to kratom's potential to enhance cognition but appears linear with the cognitive impairment described in preclinical studies with both chronic (Apyrani et al., 2010; Compton et al., 2014; Hassan et al., 2019; Ismail...
et al., 2017; Yusoff et al., 2016) and acute (10, 30, 100 mg/kg, p.o.) (Moklas et al., 2013) administration of the preparation. It is possible to say that findings are inconsistent, and kratom’s significant cognitive impact needs to be further investigated.

On the other side, clinical studies related to therapeutic applications are lacking as well. In the analyzed studies, most participants consumed ≥ two-to-three glasses of kratom 1–3 times daily with a mitragynine content ranging between a minimum of 72.5 mg and a maximum of 434.28 mg. However, data about the consumed amount was only reported in few studies (Leong Bin Abdullah et al., 2020b; Saref et al., 2019b; Singh et al., 2014, 2018a, 2018b, 2018d, 2019c).

Further, adverse events such as alterations of cholesterol level, dependence, and withdrawal symptoms were reported and described as mild and dependent on higher doses (Singh et al., 2018a, 2018c, 2018d, 2019a) or more frequent use (Leong Bin Abdullah et al., 2020a, 2020b). This suggests that these adverse events may not occur at lower doses used with less frequency. These findings confirm that those who use kratom in traditional settings regularly could experience kratom cessation related concerns, as previously reported (Saingam et al., 2016; Vicknasingam et al., 2010), but evidence suggests that most of them self-manage their symptoms (Singh et al., 2014, 2015), and experience more tolerable pain when compared to opioids (Singh et al., 2018d). However, a physical well-being impairment has been reported only in severe kratom dependence (Leong Bin Abdullah et al., 2019a). Vicknasingam et al. (2020) did not find withdrawal symptoms in the observation period (20 h), and Trakulsrichai et al. (2015) did not describe serious adverse events in humans.

Case reports in the literature showed other health problems related to chronic kratom use, such as hepatic damage, endocrinologic issues (e.g., hypogonadism and hypothyroidism), neurological disorders, such as posterior reversible leukoencefalopathy syndrome, seizure and coma, pulmonary (e.g., acute respiratory distress syndrome, ARDS), and cardiovascular problems (Alsarraf et al., 2019; Anwar et al., 2016; Schimmel and Dart, 2020). However, these concerns were mainly reported by users in Western countries, who besides the potential risks, would stress the plant’s beneficial effects as well.

4.1 | Limitations

Our review has some limitations. First, findings from preclinical studies are not always comparable due to methodological limitations linked to the studied compounds/preparations, ways of administration, and the variability of the extract composition as it may contain other alkaloids like paynantheine, corynantheidine and speciociliatine, speciogynine, mitragynaline, and corynantheidaline (Takayama, 2004). Thus, this limits strong conclusions about the effect of mitragynine on the investigated domains. Second, all clinical studies were performed in chronic kratom users, in Southern East Asia, with three out of them (only one RCT) studying kratom acute effects, while no one tested long-term effects of single/repeated administration nor RCTs have been conducted in kratom naive participants. Most of the other clinical studies had a cross-sectional design, which does not allow a definitive causal interpretation of a direct link between kratom consumption and health consequences, providing mainly retrospective information in terms of kratom’s exposure. However, this is not generalizable to the population that occasionally uses kratom in traditional settings, nor to those that use it in non-traditional settings in the West, where available kratom products may differ in terms of potency. Moreover, the almost complete absence of female participants should be considered in further studies to understand gender-related variation in metabolism and pharmacology.

5 | CONCLUSIONS

Taken together, our findings help to explain, but not endorse, the empirical medical use reported by kratom users in non-medical settings in both Asian traditional and Western countries, suggesting that kratom could be a useful aid in the treatment of acute/chronic pain, opioid and substance use disorders, and psychiatric disorders. Kratom-related safety issues must be carefully considered. Until now, mitragynine and kratom’s benefits and safety profile remain largely anecdotal. More studies should be encouraged with different populations, including kratom-naive users in controlled clinical settings, to identify better mitragynine and kratom’s risks and benefits.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this paper or that could be perceived as prejudicing the impartiality of the research reported.

ETHICS STATEMENT

The authors declare that human ethics approval was not needed for this study.

AUTHORS CONTRIBUTIONS

Conceptualization, investigation, and methodology: Elisabeth Prevete, Eef L. Theunissen, Kim P. C. Kuypers, Johannes G. Ramaekers. Writing-Original Draft Preparation: Elisabeth Prevete. Writing-Review and Editing: Elisabeth Prevete, Eef L. Theunissen, Kim P. C. Kuypers, Giuseppe Bersani, Johannes G. Ramaekers, Ornella Corazza. Supervision: Johannes G. Ramaekers.

AUTHORS NOTE

The authors confirm that this work is original and has not been published elsewhere. It is currently not under consideration for publication elsewhere, or in press at other journals.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.
Bowe, A., & Kerr, P. L. (2020). A complex case of kratom dependence, Rod, A., Chellian, R., Wilson, R., Hiranita, T., Sharma, A., Leon, F.,‐‐Behnood‐Aziddin, R. E. R., Mustafa, M. R., Mohamed, Z., & Mohd, M. A. (2005). Anti‐Basiliere, S., & Kerrigan, S. (2020). CYP450
Apryani, E., Hidayat, M. T., Moklas, M. A., Fakurazi, S., & Idayu, N. F. (2010). Effects of mitragynine from Mitragyna speciosa Korth leaves on working memory. Journal of Ethnopharmacology, 129, 357‐360. https://doi.org/10.1016/j.jep.2010.03.036
Aziddin, R. E. R., Mustafa, M. R., Mohamed, Z., & Mohd, M. A. (2005). Anti‐inflammatory properties of Mitragyna speciosa extract. MJS, 24, 191‐194. May 2020. https://mjs.um.edu.my/article/view/8971
Basiliere, S., & Kerrigan, S. (2020). CYP450‐Mediated metabolism of mitragynine and investigation of metabolites in human urine. Journal of Analytical Toxicology, 44, 301‐313. https://doi.org/10.1093/jat/bkz108
Bath, R., Bucholz, T., Buros, A. F., Singh, D., Smith, K. E., Veitri, C. A., & Grundmann, O. (2020). Self‐reported health diagnoses and demographic correlates with kratom use: Results from an online survey. Journal of Addiction Medicine, 14, 244‐252. https://doi.org/10.1097/adm.0000000000000570
Behnood‐Rod, A., Chellian, R., Wilson, R., Hiranita, T., Sharma, A., Leon, F., McCurdy, C. R., McMahon, L. R., & Bruijnzeel, A. W. (2020). Evaluation of the rewarding effects of mitragynine and 7‐hydroxymitragynine in an intracranial self‐stimulation procedure in male and female rats. Drug and Alcohol Dependence, 215, 108235. https://doi.org/10.1016/j.drugalcdep.2020.108235
Bowe, A., & Kerr, P. L. (2020). A complex case of kratom dependence, depression, and chronic pain in opioid use disorder: Effects of buprenorphine in clinical management. Journal of Psychoactive Drugs, 52, 1–452. https://doi.org/10.1080/02791072.2020.173586
Boyer, E. W., Babu, K. M., Adkins, J. E., McCurdy, C. R., & Halpern, J. H. (2008). Self‐treatment of opioid withdrawal using kratom (Mitragyna speciosa korth). Addiction, 103, 1048–1050. https://doi.org/10.1111/j.1360‐0443.2008.02209.x
Boyer, E. W., Babu, K. M., Macalino, G. E., & Compton, W. (2007). Self‐treatment of opioid withdrawal with a dietary supplement, Kramt. American Journal on Addictions, 16, 352–356. https://doi.org/10.1080/105540970152368
Brown, P. N., Lund, J. A., & Murch, S. J. (2017). A botanical, phytochemical and ethnomedical review of the genus Mitragyna korth: Implications for products sold as kratom. Journal of Ethnopharmacology, 202, 302–325. https://doi.org/10.1016/j.jep.2017.03.020
Carpenter, J. M., Criddle, C. A., Craig, H. K., Ali, Z., Zhang, Z., Khan, I. A., & Sufka, K. J. (2016). Comparative effects of Mitragyna speciosa extract, mitragynine, and opioid agonists on thermal nociception in rats. Fitoterapia, 109, 87–90. https://doi.org/10.1016/j.fitote.2015.12.001
Cheaha, D., Keawpradub, N., Sawangjaroen, K., Phukpattaranon, P., & Kumarnsit, E. (2015). Effects of an alkaloid‐rich extract from Mitragyna speciosa leaves and fluoxetine on sleep profiles, EEG spectral frequency and ethanol withdrawal symptoms in rats. Phyto‐medicine, 22, 1000–1008. https://doi.org/10.1016/j.phymed.2015.07.008
Cheaha, D., Reakkamnuan, C., Nukitram, J., Chitrtrakarn, S., Phukpattaranon, P., Keawpradub, N., & Kumarnsit, E. (2017). Effects of alkaloid‐rich extract from Mitragyna speciosa (Korth.) Havil. on naloxone‐precipitated morphine withdrawal symptoms and local field potential in the nucleus accumbens of mice. Journal of Ethnopharmacology, 208, 129–137. https://doi.org/10.1016/j.jep.2017.07.008
Chitrtrakarn, S., Keawpradub, N., Sawangjaroen, K., Kansenalak, S., & Janchawee, B. (2010). The neuromuscular blockade produced by pure alkaloid, mitragynine and methanol extract of kratom leaves (Mitragyna speciosa Korth.). Journal of Ethnopharmacology, 129, 344–349. https://doi.org/10.1016/j.jep.2010.03.035
Chitrtrakarn, S., Radenahmad, N., Kaeawsara, S., Udomkusorn, W., Keaw‐pradub, N., & Phukpattaranon, P. (2018). Gastroprotective effects of methanolic extract of kratom leaves on gastric ulcer and reflux esophagitis in rats. Songklanakarin Journal of Science and Technology, 40(2), 258–263. May 2020. https://rdo.psu.ac.th/sjstweb/journal/40‐2/40‐2‐1.pdf
Chitrtrakarn, S., Sawangjaroen, K., Praseththo, S., Janchawee, B., & Keaw‐pradub, N. (2008). Inhibitory effects of kratom leaf extract (Mitragyna speciosa Korth.) on the rat gastrointestinal tract. Journal of Ethnopharmacology, 116, 173–178. https://doi.org/10.1016/j.jep.2007.11.032
Cinis, E., Martinotti, G., Simonato, P., Singh, D., Demetrovics, Z., Roman‐Urrestarazu, A., Bersani, F. S., Vicknasingam, B., Piazzon, G., Li, J. H., Yu, W. J., Kapitany‐Foveny, M., Farkas, J., Di Gianantonio, M., & Corazza, O. (2015). Following the “roots” of kratom (mitragynine speciosa): The evolution of an enhancer from a traditional use to increase work and productivity in Southeast Asia to a recreational psychoactive drug in western countries. Biomed Research International, 2015, 968786–968811. https://doi.org/10.1155/2015/968786
Coe, M. A., Pillitteri, J. L., Sembower, M. A., Gerlach, K. K., & Hennesfield, J. E. (2019). Kratom as a substitute for opioids: Results from an online survey. Drug and Alcohol Dependence, 202, 24–32. https://doi.org/10.1016/j.drugalcdep.2019.05.005
Compton, D. M., García, C., Kamaratos, A., Johnson, B. G., & Wedge, T. (2014). An examination of the consequences of chronic exposure to Mitragyna speciosa during adolescence on learning and memory in adulthood. J Phytopharmacol, 3(5), 300–309. June 2020. http://www.phytopharmacajournal.com/Vol3_Issue5_01.pdf
Corkery, J. M., Streete, P., Claridge, H., Goodair, C., Papanti, D., Orsolini, L., Schifano, F., Sikka, K., Körber, S., & Hendricks, A. (2019). Characteristics of deaths associated with kratom use. Journal of Psychopharmacology, 33, 1102–1123. https://doi.org/10.1177/0269881119862530
Criddle, C. (2015). A comparison of mitragynine speciosa and mitragynine against opioids on thermal nociception in rats. University of Mississippi. July 2020. https://core.ac.uk/download/pdf/148695039.pdf
Eastlock, S. C., Cornett, E. M., & Kaye, A. D. (2020). Kratom‐pharmacology, clinical implications, and outlook: A comprehensive review. Pain Ther, 9, 55–69. https://doi.org/10.1007/s40122‐020‐00151‐x
Fakurazi, S., Rahman, S. A., Hidayat, M. T., Ithnin, H., Moklas, M. A., & Arulselvan, P. (2013). The combination of mitragynine and morphine prevents the development of morphine tolerance in mice. Molecules, 18, 666–681. https://doi.org/10.3390/molecules18101066
Faouzi, A., Varga, B. R., & Majumdar, S. (2020). Biased opioid ligands. Molecules, 25, 4257. https://doi.org/10.3390/molecules25184257
Forouzanfar, F., & Hosseinzadeh, H. (2018). Medicinal herbs in the treatment of neuropathic pain: A review. Iranian Journal of Basic
derivative of the indole alkaloid mitragynine: A novel dual-acting mu- and kappa-opioid agonist with potent antinociceptive and weak rewarding effects in mice. Neuropharmacology, 55, 154–165. https://doi.org/10.1016/j.neuropharm.2008.05.003

Maxwell, E. A., King, T. I., Kamble, S. H., Raju, K. S. R., Berthold, E. C., León, F., Avery, B. A., McMahon, L. R., McCurdy, C. R., & Sharma, A. (2020). Pharmacokinetis and safety of mitragynine in beagle dogs. Planta Medica, 86, 1278–1285. https://doi.org/10.1055/a-1212-5475

Meepong, R., & Sooksawate, T. (2019). Mitragynine reduced morphine-induced conditioned place preference and withdrawal in rodents. Thai Journal of Pharmaceutical Sciences (TJPS), 43, http://www.tjps.pharm.chula.ac.th/ojs/index.php/tjps/article/view/892

Metastasio, A., Prevete, E., Singh, D., Grundmann, O., Prozialeck, W. C., Veltri, C., Bersani, G., & Corazza, O. (2020). Can kratom (Mitragyna speciosa) alleviate COVID-19 pain? A case study. Frontiers in Psychiatry, 11, 594816. https://doi.org/10.3389/fpsyg.2020.594816

Moher, D., Liberati, A., Tetzlaff, J., & Altman, D. G. (2009). Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. BMJ, 339, b2535. https://doi.org/10.1136/bmj.b2535

Moklas, M. A. M., Suliman, N. A., Taib, C. N. M., Hidayat, M. T., Baharudin, S. F., Zakaria, F. N., Yusof, M. K. M., Adzhar, M. F., Rasul, M. S. M., & Akim, A. M. (2013). Sedative, cognitive impairment and anxiolytic effects of acute Mitragyna Speciosa in rodents. differences, 22, 313–317. July 2020. https://www.researchgate.net/publication/309448538

Mossadeq, W. M. S., Sulaiman, M. R., Tengku Mohamad, T. A., Chong, H. S., Zakaria, Z. A., Jabit, M. L., Baharudin, M. T., & Israf, D. A. (2009). Anti-inflammatory and antinociceptive effects of Mitragyna speciosa Korth methanolic extract. Medical Principles and Practice, 18, 378–384. https://doi.org/10.1159/000226292

Olsen, E. O., O’Donnell, J., Mattson, C. L., Schier, J. G., & Wilson, N. (2019). Notes from the field: Unintentional drug overdose deaths with kratom detected - 27 states (July 2016–December 2017). MMWR Morb Mortal Wkly Rep, 68, 326–327. https://doi.org/10.15585/mmwr.mm6814a2

Parthasarathy, S., Bin Azizi, J., Ramanathan, S., Ismail, S., Sasidharan, S., Said, M. I. M., & Mansor, S. M. (2009). Evaluation of antioxidant and antibacterial activities of aqueous, methanolic and alkaloid extracts from Mitragyna speciosa (Rubiaceae family) leaves. Molecules, 14, 3964–3974. https://doi.org/10.3390/molecules14103964

Prozialeck, W. C., Jivan, J. K., & Andurkar, S. V. (2012). Pharmacology of Mitragynine, A. Prozialeck, W. C., Singh, D., Grundmann, O., Maxwell, E. A., King, T. I., Kamble, S. H., Raju, K. S. R., Berthold, E. C., León, F., Avery, B. A., McMahon, L. R., McCurdy, C. R., & Sharma, A. (2020). Pharmacokinetis and safety of mitragynine in beagle dogs. Planta Medica, 86, 1278–1285. https://doi.org/10.1055/a-1212-5475

Reanmongkol, W., Keawpradub, N., & Sawangjaroen, K. (2007). Effects of the extracts from Mitragyna speciosa Korth. leaves on analgesic and behavioral activities in experimental animals. Songklanakarin Journal of Science and Technology, 29(Suppl. 1), 39–48. Thai Herbs II. https://rdc.psu.ac.th/sjst/journal/29-Suppl-1/05Wantana_39-48.pdf

Sabetghadam, A., Navaratnam, V., & Mansor, S. M. (2013). Dose–response relationship, acute toxicity, and therapeutic index between the alkaloid extract of M iragyna speciosa and its main active compound mitragynine in mice. Drug Development Research, 74, 23–30. https://doi.org/10.1002/ddr.21052

Sabetghadam, A., Ramanathan, S., & Mansor, S. M. (2010). The evaluation of antinociceptive activity of alkaloid, methanolic, and aqueous extracts of Malaysian Mitragyna speciosa Korth leaves in rats. Pharmacognosy Research, 2, 181–185. https://doi.org/10.4103/0974-8490.65514

Saingam, D., Assanangkornchai, S., Geater, A. F., & Lerkiatbundit, S. (2016). Factor Analytical investigation of krathom (mitragyna speciosa korth.) withdrawal syndrome in Thailand. Journal of Psychoactive Drugs, 48, 76–85. https:// doi.org/10.1080/02791072.2016.1156791

Salleh, A., Kadir, A., & Mossadeq, M. S. (2011). Antipyretic effect of crude methanolic extract of mitragyna speciosa in mice. 6th Proceedings of the Seminar on Veterinary sciences (pp. 1–6). May 2020. http://psasir.upm.edu.my/id/eprint/62521/1/Proceedings%20pdf

Saref, A., Suraya, S., Singh, D., Grundmann, O., Narayan, S., Swogger, M. T., Prozialeck, W. C., Boyer, E., & Balasingam, V. (2019a). Self-report data on regular consumption of illicit drugs and HIV risk behaviors after kratom (mitragyna speciosa korth.) initiation among illicit drug users in Malaysia. Journal of Psychoactive Drugs, 52, 1–144. https://doi.org/10.1080/02791072.2019.1686553

Saref, A., Suraya, S., Singh, D., Grundmann, O., Narayan, S., Swogger, M. T., Prozialeck, W. C., Boyer, E., Cheer, N. J. Y., & Balasingam, V. (2019b). Self-reported prevalence and severity of opioid and kratom (Mitragyna speciosa korth.) side effects. Journal of Ethnopharmacology, 238, 111876. https://doi.org/10.1016/j.jep.2019.111876

Schimmel, J., Amioka, E., Rockhill, K., Haynes, C. M., Black, J. C., Dart, R. C., & Iwanicki, J. L. (2020). Prevalence and description of kratom (mitragyna speciosa) use in the United States: A cross-sectional study. Addiction, 116, 176–181. https://doi.org/10.1111/add.15082

Schimmel, J., & Dart, R. C. (2020). Kratom (mitragyna speciosa) liver injury: A comprehensive review. Drugs, 80, pp. 1–283. https://doi. org/10.1007/s40265-019-01242-6

Senik, M. H., Mansor, S. M., Tharakan, J. K. J., & Abdullah, J. (2012). Effect of acute administration of Mitragyna speciosa Korth. standardized methanol extract in animal model of learning and memory. Journal of Medicinal Plants Research, 6, 1007–1014. https://academicjournals. org/journal/JMPR/article-full-text-pdf/83970727156

Shamma, A. R., Fakurazi, S., Hidayat, M. T., Hairusszah, I., Moklas, M. A., & Arulselvan, P. (2012). Antinociceptive action of isolated mitragynine from Mitragyna Speciosa through activation of opioid receptor system. International Journal of Molecular Sciences, 13, 11427–11442. https://doi.org/10.3390/ijms130911427

Shelldor, E. J. (1974). The alkaloids of Mitragyna with special reference to those of Mitragyna speciosa, Korth. Bull Narc, 26, 41e55.

Singh, D., Damodaran, T., Prozialeck, W. C., Grundmann, O., Karunakaran, T., & Vicknasingam, B. (2019a). Constipation prevalence and fatigue severity in regular kratom (Mitragyna speciosa Korth.) users. Journal of Substance Use, 24, 233–239. https://doi.org/10.1080/14659981.2018.156340

Singh, D., Müller, C. P., Murugaiyah, V., Hamid, S. B. S., Vicknasingam, B. K., Avery, B., Cheer, N. J. Y., & Mansor, S. M. (2018a). Evaluating the hematological and clinical-chemistry parameters of kratom (Mitragyna speciosa) users in Malaysia. Journal of Ethnopharmacology, 214, 197–206. https://doi.org/10.1016/j.jep.2017.12.017

Singh, D., Müller, C. P., & Vicknasingam, B. K. (2014). Kratom (Mitragyna speciosa) dependence, withdrawal symptoms and craving in regular users. Drug and Alcohol Dependence, 139, 132–137. https://doi.org/10.1016/j.drugalcdep.2014.03.017
Wilson, L. L., Harris, H. M., Eans, S. O., Brice-Tutt, A. C., Cirino, T. J., Stacy, H. M., Simons, C. A., León, F., Sharma, A., Boyer, E. W., Avery, B. A., McLaughlin, J. P., & McCurdy, C. R. (2020). Lyophilized kratom tea as a therapeutic option for opioid dependence. Drug and Alcohol Dependence, 216, 108310. https://doi.org/10.1016/j.drugalcdep.2020.108310

Wong, A., & Mun, M. (2020). A case of kratom overdose in a pediatric patient. Case Reports in Psychiatry, 2020, 8818095. https://doi.org/10.1155/2020/8818095

Yue, K., Kopajtic, T. A., & Katz, J. L. (2018). Abuse liability of mitragynine, a major analgesic alkaloid in kratom (mitragyna speciosa): A systematic review. Asian Journal of Psychiatry, 43, 73–82. https://doi.org/10.1016/j.jap.2019.05.016

Yee, K., Kopajtic, T. A., & Katz, J. L. (2018). Abuse liability of mitragynine assessed with a self-administration procedure in rats. Psychopharmacology, 235, 2823–2829. https://doi.org/10.1007/s00213-018-4974-9

Yuniarti, R., Nadia, S., Alamanda, A., Zubir, M., Syahputra, R., & Nizam, M. (2020). Characterization, phytochemical screenings and antioxidant activity test of kratom leaf ethanol extract (mitragyna speciosa korth) using DPPH method. Journal of Physics: Conference Series, 1462, 012026. November 2020. https://ui.adsabs.harvard.edu/link_gateway/2020JPhCS1462a2026Y/doi:10.1088/1742-6596/1462/1/012026

APPENDIX

| Abbreviations | Definition |
|---------------|------------|
| 5-HT          | serotonin  |
| 7HMG          | 7-hydroxymitragynine |
| α2            | adrenenergic |
| A2A           | adenosine   |
| ADD           | attention deficit disorder |
| ADHD          | attention deficit hyperactivity disorder |
| ARDS          | acute respiratory distress syndrome |
| CNS           | central nervous system |
| COX-2         | cyclooxygenase-2 |
| CPP           | conditioned place preference |
| CREB          | cAMP response element binding |
| CRF           | corticotrophin releasing factor |
| CSA           | controlled substances act |
| CYP450        | cytochrome P450 |
| D2            | dopamine    |
| DEA           | drug enforcement administration |
| ECG           | electrocardiogram |
| FDA           | food and drug administration |
| FSH           | follicle stimulating hormone |
| HPA           | hypothalamic-pituitary-adrenal axis |
| i.p.          | intraperitoneal administration |
| i.v.          | intravenous  |
| LH            | luteinizing hormone |
| LKT           | lyophilized kratom tea |
| MeSH          | medical subject headings |
| MG            | mitragynine |
| MRM-15        | (E)-methyl 2-((2S,3S,7aS,12aR,12bS)-3-ethyl-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindolo[2,3-a]quinolin-2-yl)-3-methoxyacrylate |
| MRM-16        | (E)-methyl 2-((2S,3S,7aS,12aR,12bS)-3-ethyl-9-fluoro-7a-hydroxy-8-methoxy-1,2,3,4,6,7,7a,12,12a,12b-decahydroindolo[2,3-a]quinolin-2-yl)-3-methoxyacrylate |
| MRM-9         | [(E)-methyl 2-(3-ethyl-7a,12a-(epoxyethanoxy)-9-fluoro-1,2,3,4,6,7,12,12b-octahydro-8-methoxyindolo[2,3-a]quinolin-2-yl]-3-methoxyacrylate] |
| MOR           | morphine    |
| MS            | Mitragyna speciosa |
| p.o.          | oral administration |
| RCT           | randomized controlled trial |
| T1/2          | half-life   |
| US            | United States |

Yusoff, S. R., Mohd Uzid, M., Teh, E. H., Hanapi, N. A., Mohideen, M., Mohamad Arshad, A. S., Mordi, M. N., Loryan, I., & Hammarlund-Udenaes, M. (2019). Rate and extent of mitragynine and 7-hydroxymitragynine blood-brain transport and their intra-brain distribution: The missing link in pharmacodynamic studies. Addiction Biology, 24, 935–945. https://doi.org/10.1111/adb.12661

Yusoff, N. H., Suhami, F. W., Vadivelu, R. K., Hassan, Z., Rümler, A., Rotter, A., Amato, D., Dringenberg, H. C., Mansor, S. M., Navaratnam, V., & Müller, C. P. (2016). Abuse potential and adverse cognitive effects of mitragynine (kratom). Addiction Biology, 21, 98–110. https://doi.org/10.1111/adb.12185

How to cite this article: Prevete, E., Kuypers, K. P. C., Theunissen, E. L., Corazza, O., Bersani, G., & Ramaekers, J. G. (2021). A systematic review of (pre)clinical studies on the therapeutic potential and safety profile of kratom in humans. Human Psychopharmacology: Clinical and Experimental, e2805. https://doi.org/10.1002/hup.2805