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ANA LAURA MASQUETTI FAVA

AVALIAÇÃO DA EFETIVIDADE DO CANABIDIOL EM FORMULAÇÕES TÓPICAS

CANNABIDIOL EFFECTIVENESS EVALUATION IN TOPICAL FORMULATION

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Tese apresentada à Faculdade de Ciências Médicas da Universidade Estadual de Campinas como parte dos requisitos exigidos para a obtenção do título de Mestra em Ciências, Área de Concentração Pesquisa Clínica.

Thesis presented to the School of Medical Sciences of the State University of Campinas as part of the requirements required to obtain the title of Master of Science, Area of Concentration in Clinical Research.

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“Se eu vi mais longe, foi por estar sobre ombros de gigantes.”

Isaac Newton

(1643-1727)

RESUMO

FAVA, Ana Laura Masquetti. *Avaliação da efetividade do canabidiol em formulações tópicas.* (Dissertação Mestrado) Programa de pós-graduação em Ciências Médicas, Faculdade de Ciências Médicas, Universidade Estadual de Campinas. Campinas, São Paulo, 2022.

O presente estudo visa desenvolver revisão escopo, referente ao tratamento tópico com canabidiol, em relatos de experimentos pré-clínicos e clínicos. Para tal, utilizou-se a metodologia recomendada pelo Instituto Joanna Briggs, e os estudos foram selecionados de acordo com critérios de inclusão e exclusão determinados previamente. *Cannabis sativa* pertence à família Cannabaceae, caracterizada por uma vasta atividade farmacológica. Isso porque, em sua composição molecular, a classe dos Canabinóides é predominante e a responsável por causar os efeitos farmacológicos. O tetrahidrocanabinol (THC) e o canabidiol (CBD) são, respectivamente, os bioativos mais concentrados na *Cannabis sativa.* Ao longo dos anos, o desenvolvimento científico permitiu determinar o potencial farmacológico do CBD em diversas doenças e síndromes, já que esta demonstra uma segurança maior que o THC, a partir da diminuição de efeitos adversos, como os efeitos psicotrópicos. A via de administração tópica, tem sido investigada por apresentar vantagens quanto a adesão do paciente, ao tratamento e redução dos efeitos. Com o canabidiol não foi diferente, e estudos foram desenvolvidos para determinar a sua efetividade por esta via de administração. Os resultados apontaram para uma boa atividade farmacológica do CBD, principalmente quando combinado com THC quando avaliados no tratamento de glaucoma, dermatite atópica, esclerose múltipla, osteoartrite e outras.

Palavras- chave: Canabidiol; *Cannabis sativa;* Administração tópica.

ABSTRACT

FAVA, Ana Laura Masquetti. *Cannabidiol effectiveness evaluation in topical formulation.* (Mesters) Post- graduation program in Medical Science, Schoolf of Medical Science, State University of Campinas. Campinas, São Paulo, 2022.

The present study aims to carry out a scope review, referent to the use of cannabidiol, in pre-clinical and clinical studies, for topical administration. For that, the methodological approach used by the Joanna Briggs Institute was applied, and the studies were selected by previous determined inclusion criteria. *Cannabis sativa* is a plant of the Cannabaceae family, whose molecular composition is known for vast pharmacological properties. That is due to the presence of cannabinoids, the predominant molecular class, responsible for *Cannabis sativa* potential effects. Tetrahydrocannabinol (THC) and Cannabidiol (CBD), are, respectively, the most concentrated bioactives. Over the years, scientifical development has been interested in the potential of cannabidiol in various health conditions, since demonstrated lower adverse events, when compared to THC, such as psychotropic effects. Topical administration route has been explored in the scientific community for presenting great benefits as improving patient adherence to treatment and reducing adverse events. Cannabidiol was no different, studies were carried out to determine its effectiveness by this route of administration. Results pointed to great success of canabidiol use, especially when combined with tetrahydrocannabinol, in condition as glaucoma, atopic dermatitis, multiple sclerosis, osteoarthritis and others.

Keywords: Cannabidiol; *Cannabis sativa*; Topical Administration.

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1. INTRODUÇÃO

Cannabis sativa, popularmente conhecida como maconha, é uma planta da família das Cannabacea. Trata-se de uma planta caracterizada por suas folhas verdes, palmadas, com sete lóbulos e margens serrilhadas (FARAG; KAYSER, 2017). A *Cannabis sativa* possui importantes propriedades terapêuticas, muitas das quais ainda estão em estudo, mas tem sido usada na medicina, majoritariamente em preparações orais, para tratar doenças do sistema nervoso (FIANI *et al.*, 2020).

As atividades farmacológicas da planta são atribuídas a uma classe específica de bioativos denominada canabinóides, em especial duas moléculas pertencentes a esta classe: o Tetraidrocanabinol (THC) e o Canabidiol (CBD), que estão presentes em maior concentração (RADWAN *et al.*, 2021).

Os canabinóides interagem com os receptores canabinóides 1 e 2 (CB1 e CB2), os quais pertencem ao sistema endocanabinóide. Os receptores estão acoplados à proteína G, que quando acionada, inibe a enzima adenilatociclase, reduzindo a concentração de AMP cíclico e resultando na inibição de canais de cálcio. De tal modo, a ativação dos receptores CB1 e CB2 levam a diminuição da liberação de neurotransmissores, tanto inibitórios, quanto excitatórios (MATOS *et al.*, 2017).

Esta interação modula os efeitos, ao THC atribui-se atividades psicoativas, antiespasmódico, relaxante muscular, antiemético, indutor do sono, estimulante do apetite e analgesia (BABSON; SOTTILE; MORABITO, 2017; LEE *et al.*, 2013; MEIRI *et al.*, 2007; RUSSO; GUY; ROBSON, 2007). A elevada potência deste bioativo, pode resultar em reações adversas importantes, portanto, pode não ser a melhor opção de tratamento, especialmente quando administradas em mucosas, como irritação local (COLASANTI; POWELL; CRAIG, 1984; ELSOHLY *et al.*, 1984; MECHOULAM *et al.*, 1970).

Por outro lado, o CBD apresenta atividade anti-inflamatória, anticonvulsionante e neuroprotetora, relevantes, além de nociceptivos e ansiolíticos (MASSI *et al.*, 2013; MATOS *et al.*, 2017). A principal vantagem do uso de CBD está no fato de que, comparado ao THC, é necessário maior concentração deste ativo para exercer uma atividade significativa e, por isso, pode ser uma boa opção pra tratamentos tópicos (CHESHER; JACKSON, 1985; FIANI *et al.*, 2020).

A administração tópica e transdérmica é bastante vantajosa, já que evita o metabolismo de primeira passagem, minimiza os efeitos adversos sistêmicos, melhora a adesão do paciente e permite uso de sistemas de liberação controlada (HAVLICKOVA; FRIEDRICH, 2008).

O presente estudo pretende buscar na literatura estudos que exploram os efeitos do canabidiol através da administração tópica. Previamente à decisão de trabalhar com este tema, o planejamento inicial incluía colocar em prática o projeto intitulado "Avaliação da atividade antioxidante da mangiferina em formulações tópicas". Devido à pandemia e ao isolamento social, o cronograma proposto foi comprometido, portanto optou-se por um trabalho que não dependesse do uso das instalações do laboratório de pesquisa. Para o tema inicial, no entanto, não haveria dados suficientes para elaborar uma revisão de escopo ou sistemática e, portanto, um novo tema foi escolhido e aprovado pela Comissão de Pós-graduação do Programa de Pós-graduação em Ciências Médicas (FCM/Unicamp).

Sendo assim, o presente trabalho apresenta revisão de literatura intitulada "*Evidence Of Cannabidiol Effectiveness In Topical Administration: A Scope Review*" (Fava, A.L.M; De Souza, C.M.; Ataide, Dos Santos, E.M.; J.A.; Mazzola, P.G., já submetido em periódico). Ademais, serão contemplados os itens e subitens utilizados tradicionalmente em dissertações. As referências do artigo de revisão e do artigo de resultados encontram-se ao final dos respectivos capítulos, e as demais referências encontram-se no *item 6*, ao final do trabalho.

2. OBJETIVOS

Realizar revisão de escopo referente a efetividade do canabidiol em formulações para uso tópico, incluindo apenas artigos com métodos pré-clínicos, em animais, e clínicos, em humanos.

Dessa forma definimos como objetivos específicos:

- Determinar as principais condições de saúde dispostas na literatura, as quais utilizam canabidiol como principal forma de tratamento;
- Obter informações farmacotécnicas e farmacológicas, tais como forma farmacêutica e dose, que interfiram na administração e na absorção da droga pela via tópica;
- Comparar os dados analíticos obtidos pelos estudos experimentais pré-clínicos e clínicos.

3. METODOLOGIA

3.1. Desenho do Estudo

O desenho do estudo seguiu o método recomendado pela Instituição Joanna Briggs (INSTITUTE, 2014), de acordo com as características específicas de cada revisão.

Para a realização da pergunta para revisão de escopo utilizou-se a seguinte pergunta: Qual a efetividade do canabidiol em formulações tópicas?

3.2. Estratégia de busca e seleção de artigos

Foi realizada uma busca de estudos *in vivo*, pré-clínicos e clínicos, que avaliaram o potencial e as consequências da utilização do extrato de *Cannabis sativa* e/ou de seus bioativos isolados, nas seguintes bases de dados: *Pubmed*, *Pubmed PMC*, *Scopus*, *Web of Science*, *BVS/Bireme*, *Cochrane Library*, *Embase* e *Ebscohost*.

A busca foi realizada em fevereiro de 2021, e atualizada em janeiro de 2022 e julho de 2022. Houve restrição de idioma, incluídos apenas estudos na língua inglesa. As palavras-chaves incluídas foram "*Cannabidiol*", "*Cannabis" "Medical marijuana", "Administration, topical", "Administration, cutaneous"* e *"Transdermal patch*", as quais foram submetidas a avaliação de descritores para desenvolvimento da estratégia de busca.

As referências foram transferidas para *EndNote Web* e *Ryyan*, para seguirem para seleção por título e resumo. A inclusão por título e resumo foi pareada por dois autores (ALMF e CMS), com cegamento. Após a seleção completa por ambos os autores, o cegamento foi quebrado e, em caso de divergências, um terceiro membro (JAA) decidia pela inclusão ou exclusão do resumo.

As referências incluídas nesta etapa foram transferidas para a planilha do Excel 2019. A seleção por texto completo foi realizada por um único autor (ALMF), assim como a extração de dados.

Para que fossem incluídos, os artigos precisavam cumprir com os seguintes critérios:

- Tipo de estudo: experimental *in vivo* pré-clínicos e clínicos;
- Via de administração: administração tópica (mucosa ou pele) e/ou outras vias de administração, desde que o intuito seja comparação entre as vias de administração.
- Formulação: Canabidiol puro ou com combinado com outros bioativos; extrato de *Cannabis sativa* com dosagem dos ativos; formulações comerciais com Canabidiol isolado ou com extrato de *Cannabis sativa* com dosagem dos ativos;

- Metodologia: descrição detalhada do tratamento aplicado de acordo com a condição de saúde estudada.

Ainda, seriam automaticamente excluídos artigos que não cumprissem com os critérios de inclusão, e ainda:

- Não apresentasse informação irrelevante ou não- clínica referente ao uso do Canabidiol;
- Não fosse classificado como estudo experimental, como revisões de literaturas, livros, colunas de revistas, resumos, resumo expandido de congresso, propagandas, cartas para o editor, protocolo de estudo clínicos e/ou que não contenham resultados e discussão;
- Apresentassem formulação contendo mais de uma planta medicinal; que a *Cannabis sativa* e/ou Canabidiol não fossem foco dos estudos;
- Apresentassem formulação com outros bioativos que não Canabidiol;
- Apresentasse moléculas sintéticas ou naturais diferentes daquelas em foco, especialmente se a atividade farmacológica fosse contrária a que é conhecida;
- Estudos não disponíveis, e mesmo que solicitados, não fossem fornecidos.
- Estudo baseados em metodologias *in vitro.*

3.3. Extração de dados

As referências incluídas seguiram para extração de dados, realizada por um único autor (ALMF). Os dados extraídos incluíram a condição de saúde estudada, regime de tratamento, formulação usada, metodologia aplicada, desfecho e resultados, discussão e conclusão. As referências foram organizadas no EndNote X8.

3.4. Interpretação dos dados

Os dados obtidos forma organizados em uma tabela, organizada de acordo com a via de administração utilizada no tratamento, e em seguida, pela condição de saúde em foco. Inicialmente, houve avaliação das concentrações e formulações utilizadas pelos artigos, e uma primeira comparação foi feita.

Tendo em vista a possibilidade de comparação entre os estudos, tanto qualitativamente quanto quantitativamente, os dados foram, então, comparados a partir dos resultados e discussão de cada estudo. Dessa forma foi possível levantar evidências que favoreçam a discussão.

4. RESULTADOS E DISCUSSÃO

4.1. Levantamento Bibliográfico

Aplicando a estratégia de busca nas plataformas de pesquisa, foram encontradas 778 referências. As referências foram transferidas para a plataforma Ryyan, que detectou 81 duplicidades. Excluindo as duplicidades, 697 referências em potencial seguiram para a etapa de inclusão por títulos de resumos.

Ao final, das 697 referências, 528 foram excluídas na fase de inclusão por título e resumo, e apenas 169 foram incluídas, seguindo para a avaliação do texto completo.

A seleção por texto completo foi feita por um único autor, e das 169 referências, 46 foram incluídas e 123 foram excluídas. Infelizmente, 2 referências não foram encontradas e foram excluídas por esse motivo, e 2 artigos foram excluídos por duplicidade. Os artigos incluídos foram divididos em estudos em pré-clínicos, em animais (n=21) e clínicos (n=25).

4.2. Manuscrito da Revisão de Escopo

A revisão de escopo está submetida na revista *Phytotherapy research*, sendo assim, o arquivo que segue é um manuscrito e impede que este seja publicado antes da autorização da revista, caso seja aceito.

EVIDENCE OF CANABIDIOL EFFECTIVENESS IN TOPICAL ADMINISTRATION: A SCOPE REVIEW

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ABSTRACT

Cannabis sativa is a plant of the Cannabaceae family, whose molecular composition is known for vast pharmacological properties. Cannabinoids are the molecules responsible for *Cannabis sativa* potential effects, especially tetrahydrocannabinol and cannabidiol. Scientific development has been interested in the potential of cannabidiol in various health conditions, since demonstrated lower adverse events and a great pharmacological potential, including when administered topically. Thus, the present study aims to carry out a scope review, referent to the use of cannabidiol, *in vivo* models, for topical administration. For that, the methodological approach used by the Joanna Briggs Institute was applied, and the studies were selected by previous inclusion criteria. In general, the molecule demonstrated great success in the treatment and prevention of glaucoma, atopic dermatitis, epidermolysis bullosa, pyoderma gangrenosum, and others. However, more information regarding the dose to achieve pharmacological potential are needed.

Keywords: Cannabidiol, *Cannabis sativa*, Topical Administration.

1. INTRODUCTION

Cannabis is a plant genus part of the Cannabaceae family, whose botanical morphology is known for the presence of hops, as much as *Humulus* genus (Farag; Kayser, 2017; Turner; Elsohly; Boeren, 1980). There are three family members of the *Cannabis* genus, such as *C. sativa* L., *C. ruderalis* and *C. indica* Lam (Farag; Kayser, 2017; Schultes *et al.*, 1974). Although, different classification is discussed among authors, since the use is basically to distinguish the wild variants form the domesticated. With that in mind, many authors refer to all *Cannabis* species as *C. sativa* only (Farag; Kayser, 2017; Hoffmann, 1961). Besides, the leaves of the plants are easily recognized by palmate form, with seven lobes and serrate leaflets, which differs according to genetic origin (Clarke, 1981).

According to previous evidence, aside from the analgesia for pain control, and anti-depressive (stress and anxiety relief), *Cannabis* and/or their bioactives demonstrate important pharmacological properties as well as anti-inflammatory, antitumor, antioxidant, antinociceptive, antimicrobial, neuroprotective, anti-convulsant, anti-parksonian and lowers blood pressure (Kumar *et al.*, 2021). With than in mind, the lack of possibilities for disorder treatment is extremely wide, thus, in countries where the *Cannabis* is illegal, there are some impossibilities for medical use as well, reducing the options.

Through the years, over 500 phytoactives were isolated and identified from *Cannabis,* including alkaloids, terpenoids, flavonoids, cannabinoids and others (Booth; Bohlmann, 2019; Farag; Kayser, 2017; Fischedick, 2017; Hanuš *et al.*, 2016; Radwan *et al.*, 2021; Turner; Elsohly, 1976; Vanhoenacker *et al.*, 2010). From those compounds, 125 were cannabinoids, in summary, cannabinoids classes are divided according to their molecule similarity, besides the Δ^9-THC and CBD types, other groups present in *Cannabis* includes: (−)-Δ8-Trans-Tetrahydrocannabinol (Δ^8-THC), cannabigerol (CBG), cannabinodiol (CBND), cannabielsoin (CBE), cannabicyclol (CBL), cannabichromene (CBC), cannabinol (CBN), cannabitriol (CBT) and Miscellaneous types Cannabinoids (Radwan *et al.*, 2021).

Those bioactives are produced by the glandular trichomes of the plant, whose pharmacological potential is contrasted in *Cannabis*, specially (−)-Δ^9-Trans-Tetrahydrocannabinol (Δ^9-THC), whose concentration is predominant (Elbatsh *et al.*, 2012).

Cannabidiol (CBD) is another cannabinoid, that has been gaining more and more space in the scientific community due to its pharmacological potential use in neurological disorders (Fiani *et al.*, 2020). The safety has been the greatest advantage in the use of cannabidiol in comparison to Δ^9-THC, due to the lower incidence of important adverse events.

Thus, *in vitro* and *in vivo* experiments are the firsts steps to better understand the mechanism of action and primordial effects of the bioactive, before following to human studies.

It is important to emphasized that in the present review topical administration was defined as formulations administrated through skin or mucosa, intending a local or systemic action. The aim of this scoping review was to describe cannabidiol pharmacological potential, when administrated topically in animals and humans.

2. METHODOLOGY

It was performed a scoping review from february to march of 2021 and actualized in January 2022, following the Joanna Briggs Institute's approach for scope reviews (Institute, 2014).

Search Parameters

Only primary and secondary studies were included in the present review, with no period or language delimitations. The reason for the parameter was based into the development of scientific and historical knowledge about *Cannabis sativa* and its worldwide use.

Search Strategy and Databases

Search terms were defined using MeSH terms and DeCS terms, previously to the search strategy development and are available in supplementary material.

Literature was raised into the following electronic database: PUBMED, PUBMED PMC, BVS/BIREME (MEDLINE and IBECS), SCOPUS, Web of Science, EMBASE, Cochrane Library, EBSCOHost (MEDLINE, MEDLINE complete, Academic Search Premier, Regional Busniss News, SocINDEX, Business Source Premier, Newswire, SPORTDiscus).

Inclusion and Exclusion Criteria

References and their abstracts were migrated to Rayyan® (Intelligent Systematic Review), where the duplicities were excluded. The selection was made in paired by two authors (ALMF and CMDS), with blinds on. Once the selection was finished by both authors, the blinds were off and a third author (JAA) solved the conflicts, according to inclusion criteria.

Secondly, to be included articles should present in the abstract the primordial keywords and terms such as “Cannabidiol”, “Cannabis”, “Medical Marijuana”, “Administration, topical”, “Administration, Cutaneous”, “Transdermal Patch” and could present cannabidiol with one or more bioactives isolated. Besides, articles should be experimental.

Therefore, the articles were excluded if were not experimental, such as reviews, books, blogs, articles from newspapers, online sites or other. As much as, if did not mention in the abstract the primordial keywords and terms.

After Abstract selection, the included references were transferred to Excel 2019 (Microsoft Office), for search by full text. Full text selection was made by one author, considered included the articles should fit the following criteria:

- Study type: Experimental studies with animals or *in vitro*;
- Administration path: topical administration (mucosal or skin), also, studies could contain more than one administration path if discussed the different results between them;
- Formulation: Cannabidiol with or without other bioactives; in dosed *Cannabis sativa* extract; in commercial formulations isolated or in dosed *Cannabis sativa* extract;
- Methodology: should contain the treatment used, according to the health condition studied;

Although, articles were excluded if:

- Presented formulation containing more than one medicinal plant and/or which *Cannabis sativa* and/or cannabidiol is not the focus of the study;
- Presented formulation with other bioactives but not cannabidiol;
- Presented synthetic or natural molecules different from those in focus, especially if presents contrary activities from those in focus;
- Full text not available or, if requested, not provided.

Data extraction

Information was extracted by one author, considering the important information regarding the use of *Cannabis sativa* extract or its isolated bioactives, such as: health conditions, treatment regimen, formulation, other methodological approaches that affect the discussion, statistical analysis, main outcomes and results, discussion, and conclusion. Finally, to organize references citation, references were extracted to EndNote X8.

3. RESULTS AND DISCUSSION

3.1. Literature selection

The search in electronic databases resulted in 778 potential references, which 81 were excluded for being duplicates (Figure 1). Then, at the first step, abstract inclusion, only 697

were analyzed in paired, resulting in 169 inclusions. Therefore, the 169 references followed to the second step, the full text analysis, which only 46 articles were included.

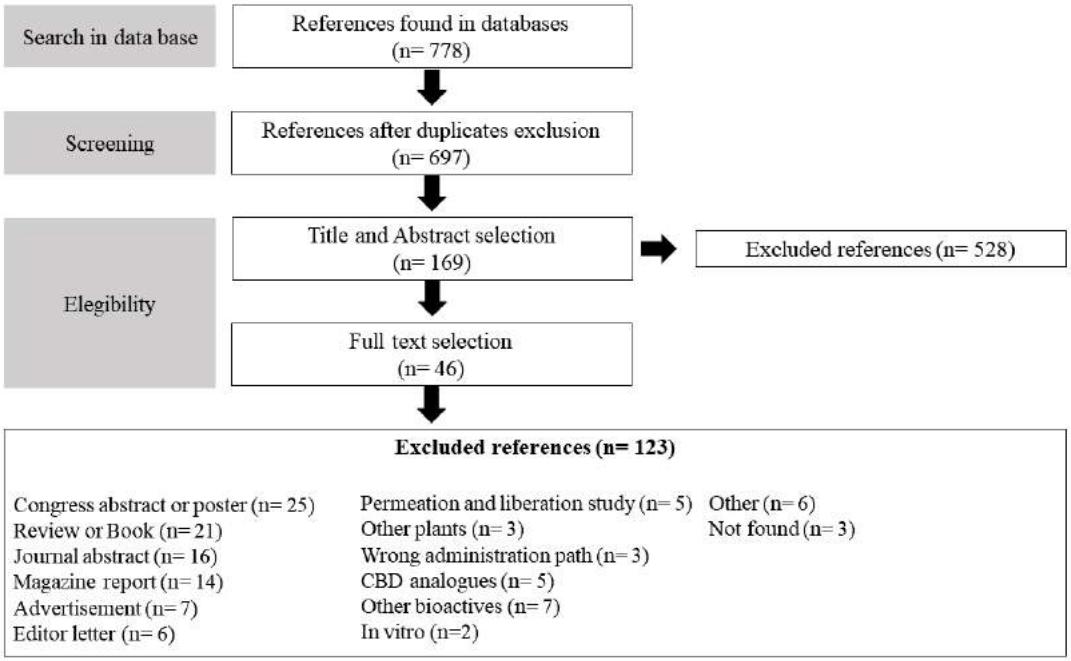

Figure 1: Methodological development and preliminarry results.

The included references were published from 1977-2022 and classified by study type as pre-clinical when conducted with animals (n=21) and clinical when conducted with humans (n=25). The articles will be discussed in the next topics, divided by study type and administration path.

3.2. Pre-clinical Studies: *In vivo*

In order to create a profound discussion about obtained articles, they were organized by similar administration path, since usually aims and methodologies were similar in the same path.

3.2.1. Eye mucosal

Administration of pharmaceutical medication trough this path is mostly employ in order to treat local diseases, more specifically, those located in the anterior segment, since the anatomical and precorneal barrier difficulties the active permeation to reach deeper layers (Ananthula *et al.*, 2009; Gaudana *et al.*, 2010; Thrimawithana *et al.*, 2011).

In literature, diseases treatment basing CBD in *Cannabis sativa* extract and its bioactives were detach in intraocular pressure and corneal hyperalgesia, discussed below. Both diseases are in posterior eyes, which requires measures to improve and facilitate permeation. As seen in

Table 1, some authors introduced an osmotic minipump to drug delivery, this method can be used to induce the drug permeation to reach posterior area of the eye (Thrimawithana *et al.*, 2011).

Intraocular pressure

Predominantly, the studies aimed to determine the effect of cannabinoids in intraocular pressure (IOP). In a few words, IOP is the pressure caused by the aqueous humor on the internal surface of the anterior eye. Pathologies associated with intraocular pressure included glaucoma, uveitis and retinal detachment, especially if not well regulated (Machiele; Motlagh; Patel, 2021). Homeostatic mechanism to maintain the intraocular pressure is associated with the sympathetic nervous system, which influence in aqueous humor secretion. The region where the regulation occur is the juxta canicular region, when stressed the cells initiates a series of responses in cascade leading to the increase of aqueous humor secretion (Machiele; Motlagh; Patel, 2021; Vranka *et al.*, 2015).

According to Green; Bowman and Wynn (1978), only Δ^9-THC and 11- OH- Δ^9 THC were effective in IOP reduction, in a concentration of 0.1% in sesame oil. The same concentration in drug in mineral oil, were able to reduce the intraocular pressure at some point. On the other hand, highest concentration (1%) showed its effectiveness after 2 and 4 hours. Importantly, dose-response spectrum was not what was usually seem at the time, there were no clear correlation among concentration and response. Although, the authors affirmed an existence of correlation between both, which influence the efficacy or potency, the explanation relies in the fact that different concentrations modulate the action of the molecule. In some cases, such as cannabinol, the molecule presented low efficacy at 0.001%, but increased the activity at 0.01%, over than this range, there were no significant activity. Differing from this case, there is cannabidiol, which reduce intraocular pressure discretely and similarly until a 0.01% concentration, but when increased the concentration, there were significant IOP reduction (Green; Bowman; Wynn, 1978).

With these results, the author arranged the molecules studied in order of potency, considering the drug concentration able to lower the IOP at 15% from baseline values was Δ^8-THC, 8α-11-diOH-Δ^9-THC, 8α-OH-Δ^9-THC, Δ^9-THC, 8β-11-diOH-Δ^9-THC, cannabidiol, 11-OH-Δ^8- THC, 8β-OH-Δ^9- THC. Further, those cannabinoids showed great reduction in intraocular pressure at the dose stablished and sustained the activity for 7 hours, which was the stablished experimental time period. Although, ganglionectomized eyes did not respond to Δ^9-THC treatment but did respond to Δ^8-THC in 80% range, when compared to the normal eye.

The ganglionectomy may influence in the nervous system ability to react against the increase of intraocular pressure after stimulation to reduce it, demonstrating lower IOP when compared to normal eyes, which have both mechanisms of intraocular pressure increase and decrease intact (Green; Bowman; Wynn, 1978).

Similar result was observed with 8α and 8β-11-diOH-Δ^9- THC, with a 95% range. As for 11-OH-Δ^8-THC, the response was even for both normal and ganglionectomized eyes (Green; Bowman; Wynn, 1978).

Recently, it has been proven that Δ^9 -THC applied topically reduced intraocular pressure in 30% after 8 hours in male mice (Miller; Daily*; et al.*, 2018). Although, a different efficiency was observed in female mice, leading to a discussion relied into gender dependency. This difference was confirmed by the level of mRNA expression in PCR, which showed reduction of CB1 and GPR18 receptors in female mice. The intraocular pressure reduction by the Δ^9 -THC use was given to the activation of CB1 receptor and GPR18 receptor in combination. As for CBD, male and female wild-type mice showed elevation in IOP at 1 and 4 hours, but in knockout CB1 mice the CDB administration resulted in reduction on IOP in the 1st hour and non-effect in the 4th hour. With those results, the authors discuss that CBD caused raise in intraocular pressure CB1-dependent and non-effect intraocular pressure in animals with GPR18 antagonist O1918. In contrary, the CBD increased the cannabinoid-related lipid species (Miller; Daily*; et al.*, 2018).

However, there are differences in IOP in mice and humans during the day and night. Humans have diurnal habit, thus, IOP peaking happens during the day, on the other hand mice have nocturnal habits, with IOP peaking at night (Miller *et al.*, 2016; Rebibo *et al.*, 2022; Straiker, 2019). A study conducted by Rebibo *et al.* (2022) considered these differences during treatment, the results pointed to significant difference between the prior to treatment measurement and after CBD- contained nanoemulsion treatment began. In fact, compared to the blank nanoemulsion 0.4% CBD decreased IOP in mice after 7, 10 and 14 days of treatment, as much as 1.6% CBD showed the same result after 14 days of treatment. In addition, 0.8% CBD did not present significant results, when compared to the other concentrations used in the study, leading to the conclusion of non-dose-response dependence for CBD activity in IOP reduction (Rebibo *et al.*, 2022).

Corneal Hyperalgesia

Neuropathic pain in the cornea, may cause itch sensation, irritation, dryness, grittiness, burning, aching and light sensitivity. Pain may be caused by corneal hyperalgesia when

stimulates the nerves endings and inducing hyper sensibility, usually, induced by moving air, minimal noxious stimulus and normal light, which is also associated to allodynia (Acosta *et al.*, 2001; Feng; Simpson, 2003; Goyal; Hamrah, 2016; Rosenthal; Baran; Jacobs, 2009)

According to the topical administration results, the application of 0.5% and 1% Δ^8-THC reduced considerably the pain scores but did not prove to be effective in lower concentrations. CBD at 5%, also showed reduction in pain score, but did not showed effectiveness in pain reduction at 3%. Finally, 1.5% HU-308 proved antinociceptive effects (Thapa *et al.*, 2018).

When used separately CBD 5%, Δ^8-THC 1% and HU-308 1.5% demonstrated reduction in neutrophil number, indicating reduction of inflammation response. When evaluates for the mechanism of action, by using an antagonist AM251, it was found that Δ^8-THC acts via CB1R to cause antinociceptive effect, although, CBD 5%, even with the antagonist, was able to reduce neutrophils in corneas. The CBD demonstrated an important action in 5-HT1A to reduce corneal pain and inflammation. As for HU-308 the mechanism of action was through CB2R (Thapa *et al.*, 2018).

With that in mind, the study presented strong information regarding the success in use of Δ^8-THC and CBD into corneal hyperalgesia to reduce pain and inflammation response (Thapa *et al.*, 2018). Unfortunately, there were no other articles to compare their findings, although, the authors provided a novel subject to test cannabinoids as an option of treatment.

3.2.2. Oromucosal

Oral mucosa presents great absorption ability, as an example, sublingual route is commonly use in cases when medications have the need to fast reach blood stream. The reason relies on the fact, that oral cavity presents many blood vessels, further its tissue presents less barriers that would difficulty permeation, such as keratin and tight junctions. In this case the molecular absorption is made by cellular transport which is a great choice for sensible molecules (Brandl; Bauer-Brandl, 2019).

With that in mind, cannabinoids were tested for certain health conditions by oromucosal path (Table 2). According to Brioschi *et al.* (2020) studies the questionnaire results determine a significant reduction in pain severity score (PSS) in dogs for spontaneous osteoarthritis (OA) after CBD administration, when compared to the control group after one, two and four weeks of treatment. Spontaneous osteoarthritis is related to genetic and mechanical impact as influence factors (Conaghan, 2002). In general, these factors, in association, leads to cartilage wear out, especially after stronger impact on joints, as a result, increase the contact between bones in articulation areas (Glyn-Jones *et al.*, 2015). Obviously, the wear of this protection causes

several pains, and may impact into patient's life quality, due to impossibility of perform basic day-life activities (Glyn-Jones *et al.*, 2015; Hiligsmann *et al.*, 2013).

Given the fact, Brioschi *et al.* (2020) introduce a cannabinoid therapy in animals, in order to test its capabilities into pain control, leading to quality of life. Along with the results described earlier, it was observed increase in quality-of-life index (QoL) at all weeks when compared to the baseline. Comparing CBD groups with the baseline results, there were a significant decrease of the pain severity score (PSS) after two and four weeks, leading to the conclusion that CBD may have reduced the pain after a regulated treatment (Brioschi *et al.*, 2020).

As for the pain interference score, the values were lower at all weeks when compared to the baseline. The control groups did not show statistically significant variables at all the scores, but in general, there were a decrease in the pain severity score, in pain interference score (PIS) and improvement on the QoL. In fact, the administration of firocoxib and prednisone, did not affect CBD and control groups in most cases (Brioschi *et al.*, 2020).

In summary, the authors emphasized the perceptive increase in quality of life after dogs received CBD oil through transmucosal path, that is because, the pain levels decrease every week with the treatment, which were not that significant in the control groups. The control groups did not present significant pain decrease, probably because the use of analgesics commonly used in osteoarthritis treatment (Brioschi *et al.*, 2020).

Mechanism of action, on the other hand, was not investigated, although, it was hypostasized that CBD may influence on chronic pain, which intensifies if a non-steroidal anti-inflammatory drug (NSAID) is added, since the inhibition of COX-2 would prolong the cannabinoids action (Brioschi *et al.*, 2020).

3.2.3. Rectal mucosal

Rectal mucosal presents two hemorrhoidal vein that may absorb the drug after administration. Middle and inferiors hemorrhoidal veins lead the drug directly to circulation, especially lipophilic drugs. Also, if inserted correctly, this path is advantageous since avoids the first-pass metabolism. On the other hand, the wrong insertion may cause the metabolism of the drug, that is because the superficial hemorrhoidal vein, also located in the rectum, may drain the drug to portal circulation (Jannin *et al.*, 2014; Purohit; Hanning; Wu, 2018; Van Cauwenberge *et al.*, 2004; Van Hoogdalem; De Boer; Breimer, 1991).

Therefore, Schicho and Storr (2012) were the only ones to determine cannabinoids, specifically CDB, effects in colitis inflammation, administrated through rectal mucosal and oral

path to compare both effectiveness as seen in Table 3. In summary, colitis, a chronic inflammatory bowel condition, is characterized by blood and mucous diarrhea, with non-determined etiology (Teixeira; Hosne; Sobrado, 2015).

After colitis induction, the results indicated relation between mice weight lost and the condition, on the other hand, the weight loss was not related to CBD treatment or its vehicles. Intraperitoneal CBD administrations improved the colitis score and decrease myeloperoxidase (MPO) activity, demonstrating reduction of inflammation and oxidative process (Schicho; Storr, 2012).

In addition, the histological assessment presented lower destruction areas of the epithelial lining, reduction in colon thickness and infiltration of monocytes than compared to its vehicle. In contrary, CBD treatment by intragastric administration did not improved colitis inflammation (Schicho; Storr, 2012).

Although, an interest result regarding the intrarectal CBD treatment was obtained, a slightly important improvement in the colon inflammation was seen, that was confirmed by the reduction in myeloperoxidase activity and the histological assessment showing reduction in the leukocyte infiltration. Further, the crypt architecture was partially preserved in comparison to its vehicle (Schicho; Storr, 2012).

According to the results obtained, the rectal administration of CBD, could be a great choice to colitis treatment, even though, the path is still behind from the CBD intraperitoneal administration (Schicho; Storr, 2012).

3.2.4. Intranasal mucosal

As well as oral mucosa cavity, intranasal cavity presents a high surface area with a considerate amount of blood vessels, which favors drug absorption and its referral to circulation. Then, this pathway is a great choice for those molecules that pass through the first-pass metabolisms or if has poor solubility and easy gastric degradation, further, may reduce side effects (Bitter; Suter; Surber, 2011; Liu; Yang; Ho, 2018).

As seen in Table 3, author determine the cannabinoids effects in multiple sclerosis (MS), which is a disabling condition of the brain and spinal cords caused by an autoimmune response. Immune system does not recognize the myelin covering the nerves, responding with an attack mechanism, leading to nerves demyelination and inflammation process. Therefore, the flux of nervous impulses is compromised, reducing the responses after a stimulus and after brain command. With time, the nerves deteriorate increasing motor and sensorial damage (Ashtari *et al.*, 2020; Frohman; Racke; Raine, 2006; Karussis, 2014; Steinman, 2001).

In order to study the multiple sclerosis, scientist develop a model by inducing autoimmune encephalitis (EAE) in animals, by administration of soluble myelin-derived proteins, myelin-derived peptides in adjuvant, or passive transfer of activated myelin- specific CD4 (Robinson *et al.*, 2014).

With that in mind, Duchi; Ovadia and Touitou (2013) applied the model MS-EAE in order to study the potential of CBD (and other drugs), into a nasal delivery system (NDS) as a new potential treatment for multiple sclerosis (methodological details described in Table 2).

According to the results, administration of the glatiramer acetate (GA), a medication for reduce multiple sclerosis relapse, into nasal delivery system demonstrated great decrease of the encephalomyelitis scores when compared to the other administration groups for this drug. In comparison, the CBD demonstrated better decrease in encephalomyelitis score when associated with glatiramer acetate by nasal delivery system, then in others administration paths (Duchi; Ovadia; Touitou, 2013).

Administration of CBD alone in nasal delivery system reduced the expression of inflammatory cytokines, IL-6 and TNF-α, in the cerebellum tissue of encephalomyelitis mice. Similar results were obtained with the nasal delivery system combination of CBD-GA in the preventive use of prednisolone. Further, the nasal administration of glatiramer acetate (13.7 mg/mL) and prednisolone for preventive purpose, reduced the inflammatory cell infiltration (Duchi; Ovadia; Touitou, 2013).

It is possible to assume that the combination of CBD-GA could bring benefits to multiple sclerosis treatment, including increasing neurogenesis, and could be considered for clinical trials as a novel treatment for multiple sclerosis condition (Duchi; Ovadia; Touitou, 2013).

3.2.5. Transdermal

Recently, many studies have been experimenting transdermal route, as pertinent pathway to drug delivery, since it does not impose administration discomfort, reduces adverse events, sustains drug delivery for a long time, reduces drug interaction and first pass metabolism. Transdermal delivery counts with cell and drug interaction in order to absorb the molecule to reach circulation, using mechanisms of transcellular and/or intercellular route (Frasch; Barbero, 2013; Pan *et al.*, 2020; Prausnitz; Langer, 2008).

References using transdermal delivery of cannabinoids focused mostly into the pharmacokinetics of the formulation, mentioning a discussion regarding the molecule permeation into the skin, its accumulation and plasma concentration (Table 3).

Pharmacokinetics

According to Lodzki *et al.* (2003) CBD-in-ethosomes permeation studies showed great accumulation of the drug in the abdominal muscle, skin, and hip skin. The distribution of CBD was observed, predominantly at hip muscle, liver, and pancreas.

After application, the steady state of transdermal formulation of CBD was reached in 24 hours and lasted until 72 hours, stabilizing in a concentration of 0.67 µg/mL. Although, the transdermal dose of CBD detected after 12 hours was 1.37 mg ± 0.72 mg, a dose equivalent of 45.7 mg/kg, considering the mice body weight of 300 g, and after 72 hours was 2.60 mg ± 0.79 mg, a dose equivalent of 86.7 mg/kg, considering mice body weight of 300g.

In the same way, Hammell *et al.* (2016), used a gel with 1% or 10% of CBD, administrated in different doses (0.62; 3.1; 6.2 and 62.3 mg/day) and observed the following plasma concentration of CBD: 3.8 µg/mL ± 1.4 µg/mL (in 9 rats, at 0.62 mg/day), 17.5 µg/mL ± 4.4µg/mL (in 8 rats, at 3.1 mg/day), 33.3 µg/mL ± 9.7 µg/mL (in 8 rats, at 6.2 mg/day) and 1629.9 µg/mL ± 379.0 µg/mL (in 4 rats, at 62,3 mg/day), after 4 days of treatment.

In addition, Hannon *et al.* (2020) found, after six hours from transdermal formulation application, traces of CBD in plasma samples. Concentration reached an amount of 12.8 µg/mL at the end of 1 week and 10.6 µg/mL in the end of 2 weeks. Results regarding THC detection demonstrated concentration values below the lower limit of quantification in both weeks (Hannon *et al.*, 2020).

The authors compared the pharmacokinetics of CBD and THC to its acid derivates, cannabidiolic acid (CBDA) and tetrahydrocannabinolic acid (THCA), respectively. CBDA showed mean concentration of 32.4 µg/mL and 21.7 µg/mL in the first and second weeks, respectively, and THCA 3.8 µg/mL and 3.1 µg/mL, respectively (Hannon *et al.*, 2020).

Statistical analysis confirmed that there was no difference in cannabinoids concentration when compared along the weeks. However, CBDA and THCA showed better absorption results then CBD and THC. Importantly, some animals experimented erythema, a common side effect to transdermal ointment formulation specially with extracts containing terpenes or ethanolic extracts that may cause skin sensitivity (Hannon *et al.*, 2020).

Considering the previous studies, it is important to emphasized that differences among formulation content and administration path could affect the cannabinoids pharmacokinetics. Therefore, Bartner *et al.* (2018) after comparing formulations of transdermal cream, infuse-oil and microcapsules for oral administration (all containing CBD), observed that the media of the maximum concentration and standard deviation of all three formulation were lower at 75 mg q12h groups when compared to 150 mg q12h group values. Among the formulations CBD-

infused oil was the one with less inter-individual variability of CBD exposure. Statistical difference of the plasma concentration among the formulation's groups showed a higher amount of CBD levels in the CBD-infused oil then other formulations, without significant difference at 4 and 6 weeks in the 75 mg q12h group. Besides, all the statistical differences were found in the time points, doses, and all formulations, with exception of the 6 weeks at 150 mg q12h of all formulations.

With that in mind, the oral administration was the worst path of CBD absorption, and, in contrary, transdermal paths avoided the first path effect from the liver. Although, the transdermal formulation did not achieve similar plasma concentration as the other formulations, the authors discuss the event due to the thickness of the pinnae skin and a reduced capability of the cream to permeate. Formulation behavior will be discussed in a specific topic, disposed ahead.

Dose difference could influence in the cannabinoid's effects and its permeation into the skin, thus, the difference among studies could influence the comparison between results. Further, different animal and number of populations could influence data collection.

Importantly, in the Gonzalez-Cuevas *et al.* (2018) experiment, there were long lasting CBD concentration in plasma, authors attributed this fact due to its high lipophilicity, which allows it to stay longer available in the brain and plasma even for a few days past the treatment. Corroborating to the results of Hannon *et al.* (2020), who found traces of CBD, 6 hours after treatment. Also, the accumulation rate found by Lodzki *et al.* (2003) may be related to the fact that ethosomes has lipophilic characteristic, which associated to the CBD, increase its accumulation capability.

On another point of view comparing intranasal and transdermal administration paths, Paudel *et al.* (2010) studied the CBD absorption and pharmacokinetics. First, nasal absorption of CBD was faster, identified in less than 10 minutes and was detected after 0.5 minutes of treatment, specifically. On the other hand, transdermal absorption resulted in a mean plasma concentration of 6.3 ng/mL, which were attained at 15.5 hours, and maintained for 48 hours after gel application, declining only after 6 hours of the gel removal. These results indicate a skin reservoir of CBD, due to its high lipophilic properties, that allows permeation through the stratum corneum but slow permeation trough dermis.

In this perspective, in treatments using CBD intranasal or transdermal formulation, if the need of CBD use is immediate, that is, the needed to reach immediate results, intranasal showed to be the best choice. On the opposite, in chronic treatments that reacquires slow drug delivery, the transdermal pathway is the best choice.

Anti-inflammatory

Considering the pharmacokinetics studies, Lodzki *et al.* (2003) and Hammell *et al.* (2016) determined the potential anti-inflammatory effects of cannabinoids in the concentration found. Initially, permeation and distribution were considered adequate to the anti-inflammatory effect expected (Lodzki *et al.*, 2003).

In this case, the anti-inflammatory effect of CBD, showed that the edema caused by the stimulation with carrageenan, was prevented in the pretreated CBD groups, starting significantly after 1 hour and lasted until the end of the inflammation course (Lodzki *et al.*, 2003).

In addition, Hammell *et al.* (2016) determinate the anti-inflammatory effect of CBD in monoarthritic induced rats, using complete Freud's adjuvant (CFA). After induction, there were noticed significant swelling in the side ipsilateral. Although, after four days of 6.2 mg/day and 62.3mg/day CBD treatment, the knee joint showed reduction of circumference, supposing a reduction in the inflammatory process, the opposite was seen with lower dose administration of CBD (Hammell *et al.*, 2016).

In accordance to that result, the histological evaluation showed that the synovial membrane was thickened after 7 days of intra-articular complete Freud's adjuvant injection, and reduced after 4 days of CBD 6.2 mg/mL treatment, and no changes was observed with low dose CBD administration (Hammell *et al.*, 2016). In addition, the spontaneous pain was higher in day 3 in all animals, but after 4 days of 6.2 mg/mL CBD administration, the pain score reduced significantly, especially when compared to the control group (Hammell *et al.*, 2016).

In association to the anti-inflammatory effect, the CBD in 62.3 mg/day demonstrated great reduction of pro-inflammatory cytokines such as TNFα when compared to the non-treated monoarthritic groups. Other important effect of CBD were the recoveries of heat hypersensitivity in the rats' paws, caused by the monoarthritic induction, further, CBD treatment did not alter the activity levels or motor abilities (Hammell *et al.*, 2016).

With these results, both authors brought up the fact that the transdermal administration of CBD were effective against the inflammation caused by edema induced by carrageenan (Lodzki *et al.*, 2003) and arthritis induced by complete Freud's adjuvant (Hammell *et al.*, 2016).

Relapse-promoting conditions

Other CBD effects were detected by Gonzalez-Cuevas *et al.* (2018), in general the cannabinoid exhibited positive effect in relapse-promoting conditions, such as: stress, anxiety and impaired impulse control. According to the authors results, the CBD could be useful as

therapy for addiction treatment across several drugs of abuse, due to reduction of vulnerability states that may cause relapse.

In alcohol and cocaine groups, CBD showed reduction of yohimbine effect of stress induction which increases the drug seeking. When compared to vehicle groups, demonstrated reduction of reinstatement in cocaine and alcohol groups.

Neurodegeneration

Neurodegeneration process may be caused by a diversity of conditions, especially motor neuron diseases, such as Alzheimer, Parkinson, Multiple Sclerosis, Amyotrophic Lateral Sclerosis (ALS) and others, leading to progressive motor disability (Mayeux, 2003). As seen before, previous authors studied the effect of CBD nasally applied in induced- multiple sclerosis.

In the study conducted by Liput *et al.* (2013), the animals achieved an intoxication level in which they delayed righting reflex and ataxia. There was a significant difference among ethanol/vehicle groups and ethanol/1% CBD, indicating great effect of the treatment. However, according to the statistical analysis, the treatment with CBD transdermal gel and transdermal vehicle, did not show alteration in intoxication effect or pharmacokinetics of ethanol in both experiments.

Neurodegeneration in the entorhinal cortex detection assessed by Fluoro- Jade B (FJB), described as FJB+ cells, revealed statistically similar relevance in ethanol only and ethanol/vehicle gel groups, the same relevance was observed among animals treated with 1% or 2.5% CBD gels (Liput *et al.*, 2013).

On the opposite side, 5% CBD gel treatment reduced in 48.8% the amount of FJB+ cells, which characterize neuroprotective effects. In accordance with that, pharmacokinetics tests showed higher CBD plasma concentration at 5% then at 1% in gel. Interestingly, diet showed interaction to CBD (Liput *et al.*, 2013).

Further, FJB+ cells were found in neurodegenerative tissue along the entorhinal cortex, after four days with ethanol exposure. In fact, when compared to the control (ethanol only and ethanol/vehicle gel) the other groups with treatment collapsed since the control groups received the sample trough different administration path. Both CBD intraperitoneal injection and transdermal delivery showed reduction of FJB+ cells when compared to its control groups, although, that were not statistically significant. Although, it was possible to measure its neuroprotective effects and both showed similar neuroprotection capacity (Liput *et al.*, 2013).

Information presented earlier proved that CBD associated with glatiramer acetate could be a great choice to multiple sclerosis treatment after intranasal administration, in encephalomyelitis models (Duchi; Ovadia; Touitou, 2013). In this case, after topical administration, the pharmacokinetics showed that highest concentration of CBD in plasma, was approximately 8.3 ng/mL (Giacoppo *et al.*, 2015).

During treatment, CBD treated groups showed allergic reactions in the application location or in systemic level. However, the concentration in animals with encephalomyelitis treated with CBD, showed great recovery throw time and less disability to encephalomyelitis when compared to untreated encephalomyelitis mice. In fact, encephalomyelitis mice treated with CBD-cream presented significant response in the needle test mechanical allodynia, when compared to encephalomyelitis untreated group (Giacoppo *et al.*, 2015).

Histopathology assay of encephalomyelitis, untreated groups presented significant demyelination and axonal structures in the spinal cord, on the other hand, 1% CBD-cream treated groups showed reduction of the demyelination process and axonal loss caused by encephalomyelitis. In the same way, cell infiltration was detected in EAE untreated mice groups when compared to the encephalomyelitis treated with CBD groups, the last one, showed complete resolution of inflammatory cell infiltration (Giacoppo *et al.*, 2015).

So, evidence pointed a great response of CBD treatment in multiple sclerosis in both administration paths: mucosal and skin, especially for reduction in nerve preservation, inflammation reduction and for pain control.

3.2.6. Topical Skin

Usually, topical skin drug administration is used to treat local conditions, in this case there are no interest into absorption, or a minimum absorption. Although, that path can be used as a transdermal route, according to the formulation used, as seen in Table 4 authors usually went for local conditions, but in some cases, absorption was required.

Anti-inflammatory

According to previous information presented, cannabinoids performed interesting anti-inflammatory effects, the same effect was observed in studies applying cannabinoids through topical skin. For instance, Tubaro *et al.* (2010) studied cannabinoids potential in reduce oedema induced previously in animals' ears, and determine that, cannabinoids, considered psychoactive, such as Δ^9-THC, Δ^8-THC and Δ^8-THCV (tetrahydrocannabivarin), reduced

significantly oedema with a dose-dependent response, meaning better results in the maximum concentration (1 μmol/cm²).

On the other hand, the non-psychoactive cannabinoids, CBC, CBD, CBCV (cannabichromevarin) and CBDV (canabidivarina) demonstrated lower potency in the highest dose (1 μmol/cm²) dose, with 40% of oedema reduction for CBC and CBCV, 36% for CBD and 29% for CBDV. When compared to the psychoactive cannabinoids the response was lower (20-70%), although the oedema reduction was significant (Tubaro *et al.*, 2010).

Authors used indomethacin as a comparative medicine that is usually used for anti-inflammatory effects and obtained an anti-inflammatory response of 22%-86% of oedema reduction, also dose-dependent. In comparison, the indomethacin group demonstrated better oedema reduction and more potency then both cannabinoid groups, although, both cannabinoid groups demonstrated relevant oedema reduction (Tubaro *et al.*, 2010).

In addition, Mciver *et al.* (2020), reinforce the anti-inflammatory effect of CBD by CB2 receptor suppression. In this case, the authors aimed to determine the wound healing potential of CBD extract, as pointed, there were no differences in the daily mean wound area after treatments. The differences appeared after 7 days of treatment and the mean area decreased after 14 to 42 days.

Overall, healing process was slow until day 21 and increased faster after this period. In contrary, there were no significant differences in the overall healing rate between any of the treatment groups, the same thing happens to total days to healing. Experiments demonstrated that inflammation reduction was probable due to CBD activation of CB2 receptor, although, it did not demonstrate influence in healing process (Mciver *et al.*, 2020).

Besides, a study presented a possible multidirectional anti-inflammatory effect of CBD after topical application in UVA/B exposure skins. Plasma samples demonstrated downregulation of relevant phospholipids (lyso-PE and lyso-PC), prostaglandin and thromboxane, although, demonstrated upregulation of anti-inflammatory lipoxin ($LPXA_4$) (Łuczaj *et al.*, 2021).

With the in mind, the evidence of CBD experiments in animals, showed a significant anti-inflammatory effect, after its skin administration.

Antibacterial

Considering the wound healing process discussed before, it is important to emphasized that wound contamination may accelerate the healing process by increasing the inflammatory activation in the first hours. In that case, if CBD causes systemic effects, it is likely that reduced

the number of bacteria and inflammatory activity, which caused the wound healing retardation. So, if CBD is applied into an open wound, there are big possibility of systemic effects (Mciver *et al.*, 2020).

In fact, Blaskovich *et al.* (2021), demonstrated that CBD reduced gram-negative bacteria, including the *N. gonorrhoeae,* one of the most resistant bacteria, at inoculated mice, after topical application. Further, in gram-positive bacteria, CBD showed high spectrum. The authors specified that the CBD repeatedly application did not lead to resistance.

The antimicrobial effect is a different activity related to CBD with promising results, however, it would be necessary more experimental information to be deepen this knowledge.

Antioxidant activity

This function is mostly related to the oxidative process caused by the ultraviolet (UVA and UVB) rays with sun contact. In summary, the ultraviolet rays are part absorbed through the skin, induce generation of reactive oxygen species (ROS) leading to an oxidation reaction in cells. In long term, the ROS may promote DNA damage, which is associated to genetic mutations and, possibly, melanoma formation (Romanhole *et al.*, 2020; Trojahn *et al.*, 2015).

According to Atalay *et al.* (2021) when compare the control groups (with non-exposure to UVA/B rays) to UVA/B exposure groups, the first one demonstrated better number of certain proteins then the number of proteins in the second groups. In this case, results indicate protein synthesis stimulation and protein degradation, respectively.

Cannabidiol treatment group, after UVA and UVB sun exposure demonstrated decreased of pro-oxidant activity, not only by reducing the NADPH-dependent diflavin oxidoreductase 1, which generates superoxide anion radical, but also, increasing the antioxidant enzymes expression such as Cu,Zn-SOD (Atalay *et al.*, 2021).

Further, CBD also improved the expression of ubiquination-related proteins, which regulates the proteins in cells. In fact, the CBD affects the apoptosis paths, by reducing the BcL-2 and the caspases recruitment, causing the opposite of UVA exposure effects (Atalay *et al.*, 2021).

On the other hand, Łuczaj *et al.* (2021) provided information regarding the CBD effect in the changes of phospholipid and ceramide metabolism in plasma, after UVA/B exposure. In general, metabolic disturbance were prevented when CBD was applied, after UV radiation. According to the results, the downregulation of phospholipids types lyso-PE and lyso-PC indicates an anti- inflammatory effect of CBD, and the upregulation of

phosphatidylethanolamines (PEo), which reduced the ROS property, indicates antioxidant stimulation (Łuczaj *et al.*, 2021).

Even though the study informed promising UV protective effects from CBD application, more information regarding this use is necessary to consolidate the potential use.

3.3. Clinical studies

3.3.1. Eye mucosal

After an extensive pre-clinical investigation of Δ^9-THC in IOP, discussed before, Green and Roth (1982) improve the method to a clinical case-control trial, to investigate the effect of Δ^9-THC in IOP (Table 5). First, the toxicity study presented a slight mydriasis, for both treated and vehicle eyes, after 1 hour of treatment. At the same time, there were a slily increase in conjunctival injection.

Burning and oily eye sensation, with tearing and runny no lasting about 30 minutes was described by the volunteers, after Δ^9-THC treatment, although, no systemic changes were evidenced (Green; Roth, 1982).

More importantly, no changes were observed in IOP, neither in treated nor vehicle eyes. Then, authors discuss that the topical application of Δ^9-THC may not cause effect in IOP but may depend on the amount of application dose and posology. One more time, emphasized the hypothesis that the drug may have difficulty in penetrate in the cornea in a concentration that would cause significant effect (Green; Roth, 1982).

In comparison to the promising results in pre-clinical studies, the effects in IOP after clinical study conduction, were not promising. On the other hand, considering the authors position about the influence of formulation and posology, further investigation is needed.

3.3.2. Oromucosal

Stiff-person Syndrome (SPS)

A case report described by Vicente-Valor *et al.* (2013) has Sativex® (Sativex®, GW Pharma Ltd., Wiltshire, UK) introduced as therapy in order to control the extreme pain. Sativex® is a commercial spray formulation of *Cannabis* extract, that contains, mostly, THC and CBD, but also, minor cannabinoids and other bioactives (Boivin, 2021).

In stressful days, the patient self-adjusted his doses and achieved the maximum of 6 sprays without adverse events reported. Importantly, quality of life evaluation capture improvement; first, considering that before treatment patient was wheel-chair dependent and

suffered from muscle spasms and pain, in comparison, after treatment, the wheel- chair was not needed and the patient reported reduction in pain. Then, there were improvement in gait and range of motion. Unfortunately, the SPS etiology remain uncertain, from the exams presented, autoimmune-mediated mechanism is suggested by antibodies and other autoimmune comorbidities (Vicente-Valor *et al.*, 2013). This case report may be a tool for further studies to consider Sativex® as a treatment for pain control caused by this condition.

Multiple Sclerosis (MS)

Effectiveness of *Cannabis sativa* bioactives were observed in spasticity and neuropathic pain caused by MS.

Spasticity For instance, bladder spasticity, is an uncomfortable event caused by multiple sclerosis, among the symptoms is possible to cite the urinary emergency and frequency, pain or discomfort, leakage of urine and others. Administration of THC:CBD oromucosal spray showed important reduction in the symptoms, according to the overactive bladder symptom score (Maniscalco *et al.*, 2018).

According to the study of Maniscalco *et al.* (2018), post residual volume reduced after treatment started, as much as the MS spasticity, with a score improvement from 8 to 6 in the number rate scale. Moreover, 14 patients (out of 15) showed improvement of 20% or more at 0-10 number rate scale spasticity score measured after 4-week treatment, also mobility was improved from 33 to 24 seconds (Maniscalco *et al.*, 2018).

A treatment advantage was that no significant adverse events were recorded by patients. These results showed that THC:CBD facilitated bladder emptying as the urodynamic showed reduction of post-void residual volume and increased bladder volume and its compliance (Maniscalco *et al.*, 2018).

In accordance, Riva *et al.* (2019) used the Modified Ashworth Scale, which rate the spasticity score, the results pointed to improvement in patients treated with Nabiximol spray when compared to placebo group. Nabiximol is another nomenclature adopted to refers to Sativex®, then, formulation and bioactives concentration remains the same (Boivin, 2021).

Although, a few patients discontinued treatment with Nabiximol due to adverse events, such as nausea and anxiety, disease progression, asthenia, dizziness, somnolence, vertigo, muscle spasticity or rigidity and dry mouth. Even with these adverse events, in general, authors reported that in both phases Nabiximol was well tolerable (Riva *et al.*, 2019).

Neuropathic pain Considering patients with an advanced MS condition, in the clinical trial conducted by Rog *et al.* (2005), the treatment with THC: CBD spray (2,7mg: 2,5mg)

reduced neuropathic pain scale and numerical rating scale for pain. Intention to treat data analysis presented 89% of patients added reporting dysesthetic pain and 11% painful spasms.

Other effects evaluated by the neuropathic pain scale pointed improvement such as sleep disturbance. However, at least one adverse event was found in 30% of treatment patients, and only 68,8% in placebo group, the most common event reported was nervous system disorders (dizziness) in treated and placebo groups. No fatal, life-threatening, or resulting in persistent disability or hospitalization occurred, further, there were no significant biochemical differences in blood samples and vital signs. With that in mind, it is possible to assume that the treatment was well tolerable (Rog *et al.*, 2005).

No significant differences in the neuropsychological outcomes were found by the Selective Reminding Test, although a unique difference was found in a mean improvement in placebo group that did not match the treated groups. Then, the treatment with THC: CBD did not appear to significantly affect the MS-related neuropsychological outcomes measured. Furthermore, long term use of THC: CBD did not show significant effects, it requires more studies to evaluate this information (Rog *et al.*, 2005).

Clinical trials conducted by Nurmikko *et al.* (2007) and Langford *et al.* (2013) obtained similar results. Regarding Rog *et al.* (2005) study in long-term use of THC:CBD showed non pain control, Nurmikko *et al.* (2007) determined that long-term use (from the period of 871 days) of Sativex® was effective in maintenance of pain reduction.

In addition, the group investigate Sativex® effects in allodynia, but the difference among control and treated groups were not significant. Besides, patients in Sativex® treatment group discontinued the treatment due to adverse events, non-compliance, and lack of efficacy (Nurmikko *et al.*, 2007).

Among the adverse events, patients reported several gastrointestinal discomforts (nausea, vomit, diarrhea and constipation), nervous system (severe and mild- moderate psychiatric) and oral discomfort. Ischemic attack, a serious adverse event, was reported by one patient (Nurmikko *et al.*, 2007). Gastrointestinal, oral discomforts and nervous system affections (dizziness, impairment of balance, nausea and intoxication feeling) was also reported as adverse events by patients in Rog; Nurmikko and Young (2007) and Langford *et al.* (2013) clinical trials.

Specifically, in the Langford *et al.* (2013) study, the phase A of the trial, did not proved effectiveness of THC: CBD spray in chronic neuropathic pain, although, the opposite was observed in the phase B of the trial. That is because, in phase B, patients were asked to maintain

certain dose of the spray, which were not requested in phase A, in that case, the analgesic effect was significantly higher.

On the other hand, a case reported a 53-year-old patient with relapsing multiple sclerosis diagnosis in 1999, treated with the highest dose of tizanidine, baclofen and benzodiazepines. In 2012, started a treatment with Sativex®, adjusting the optimal tolerance of 6 sprays a day. However, 4 weeks after treatment started patient presented convulsive seizure, thus, treatment with Sativex® was discontinued. Discussing this case, authors determined an impossibility of seizure induction by Sativex® use, due to the patient conditions, there were a like hood of such situation happens caused by its previous conditions (Aparicio Rosana; Polo Virginia, 2013).

According to the literature information presented, neuropathic pain and spasticity showed to be controlled using THC: CBD spray combination, including the commercial formulation. However, important reports of adverse events were mentioned and could lead to treatment discontinuation in long-term use. To minimize adverse events, an option is an adjustment in formulation, or even, addition of co-medication to reduce the specifical adverse events caused by the spray.

Epidermolysis bullosa (EB)

In general, the epidermolysis bullosa (EB) is a rare genetic condition characterized by the fragility of affected tissues. In this condition, any trauma causes painful ulceration, erosions and mucocutaneous blistering (Bardhan *et al.*, 2020). Literature raised targeted the *Cannabis sativa* bioactives capacity of wound healing, besides the pain control.

A case series conducted by Schräder (2020) presented the improvement in wound healing of patients with EB after sublingual administration of *Cannabis* based medicine containing THC and CBD. All patients did not present satisfactory effects with preview analgesia prescribed. Patients using sublingual CBM oil reported pain relief, reduction of pruritus and urges to scratch. Although, one patient had to supplement the therapy with oxycodone after 6 months of single use of CBM oil (Schräder, 2020).

A third patient reported side effects with previous prescribed treatment, leading to the use of CBM, which decrease pain and side effects. Specifically, this patient had skin carcinoma in advanced phase, then, the concentration of CBM oil had to be adjusted, reducing pain in a few days after the adjustment. Unfortunately, the carcinoma caused metastasis and leaded to his death (Schräder, 2020).

Considering the effects of CBM in patients with EB, it is possible to assume that it may increase life quality, by reducing pain from the mechanism already known. The combination of

CBD and THC may strengthen medication, inducing the cannabinoids 1 and 2 receptors and induce beta-endorphin production. Further CBD may also reduce the undesired effects of THC, such as sedation and intoxication (Schräder, 2020).

3.3.3. Transdermal

A clinical trial conducted by Heussler *et al.* (2019) (Table 7) aimed to test a novel formulation of CBD gel in volunteers with Fragile X Syndrome (FXS), a rare genetic condition caused by repetition of cytosine-guanine-guanine in FMR1 gene on X chromosome (Heussler *et al.*, 2019; Steinberg; Webber, 2013). Safety and tolerability studies indicated that 85% of patients demonstrated at least one adverse effect such as, gastroenteritis, vomiting, upper respiratory tract infection or other. during the treatment period, from which 30% of patients experienced at least one adverse effect that could be related to CBD gel treatment. The adverse effects were resolved by dose adjustment. Other adverse effects classified as no serious included: gastroenteritis, vomiting and upper respiratory tract infection in about 10% of patients.

However, authors affirmed that ZYN002 (CBD gel) use was well tolerable and clinically safe. Parameters such as echocardiograms, physical, neurological exams, and vital signs showed no relevant changes. Further, laboratory test only reported increased of eosinophil count after 83 days in patients with a moderate rash but normalized one month after the last dose administration (Heussler *et al.*, 2019).

In addition, subscales demonstrated great results after 12 weeks of treatment when compared to the screening. Anxiety, Depression and Mood Scale subscales presented significant reduction in all parameters: manic/hyperactive behavior, social avoidance, general anxiety, and compulsive behavior. In accordance with these results, the depressive mood did not reach statistical significance. Further, Aberrant Behavior Checklist-Community for FXS subscale, showed reduction in its parameters, including stereotype, social unresponsive, irritability, hyperactivity, and inappropriate speech, as much as in Pediatric Anxiety Rating Scale (PARS-S). On the other hand, a few parameters of the Pediatric Quality of Life Inventory (PedQLI) subscale did not reached improvement such as physical functioning, school functioning and social functioning (Heussler *et al.*, 2019).

With that in mind, transdermal use of CBD in gel formulation manage to improve emotional and psychological symptoms related to Fragile X Syndrome patients, showing great bioactive bioavailability after absorption, and great tolerance after administration.

On the other hand, a study conducted by Scheffer *et al.* (2021) aimed to evaluate the CBD transdermal gel treatment to children presenting epileptic encephalopathies. Overall, 46 patients were included in the study, and after 6.5 month of treatment the median seizure reduction was 12.3%. Among seizure types, that showed relevant response, was focal impaired awareness seizures (44.5%) and tonic-clonic seizures (22.7%), compared to the baseline. In addition, 33 patients had both seizure types and showed reduction in 43.5%, at 2 months a reduction of 44.4% and 5 months 57.7%. Concomitant use of clobazam improved seizure frequency. Besides, seizure severity, children's behavior and mood were also modified after implementation of CBD transdermal gel therapy. Further, parents and/or caregivers rated qualitatively an important improvement in school, engagement or participation, cognition, energy, and other measurements (Scheffer *et al.*, 2021).

Further, topical CBD for thumb basal joint arthritis-related pain, resulted in significant improvements in pain, disabilities of the arm, shoulder, and hand compared to the control. According to the score previous stablished, pain (rate of 0 to 10) was reduced from 5 at baseline to 2 after CBD cream treatment, indicating 60% pain reduction (as lower the rate score, less disability). Also, disabilities of the arm, shoulder, and hand (rate of 0 to 100) were from 36 at baseline to 22 with the CBD cream, referent to 39% reduction (Heineman *et al.*, 2022). Single assessment numeric evaluation (range 0 to 100), increased from 67.5 at baseline to 78.5 with the CBD cream treatment, meaning 16% increase at global well-being. Thus, this trial resulted in improvements in thumb basal joint arthritis-related pain and disability without adverse events (Heineman *et al.*, 2022).

3.3.4. Topical Skin

Wound Healing

Case series reported the use of commercial formulations in patients with Pyoderma Gangrenosum (PG), which is a neutrophilic dermatosis responsible for the development of painful wounds (Maverakis *et al.*, 2020; Wang; Maverakis, 2018). Patients were treated with Bedrolite® and Argyle™ (THC: CBD) in patient-dependent concentration and posology. Bedrolite® is a *Cannabis* derived product, considered CBD-only (it contains less than 1% THC and 9% CBD), leading to a less psychoactive product (Bedrocan® international, 2021). Argyle™ on the other hand, are gel capsules containing THC and CBD in similar concentrations (Tweed Inc., 2022).

Included patients did not showed pain control with previous used corticosteroids. Cannabinoids medications use pointed to pain scored reduction after a few weeks of treatment,

although no wound healing improvement was reported (Kiefer, 2017; Maida; Corban, 2017). Analgesic effect was attribute to the interaction of cannabinoids with receptors expressed on peripheral nociceptors and immune cells (Maida; Corban, 2017).

On the other hand, case series study reported the CBD self-medication to painful wound caused by Epidermolysis bullosa (EB) in children, patients reported blisters reduction, wound healing, pain and itching sensation reduction (Chelliah *et al.*, 2018). Although the mechanism of wound healing was not elucidated, but on the bright side, no adverse events was reported (Chelliah *et al.*, 2018). Same results were observed in patients using *Cannabis* based medicine, containing THC and CBD (Schräder, 2020).

Wound healing was also reported by Maida *et al.* (2020), after the use of *Cannabis*. Ulceration in legs in both reported patients was absolutely closed in a range of 75,6 days, and 50% closed in a range of 35 days. Patients demonstrated similar period of healing and pain control, which was achieved after about 18-19 days after treatment beginning. In addition, planimetric wound image analysis showed that granulation dominates the first treatment half, and them, second half showed reepithelization covering the granulated tissue.

Patients did not record side effects, which was attributed to its non-invasive and non-systemic capacity. Also, the benefits caused by the treatment was related to synergism among the bioactives present in the formulation: cannabinoids, flavonoids, and terpenes. The authors discuss that cannabinoid, such as CBD and THC, caused many physiologic effects that may improve the healing (Maida *et al.*, 2020).

With that in mind, studies are improving to determine the capacity of *Cannabis sativa* extract and/or THC and CBD in wound healing, including its mechanism of action.

Analgesia

In outpatients included in palliative care due to cancer, CBD administration showed to be a great form of increase life quality, due to pain reduction. Topical use of CBD showed to be the most common application form, and most patients used CBD daily (Highet *et al.*, 2020).

Further, a clinical trial conducted by Xu *et al.* (2020), counted with patients containing peripheral neuropathy secondary to diabetes mellitus, idiopathic peripheral neuropathy, and neuropathy secondary to medications (chemotherapy), one participant had nonpalpable pulse and two had capillary refill time greater than three seconds. From 29 patients, 23 presented no lower extremity edema, five showed a light edema and one had severe edema.

Neuropathic Pain Scale (NPS) score obtained across the weeks in control and treated groups was 3,93 with a medium baseline score of 3,76, with reported sensations of surface pain, deep pain, and unpleasant pain (Xu *et al.*, 2020).

Although no improvement was detected for deep pain, there were a significant reduction of NPS domains in CBD group, such as intense, sharp, itchy sensations, and unpleasant surface pain. This corroborates to the thought of CBD capacity in pain control, and importantly patients did not report adverse events (Xu *et al.*, 2020).

Dermatological conditions

Cannabis sativa was described as antioxidant after extract application or seed extract cream (Ali; Akhtar, 2015). Antioxidant effect may influence in dermatological conditions improvement, such as irritation, due to the ROS inhibition (Romanhole *et al.*, 2020; Trojahn *et al.*, 2015). According to Ali and Akhtar (2015), the use of cream base containing 3% *Cannabis sativa* seed extract, decreased erythema after 48 hours of application, in comparison with the skin without the extract cream or control cream.

Importantly, all volunteers presented non irritancies, ensuring its safety for human use. Further, sebum treatment with pure base and seed extract base formulations decreased in the period of the study, although, with a marked decay of sebum after 3% *Cannabis sativa* seed extract use. Authors attribute these fact to the constituent of the *Cannabis sativa* extracts, including fatty acids and phytosterols, which inhibits 5-alfa-reductase, responsible to convert testosterone into dihydrotestosterone, metabolites that stimulates the skin sebum secretion (Ali; Akhtar, 2015).

More recently, the authors determined if *Cannabis sativa* extract would improve skin surface due to the preview results. Then, study revealed that energy values were increased after dermocosmetic application for all volunteers, when compared to the base application, especially after 3 months. Although, the other parameter such as contrast and variance effects for dermocosmetic were insignificant for the base. In this case, the parameter determined, energy, contrast and variance should inform the improvement in skin texture (Ali *et al.*, 2020).

The lack of contrast and variance is characterized by the increase rise in skin hydration level, which creates a homogeny image. To support that, the surface evaluation of the living skin (SELS) results demonstrated that the base formulation did not produce significant effects, although, there were significant effects demonstrated by the statistical analysis for the results of dermocosmetic on SELS. In summary, the results pointed to a potential use of *Cannabis sativa extract* in the improvement of skin surface, and even, in skin aging (Ali *et al.*, 2020).

On the other hand, Maghfour *et al.* (2020) study pointed out the extract potential use in eczema administration. First, at baseline patients scores by the Patient Oriented Eczema Measure (POEM) demonstrated the severity of eczema, in which, 9 volunteers scored from 18 to 28 indicating severe to very severe eczema, 10 patients had a score from 8 to 16 indicating moderate eczema, and one volunteer scored 3 to 7 indicating mild eczema.

After treatment, questionary pointed out that from 16 volunteers, 3 presented reductions of their eczema from severe to mild range. The same result was observed with 3 volunteers that reported moderate eczema at the baseline, and at the end, reported resolution of their skin disease after using topical CBD for 2 weeks. Further, all the volunteers experienced improvement in the POEM score, significantly in the itch sensation, scaliness and dry skin. Furthermore, Quality of Life Hand Eczema Questionnaire (QOLHEQ) scores, resulted in a significant reduction in anxiety, sadness, and depression, and at the embarrassment about their conditions after the treatment. CBD efficacy was rated by the volunteers, demonstrating at least 60% improvement, and no volunteers reported adverse events (Maghfour *et al.*, 2020).

According to the results, is possible to discuss the significant positive results caused by CBD in the treatment of atopic dermatitis, due to suppression of cytokines involved in the inflammatory process (Maghfour *et al.*, 2020).

One more use for *Cannabis sativa* plant, reported in literature, was in skin burns relief. However, no scientifical background support the use of the plant in skin burns, since the use was reported by local inhabitants of North- West frontier province in Pakistan, probably using the plant as a traditional therapy. According to the interview, 2 out of the 328 interviewed reported the use of *Cannabis sativa L*. crushed fresh leaves in skin burns, together with Allium cepa fresh scales, applied directly (Abbasi *et al.*, 2010).

Recently, a randomized double-blinded placebo-controlled interventional pilot study determined the efficacy of CBD and aspartame formulation, as a novel topical treatment for atopic dermatitis. After 2 weeks of treatment, investigator's static global assessment reduced significantly, obtaining a score of 1.28 in the treated group compared to 0.70 in the placebo group. In addition, CBD only group did not obtain as much improvement as the CBD as aspartame group, and the investigator's static global assessment score were not significant comparing to the placebo group. Besides the limitation of this study, due to the small sample size, the result pointed to important results that aspartame may be an interesting combination to improve CBD treatment in atopic dermatitis (Gao *et al.*, 2022).

Pharmacokinetics

After a suspicious case reported by a driver, that was responsible for a car accident, claiming the use of *Cannabis sativa* based medicine to treated his bruises, Hess; Krämer and Madea (2017) conducted a study in order to verify the capacity of *Cannabis sativa* bioactives previously detected in the driver to absorb in the skin. In the driver, cannabinoids detected were 7.3 ng/mL of THC, 3.5 ng/mL of 11-Hydroxy-THC and 44.6 ng/mL of 11- nor-9-carboxy-THC (THC-COOH), which confirms the presence of cannabinoids in blood circulation.

During the experiment, volunteers using the same amount and the same formulation were tested for THC and its metabolites in blood circulation, and nothing was found. Possibly, the lack of identification was due to the concentration of the cannabinoids in the formulations administrated, since in analytical methods, the concentration of THC could be detected in both formulations tested 1,7 ng/mg of 11-Hydroxy-THC and 102 ng/mg of 11- nor-9-carboxy-THC (THC-COOH) (Hess; Krämer; Madea, 2017).

However, this study contraposes with the other studies previously discussed, in which skin absorption of *Cannabis sativa* bioactives has been detected, especially after authors determined improvement in health conditions studied. Although, the absorption may be dependent on the concentration of bioactive as much as formulation dependent.

3.4. Formulation influence in bioactives absorption

Δ^9-THC and CBD and its derivates are mostly apolar and considered hydrophobic/lipophobic molecules, as seen in the Figure 2. Besides molecule characteristics as polarity, administration pathway and barrier constitution are some factors that influence actives absorption. For example, mucosal pathways are mostly composed by water and proteins (Lane, 2013) while skin path is constituted by epidermis and dermis, that act as barrier for microorganisms and other substances entrance (Lane, 2013; Romanhole *et al.*, 2020).

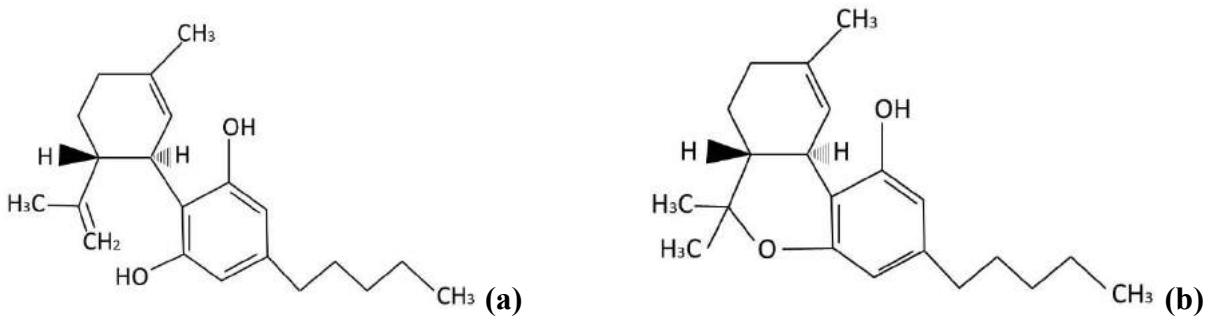

Figure 2: Molecule structure of cannabidiol- CBD (a) and (-)-delta 9-Tetrahydrocannabinol -Δ^9-THC (b).

As discussed along the review, the following studies reported in some point, the possibility of formulation interference in molecules absorption. In pre-clinical studies included using eye mucosal path, for example, different formulations were used, and mineral oil was the prevalent excipient to carry CBD and/or Δ^9-THC. When compared to sesame oil, mineral oil delivered the cannabinoids better, since permeation detection demonstrated higher concentration of cannabinoids (Green; Bowman; Wynn, 1978). In agreement, mineral oil with 1% of cannabinoids showed IOP reduction, after eye mucosal administration (Elsohly *et al.*, 1984).

When prepared as a water-soluble formulation, cannabinoids derivates also improved IOP condition, especially those derivates from Δ^9-THC. Importantly, inflammation was observed after administration of water-soluble formulation, symbolizing important effect of the molecules contact with the mucosal (Elsohly *et al.*, 1984). The authors affirmed that because of the absorption in both type of formulation, cannabinoids must be polar, although, considering the year of publication, that has been proven contrary with time and improvement of experimental tests.

Formulations may also contain enhancers that could improve actives permeation. For example, the use of Transcutol® HP (diethylene glycol monoethyl ether EP/USP-NF from Gattefosse (Cedex, France)) at 6% in gel formulation containing CBD improved the permeation significantly, given the plasma concentration found. On the other hand, CBD permeation in PEG-400, with DM-β- CD (Dimethyl-b-cyclodextrin) or 1% glycochocolate enhancers, did not proved to be higher. Possibly the application caused a cumulus of enhancers in the local applied, which may have modified the CBD thermodynamic and increased its saturation, reducing the permeation rate (Paudel *et al.*, 2010).Furthermore, the bioavailability of CBD decreased when PEG- 400 was adjusted from 100% to 50% (Paudel *et al.*, 2010).

Even more, delivery systems found in literature showed to be a great choice for cannabinoids delivery, for instance, ethosomes loaded with CBD, improved its delivery and permeation (Lodzki *et al.*, 2003).

In the same way, sterile microparticles loaded with CBD, in concentrations of 20-Mps and 10-Mps, released the bioactive faster when compared to non-sterile. In contrary, formulation with higher concentration (20-Mps) had slower erosion when compared to the lowest concentration (10-Mps). That may due to the fact that in the higher concentration of CBD improve its hydrophobic characteristic, which slows the water entrance in the microparticles (Fraguas-Sánchez *et al.*, 2020). In general, the increase of delivery after

bioactive encapsulation could be due to the better interaction of the particle with the barrier, leading to bioactive delivery.

In contrary, cannula implantation used by Colasanti; Powell and Craig (1984), demonstrated reduction in systemic absorption of Δ^9-THC, but increased its contact with the mucosal, leading to important adverse events. In this case, leads to questioning the effectiveness of the cannula use, which may force the permeation into the mucosal barrier.

Other formulation containing *Cannabis* based medicine, with content not specified, applied topically, were able to promote absorption into to the wound bed (VS-12) and to the peri-wound tissues (VS-14) (Maida *et al.*, 2020). However, in this last case, the skin barrier were not intact, thus, contributes to greater formulation absorption.

In summary, the information reported leads to the thought that the formulation plays an important role in the bioactive permeation, especially after enhancers use and encapsulations particles.

3.5. Adverse events

Along this review, adverse events were reported, and the main events are summarized in Table 9. In addition, Schmitz et al. (2020) complied information regarding the adverse events reported after PlusCBD™ use. The PlusCBD™ is a brand name given to full spectrum hemp extract of ingestible and topical products (Schmitz; Lopez; Marinotti, 2020). During the period of their search (2018-2019), an amount of 1,429 adverse events were detected in 1,151 cases. Most common effects were due to topical administration and oral administration. In fact, among oral administration adverse events, indigestion, as much as abdominal discomfort, overcame any other topical administration related, adverse events (Schmitz; Lopez; Marinotti, 2020).

Among the topical adverse events, the most reported are hypersensitivity, dermatitis, and rash. Other effects included burning sensation, pruritus, pain, urticaria, dermatitis contact and blister. Also, non-dermatologic effects after topical administration were not significant, but included headache, anxiety and abdominal discomfort. Further, 99.8% of the records reported were non-serious, and only 2 events were serious. Thus, authors could conclude that product showed to be safe and efficient when topically applied (Schmitz; Lopez; Marinotti, 2020).

The first serious case was in an 83-year-old man, that experienced rash and hallucinations, after 2 months of PlusCBD™, in a spray dose of 1.5mg of CBD/day. The results suggested that the product may not be the cause of the events since CBD is known to be non-psychoactive. The second case involved a 64-year-old woman reporting shortness of breath and

tongue swelling after four sublingual doses of PlusCBD™ oil peppermint liquid. In that case, the patient was treated for hypersensitivity reaction (Schmitz; Lopez; Marinotti, 2020).

Further, 60% out of 48 patients treating epileptic encephalopathies with CBD reported treatment- related adverse events. Among the adverse events 70% (32 patients) were considered mild and 26% (12 patients) moderate. 5% patients reported application site pain and somnolence, however, somnolence was reported by patients using clobazam in concomitance with CBD treatment. Adverse events caused by CBD was most related to gastrointestinal system but did not lead to interruption or changes in the CBD treatment. On the other hand, there were no clinically significant changes in vital signs or electrocardiograms (Scheffer *et al.*, 2021).

4. LIMITATIONS

The different methodological approach used by the studies reviewed, could interfere into the analysis, due to different experimental conditions. Thus, most of comparisons was made by qualitative analysis.

5. CONCLUSIONS

In conclusion, results pointed to potential use of cannabidiol in topical administration in all conditions studied such as: intraocular pressure, corneal hyperalgesia, osteoarthritis, colitis inflammation, multiple sclerosis, topical inflammation, relapse- promoting conditions and neurodegeneration. Antibacterial and antioxidant activities were proven as well. However, CBD alone showed lower activity, but safety results would provide a potential use in humans, differently alone brought important adverse events, especially in mucosal topical pathway. Thus, THC and CBD combination could be a great choice of treatment since the synergy among the bioactives improved pharmacological potential and did not present important adverse events. Although the methodological difference in the studies review in the present study, should be considered before moving to clinical trials. Finally, this review presented relevant information regarding the known application of CBD by topical administration, increasing perspectives for future studies. More importantly, brought out the fact that: even though is a potential and relatively famous bioactive, there's only 46 viable studies conducting experiments for topical administration. Thus, formulation details are relevant to new bioactive CBD products.

6. ACKOWLOGMENTS

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7. REFERENCES

Abbasi, A. M.; Khan, M. A.; Ahmad, M.; Zafar, M. *et al.* Ethnopharmacological application of medicinal plants to cure skin diseases and in folk cosmetics among the tribal communities of North-West Frontier Province, Pakistan. **Journal of Ethnopharmacology**, 128, n. 2, p. 322-335, 2010. Article.

Acosta, M. C.; Tan, M. E.; Belmonte, C.; Gallar, J. Sensations evoked by selective mechanical, chemical, and thermal stimulation of the conjunctiva and cornea. **Invest Ophthalmol Vis Sci**, 42, n. 9, p. 2063-2067, Aug 2001.

Ali, A.; Akhtar, N. The safety and efficacy of 3% Cannabis seeds extract cream for reduction of human cheek skin sebum and erythema content. **Pakistan Journal of Pharmaceutical Sciences**, 28, n. 4, p. 1389-1395, 2015. Article.

Ali, A.; Akhtar, N.; Khan, H.; Bin Asad, M. H. H. *et al.* The improvement on the skin surface by a new type of dermocosmetic loaded plant extract: A split face skin topographic study. **Pakistan Journal of Pharmaceutical Sciences**, 33, n. 2, p. 531-535, 2020. Article.

Ananthula, H.; Vaishya, R.; Barot, M.; Mitra, A. Duane's ophthalmology. **Bioavailability. Philadelphia: Lippincott Williams & Wilkins**, 2009.

Aparicio Rosana, R.; Polo Virginia, O. AN EPILEPTIC SEIZURE IN A PATIENT WITH MULTIPLE SCLEROSIS TREATED WITH THC/CBD (SATIVEX (TM)). **ATENCION FARMACEUTICA**, 15, n. 6, p. 439-442, 2013.

Ashtari, F.; Mokhtari, F.; Soudavi, M.; Saadat, H. *et al.* Effect of MS on pregnancy and the effect of pregnancy on MS patients in Isfahan. 2020.

Atalay, S.; Gęgotek, A.; Wroński, A.; Domigues, P. *et al.* Therapeutic application of cannabidiol on UVA and UVB irradiated rat skin. A proteomic study. **Journal of Pharmaceutical and Biomedical Analysis**, 192, 2021. Article.

Bardhan, A.; Bruckner-Tuderman, L.; Chapple, I. L. C.; Fine, J.-D. *et al.* Epidermolysis bullosa. **Nature Reviews Disease Primers**, 6, n. 1, p. 78, 2020/09/24 2020.

Bartner, L. R.; Mcgrath, S.; Rao, S.; Hyatt, L. K. *et al.* Pharmacokinetics of cannabidiol administered by 3 delivery methods at 2 different dosages to healthy dogs. **Canadian journal of veterinary research = Revue canadienne de recherche veterinaire**, 82, n. 3, p. 178-183, 2018.

Bitter, C.; Suter, K.; Surber, C. Nasal Drug Delivery in Humans. **Current problems in dermatology**, 40, p. 20-35, 02/01 2011.

Blaskovich, M. A. T.; Kavanagh, A. M.; Elliott, A. G.; Zhang, B. *et al.* The antimicrobial potential of cannabidiol. **Communications biology**, 4, n. 1, p. 7, 2021. Article.

Boivin, M. Nabiximols (Sativex®). *In*: **Cannabinoids and Pain**: Springer, 2021. p. 119-126.

Booth, J. K.; Bohlmann, J. Terpenes in Cannabis sativa – From plant genome to humans. **Plant Science**, 284, p. 67-72, 2019/07/01/ 2019.

Brandl, M.; Bauer-Brandl, A. Oromucosal drug delivery: Trends in in-vitro biopharmaceutical assessment of new chemical entities and formulations. **European Journal of Pharmaceutical Sciences**, 128, p. 112-117, 2019/02/01/ 2019.

Brioschi, F. A.; Di Cesare, F.; Gioeni, D.; Rabbogliatti, V. *et al.* Oral transmucosal cannabidiol oil formulation as part of a multimodal analgesic regimen: Effects on pain relief and quality of life improvement in dogs affected by spontaneous osteoarthritis. **Animals**, 10, n. 9, p. 1-14, 2020. Article.

Chelliah, M. P.; Zinn, Z.; Khuu, P.; Teng, J. M. C. Self-initiated use of topical cannabidiol oil for epidermolysis bullosa. **Pediatr Dermatol**, 35, n. 4, p. e224-e227, 2018/05 2018.

Clarke, R. C. **Marijuana botany: An advanced study: The propagation and breeding of distinctive cannabis**. Ronin publishing, 1981. 1579511090.

Colasanti, B. K. A comparison of the ocular and central effects of Δ9-tetrahydrocannabinol and cannabigerol. **Journal of Ocular Pharmacology**, 6, n. 4, p. 259-269, 1990. Article.

Colasanti, B. K.; Powell, S. R.; Craig, C. R. Intraocular pressure, ocular toxicity and neurotoxicity after administration of Δ9-Tetrahydrocannabinol or cannabichromene. **Experimental Eye Research**, 38, n. 1, p. 63-71, 1984. Article.

Conaghan, P. Update on osteoarthritis part 1: current concepts and the relation to exercise. **British journal of sports medicine**, 36, n. 5, p. 330-333, 2002.

Duchi, S.; Ovadia, H.; Touitou, E. Nasal administration of drugs as a new non-invasive strategy for efficient treatment of multiple sclerosis. **Journal of neuroimmunology**, 258, n. 1-2, p. 32-40, 2013.

Elbatsh, M. M.; Moklas, M. A. A.; Marsden, C. A.; Kendall, D. A. Antidepressant-like effects of Δ9-tetrahydrocannabinol and rimonabant in the olfactory bulbectomised rat model of

depression. **Pharmacology Biochemistry and Behavior**, 102, n. 2, p. 357-365, 2012/08/01/ 2012.

Elsohly, M. A.; Harland, E. C.; Benigni, D. A.; Waller, C. W. Cannabinoids in glaucoma II: the effect of different cannabinoids on intraocular pressure of the rabbit. **Curr Eye Res**, 3, n. 6, p. 841-850, 1984/06 1984.

Farag, S.; Kayser, O. The cannabis plant: botanical aspects. *In*: **Handbook of Cannabis and Related Pathologies**: Elsevier, 2017. p. 3-12.

Feng, Y.; Simpson, T. L. Nociceptive sensation and sensitivity evoked from human cornea and conjunctiva stimulated by CO2. **Invest Ophthalmol Vis Sci**, 44, n. 2, p. 529-532, Feb 2003.

Fiani, B.; Sarhadi, K. J.; Soula, M.; Zafar, A. *et al.* Current application of cannabidiol (CBD) in the management and treatment of neurological disorders. **Neurological Sciences**, 41, n. 11, p. 3085-3098, 2020/11/01 2020.

Fischedick, J. T. Identification of terpenoid chemotypes among high (−)-trans-Δ9-tetrahydrocannabinol-producing Cannabis sativa L. cultivars. **Cannabis and cannabinoid research**, 2, n. 1, p. 34-47, 2017.

Fraguas-Sánchez, A. I.; Fernández-Carballido, A.; Delie, F.; Cohen, M. *et al.* Enhancing ovarian cancer conventional chemotherapy through the combination with cannabidiol loaded microparticles. **Eur J Pharm Biopharm**, 154, p. 246-258, 2020/07 2020.

Frasch, H. F.; Barbero, A. M. Application of numerical methods for diffusion-based modeling of skin permeation. **Advanced Drug Delivery Reviews**, 65, n. 2, p. 208-220, 2013/02/01/ 2013.

Frohman, E. M.; Racke, M. K.; Raine, C. S. Multiple sclerosis—the plaque and its pathogenesis. **New England Journal of Medicine**, 354, n. 9, p. 942-955, 2006.

Gaffal, E.; Cron, M.; Glodde, N.; Tüting, T. Anti-inflammatory activity of topical THC in DNFB-mediated mouse allergic contact dermatitis independent of CB1 and CB2 receptors. **Allergy**, 68, n. 8, p. 994-1000, 2013/07 2013.

Gao, Y.; Li, Y.; Tan, Y.; Liu, W. *et al.* Novel cannabidiol aspartame combination treatment (JW-100) significantly reduces ISGA score in atopic dermatitis: Results from a randomized double-blinded placebo-controlled interventional study. **Journal of Cosmetic Dermatology**, 21, n. 4, p. 1647-1650, 2022.

Gaudana, R.; Ananthula, H. K.; Parenky, A.; Mitra, A. K. Ocular Drug Delivery. **The AAPS Journal**, 12, n. 3, p. 348-360, 2010/09/01 2010.

Giacoppo, S.; Galuppo, M.; Pollastro, F.; Grassi, G. *et al.* A new formulation of cannabidiol in cream shows therapeutic effects in a mouse model of experimental autoimmune encephalomyelitis. **Daru**, 23, p. 48-48, 2015/10 2015.

Glyn-Jones, S.; Palmer, A. J. R.; Agricola, R.; Price, A. J. *et al.* Osteoarthritis. **The Lancet**, 386, n. 9991, p. 376-387, 2015/07/25/ 2015.

Gonzalez-Cuevas, G.; Martin-Fardon, R.; Kerr, T. M.; Stouffer, D. G. *et al.* Unique treatment potential of cannabidiol for the prevention of relapse to drug use: preclinical proof of principle. **Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology**, 43, n. 10, p. 2036-2045, 2018.

Goyal, S.; Hamrah, P. Understanding Neuropathic Corneal Pain--Gaps and Current Therapeutic Approaches. **Seminars in ophthalmology**, 31, n. 1-2, p. 59-70, 2016.

Green, K.; Bowman, K.; Wynn, H. Intraocular pressure-reducing effects of naturally occurring cannabinoids. **Investigative Ophthalmology and Visual Science**, 17, n. Suppl., p. 162, 1978.

Green, K.; Roth, M. Ocular Effects of Topical Administration of Δ9-Tetrahydrocannabinol in Man. **Archives of Ophthalmology**, 100, n. 2, p. 265-267, 1982. Article.

Green, K.; Zalkow, L. H.; Deutsch, H. M. Ocular and systemic responses to water soluble material derived from Cannabis sativa (marihuana). **Current Eye Research**, 1, n. 2, p. 65-75, 1981. Article.

Hammell, D. C.; Zhang, L. P.; Ma, F.; Abshire, S. M. *et al.* Transdermal cannabidiol reduces inflammation and pain-related behaviours in a rat model of arthritis. **European journal of pain (London, England)**, 20, n. 6, p. 936-948, 2016.

Hannon, M. B.; Deabold, K. A.; Talsma, B. N.; Lyubimov, A. *et al.* Serum cannabidiol, tetrahydrocannabinol (THC), and their native acid derivatives after transdermal application of a low-THC Cannabis sativa extract in beagles. **Journal of veterinary pharmacology and therapeutics**, 43, n. 5, p. 508-511, 2020.

Hanuš, L. O.; Meyer, S. M.; Muñoz, E.; Taglialatela-Scafati, O. *et al.* Phytocannabinoids: a unified critical inventory. **Nat Prod Rep**, 33, n. 12, p. 1357-1392, Nov 23 2016.

Heineman, J. T.; Forster, G. L.; Stephens, K. L.; Cottler, P. S. *et al.* A Randomized Controlled Trial of Topical Cannabidiol for the Treatment of Thumb Basal Joint Arthritis. **The Journal of Hand Surgery**, 2022.

Hess, C.; Krämer, M.; Madea, B. Topical application of THC containing products is not able to cause positive cannabinoid finding in blood or urine. **Forensic Science International**, 272, p. 68-71, 2017. journal article.

Heussler, H.; Cohen, J.; Silove, N.; Tich, N. *et al.* A phase 1/2, open-label assessment of the safety, tolerability, and efficacy of transdermal cannabidiol (ZYN002) for the treatment of pediatric fragile X syndrome. **Journal of neurodevelopmental disorders**, 11, n. 1, p. 16, 2019.

Highet, B. H.; Lesser, E. R.; Johnson, P. W.; Kaur, J. S. Tetrahydrocannabinol and Cannabidiol Use in an Outpatient Palliative Medicine Population. **The American journal of hospice & palliative care**, 37, n. 8, p. 589-593, 2020. Article.

Hiligsmann, M.; Cooper, C.; Arden, N.; Boers, M. *et al.* Health economics in the field of osteoarthritis: an expert's consensus paper from the European Society for Clinical and Economic Aspects of Osteoporosis and Osteoarthritis (ESCEO). **Semin Arthritis Rheum**, 43, n. 3, p. 303-313, Dec 2013.

Hoffmann, W. **Hanf, Cannabis sativa L.** . Handbuch der Pflanzenzüchtung. Berlin: Paul Parey: 1961.

Institute, J. B. Joanna Briggs Institute reviewers' manual 2014. **Adelaide: The Joanna Briggs Institute**, 2014.

Jannin, V.; Lemagnen, G.; Gueroult, P.; Larrouture, D. *et al.* Rectal route in the 21st Century to treat children. **Adv Drug Deliv Rev**, 73, p. 34-49, Jun 2014.

Karussis, D. The diagnosis of multiple sclerosis and the various related demyelinating syndromes: a critical review. **Journal of autoimmunity**, 48, p. 134-142, 2014.

Kiefer, D. Topical Cannabis for Wound Pain: A Case Series...Maida V, Corban J. Topical medical cannabis: A new treatment for wound pain — three cases of pyoderma gangrenosum. J Pain Symptom Manage. 2017;54:732-736. **Integrative Medicine Alert**, 20, n. 12, p. 14-15, 2017.

Kumar, P.; Mahato, D. K.; Kamle, M.; Borah, R. *et al.* Pharmacological properties, therapeutic potential, and legal status of Cannabis sativa L.: An overview. **Phytotherapy Research**, n/a, n. n/a, 2021.

Lane, M. E. Skin penetration enhancers. **International journal of pharmaceutics**, 447, n. 1-2, p. 12-21, 2013.

Langford, R. M.; Mares, J.; Novotna, A.; Vachova, M. *et al.* A double-blind, randomized, placebo-controlled, parallel-group study of THC/CBD oromucosal spray in combination with the existing treatment regimen, in the relief of central neuropathic pain in patients with multiple sclerosis. **Journal of neurology**, 260, n. 4, p. 984-997, 2013. Journal Article; Multicenter Study; Randomized Controlled Trial; Research Support, Non-U.S. Gov't.

Liput, D. J.; Hammell, D. C.; Stinchcomb, A. L.; Nixon, K. Transdermal delivery of cannabidiol attenuates binge alcohol-induced neurodegeneration in a rodent model of an alcohol use disorder. **Pharmacology, biochemistry, and behavior**, 111, p. 120-127, 2013.

Liu, S.; Yang, S.; Ho, P. C. Intranasal administration of carbamazepine-loaded carboxymethyl chitosan nanoparticles for drug delivery to the brain. **Asian Journal of Pharmaceutical Sciences**, 13, n. 1, p. 72-81, 2018/01/01/ 2018.

Lodzki, M.; Godin, B.; Rakou, L.; Mechoulam, R. *et al.* Cannabidiol-transdermal delivery and anti-inflammatory effect in a murine model. **Journal of controlled release : official journal of the Controlled Release Society**, 93, n. 3, p. 377-387, 2003.

Łuczaj, W.; Jastrząb, A.; Do Rosário Domingues, M.; Domingues, P. *et al.* Changes in Phospholipid/Ceramide Profiles and Eicosanoid Levels in the Plasma of Rats Irradiated with UV Rays and Treated Topically with Cannabidiol. **International journal of molecular sciences**, 22, n. 16, p. 8700, 2021.

Machiele, R.; Motlagh, M.; Patel, B. Intraocular Pressure. [Updated 2021 Jul 31]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing 2021.

Maghfour, J.; Rietcheck, H. R.; Rundle, C. W.; Runion, T. M. *et al.* An Observational Study of the Application of a Topical Cannabinoid Gel on Sensitive Dry Skin. **Journal of drugs in dermatology : JDD**, 19, n. 12, p. 1204-1208, 2020. Article.

Maida, V.; Corban, J. Topical Medical Cannabis: A New Treatment for Wound Pain—Three Cases of Pyoderma Gangrenosum. **Journal of Pain and Symptom Management**, 54, n. 5, p. 732-736, 2017. Article.

Maida, V.; Shi, R. B.; Fazzari, F. G. T.; Zomparelli, L. Topical cannabis-based medicines – A novel paradigm and treatment for non-uremic calciphylaxis leg ulcers: An open label trial. **International Wound Journal**, 17, n. 5, p. 1508-1516, 2020.

Maniscalco, G. T.; Aponte, R.; Bruzzese, D.; Guarcello, G. *et al.* THC/CBD oromucosal spray in patients with multiple sclerosis overactive bladder: a pilot prospective study. **Neurological Sciences**, 39, n. 1, p. 97-102, 2018.

Maverakis, E.; Marzano, A. V.; Le, S. T.; Callen, J. P. *et al.* Pyoderma gangrenosum. **Nature Reviews Disease Primers**, 6, n. 1, p. 81, 2020/10/08 2020.

Mayeux, R. EPIDEMIOLOGY OF NEURODEGENERATION. **Annual Review of Neuroscience**, 26, n. 1, p. 81-104, 2003.

Mciver, V. C.; Tsang, A. S.; Symonds, N. E.; Perkins, N. R. *et al.* Effects of topical treatment of cannabidiol extract in a unique manuka factor 5 manuka honey carrier on second intention

wound healing on equine distal limb wounds: a preliminary study. **Australian Veterinary Journal**, 98, n. 6, p. 250-255, 2020. Article.

Miller, S.; Daily, L.; Leishman, E.; Bradshaw, H. *et al.* Δ9-tetrahydrocannabinol and cannabidiol differentially regulate intraocular pressure. **Investigative Ophthalmology and Visual Science**, 59, n. 15, p. 5904-5911, 2018. Article.

Miller, S.; Kulkarni, S.; Ciesielski, A.; Nikas, S. P. *et al.* Controlled-deactivation CB1 receptor ligands as a novel strategy to lower intraocular pressure. **Pharmaceuticals**, 11, n. 2, 2018. Article.

Miller, S.; Leishman, E.; Oehler, O.; Daily, L. *et al.* Evidence for a GPR18 role in diurnal regulation of intraocular pressure. **Investigative ophthalmology & visual science**, 57, n. 14, p. 6419-6426, 2016.

Nurmikko, T. J.; Serpell, M. G.; Hoggart, B.; Toomey, P. J. *et al.* Sativex successfully treats neuropathic pain characterised by allodynia: a randomised, double-blind, placebo-controlled clinical trial. **Pain**, 133, n. 1-3, p. 210-220, 2007. Clinical Trial; Journal Article; Multicenter Study; Randomized Controlled Trial.

Pan, Q.; Yu, Y.; Chen, D.; Jiao, G. *et al.* Enhanced penetration strategies for transdermal delivery. **Frontiers of Chemical Science and Engineering**, 14, n. 3, p. 378-388, 2020.

Paudel, K. S.; Hammell, D. C.; Agu, R. U.; Valiveti, S. *et al.* Cannabidiol bioavailability after nasal and transdermal application: effect of permeation enhancers. **Drug development and industrial pharmacy**, 36, n. 9, p. 1088-1097, 2010.

Prausnitz, M. R.; Langer, R. Transdermal drug delivery. **Nature biotechnology**, 26, n. 11, p. 1261-1268, 2008.

Purohit, T. J.; Hanning, S. M.; Wu, Z. Advances in rectal drug delivery systems. **Pharmaceutical Development and Technology**, 23, n. 10, p. 942-952, 2018/11/26 2018.

Radwan, M. M.; Chandra, S.; Gul, S.; Elsohly, M. A. Cannabinoids, Phenolics, Terpenes and Alkaloids of Cannabis. **Molecules**, 26, n. 9, May 8 2021.

Rebibo, L.; Frušić-Zlotkin, M.; Ofri, R.; Nassar, T. *et al.* The dose-dependent effect of a stabilized cannabidiol nanoemulsion on ocular surface inflammation and intraocular pressure. **International Journal of Pharmaceutics**, 617, p. 121627, 2022.

Riva, N.; Mora, G.; Sorarù, G.; Lunetta, C. *et al.* Safety and efficacy of nabiximols on spasticity symptoms in patients with motor neuron disease (CANALS): a multicentre, double-blind, randomised, placebo-controlled, phase 2 trial. **The Lancet Neurology**, 18, n. 2, p. 155-164, 2019. Article.

Robinson, A. P.; Harp, C. T.; Noronha, A.; Miller, S. D. The experimental autoimmune encephalomyelitis (EAE) model of MS: utility for understanding disease pathophysiology and treatment. **Handb Clin Neurol**, 122, p. 173-189, 2014.

Rog, D. J.; Nurmikko, T. J.; Friede, T.; Young, C. A. Randomized, controlled trial of cannabis-based medicine in central pain in multiple sclerosis. **Neurology**, 65, n. 6, p. 812-819, 2005. Article.

Rog, D. J.; Nurmikko, T. J.; Young, C. A. Oromucosal Δ9-tetrahydrocannabinol/cannabidiol for neuropathic pain associated with multiple sclerosis: An uncontrolled, open-label, 2-year extension trial. **Clinical Therapeutics**, 29, n. 9, p. 2068-2079, 2007. Article.

Romanhole, R. C.; Fava, A. L. M.; Tundisi, L. L.; De Macedo, L. M. *et al.* Unplanned absorption of sunscreen ingredients: Impact of formulation and evaluation methods. **International Journal of Pharmaceutics**, p. 120013, 2020.

Rosenthal, P.; Baran, I.; Jacobs, D. S. Corneal pain without stain: is it real? **Ocul Surf**, 7, n. 1, p. 28-40, Jan 2009.

Scheffer, I. E.; Hulihan, J.; Messenheimer, J.; Ali, S. *et al.* Safety and Tolerability of Transdermal Cannabidiol Gel in Children with Developmental and Epileptic Encephalopathies: A Nonrandomized Controlled Trial. **JAMA network open**, 4, n. 9, p. e2123930-e2123930, 2021.

Schicho, R.; Storr, M. Topical and systemic cannabidiol improves trinitrobenzene sulfonic acid colitis in mice. **Pharmacology**, 89, n. 3-4, p. 149-155, 2012/03 2012.

Schmitz, S. M.; Lopez, H. L.; Marinotti, O. Post Marketing Safety of Plus CBD™ Products, a Full Spectrum Hemp Extract: A 2-Year Experience. **Journal of dietary supplements**, 17, n. 5, p. 587-598, 2020. Article.

Schräder, N. Cannabinoid-based medicines for pain and pruritus in EB. **Acta Dermato-Venereologica**, 100, n. SUPPL 220, p. 19-20, 2020. Conference Abstract.

Schultes, R. E.; Klein, W. M.; Plowman, T.; Lockwood, T. E. Cannabis: an example of taxonomic neglect. *In*: **Cannabis and culture**: De Gruyter Mouton, 1974. p. 21-38.

Steinberg, J.; Webber, C. The roles of FMRP-regulated genes in autism

spectrum disorder: single- and multiple-hit genetic etiologies. . **Am J Hum Genet.**, 93, p. 825–839, 2013.

Steinman, L. Multiple sclerosis: a two-stage disease. **Nature immunology**, 2, n. 9, p. 762-764, 2001.

Straiker, A. What is currently known about cannabidiol and ocular pressure. **Expert Review of Ophthalmology**, 14, n. 6, p. 259-261, 2019.

Teixeira, F. V.; Hosne, R. S.; Sobrado, C. W. Management of ulcerative colitis: a clinical update. **Journal of Coloproctology (Rio de Janeiro) [online]** 35, n. n. 04, p. pp. 230-237, 2015.

Thapa, D.; Cairns, E. A.; Szczesniak, A. M.; Toguri, J. T. *et al.* The Cannabinoids Δ8THC, CBD, and HU-308 Act via Distinct Receptors to Reduce Corneal Pain and Inflammation. **Cannabis and Cannabinoid Research**, 3, n. 1, p. 11-20, 2018. Article.

Thrimawithana, T. R.; Young, S.; Bunt, C. R.; Green, C. *et al.* Drug delivery to the posterior segment of the eye. **Drug Discovery Today**, 16, n. 5, p. 270-277, 2011/03/01/ 2011.

Trojahn, C.; Dobos, G.; Lichterfeld, A.; Blume-Peytavi, U. *et al.* Characterizing facial skin ageing in humans: disentangling extrinsic from intrinsic biological phenomena. **BioMed research international**, 2015, 2015.

Tubaro, A.; Giangaspero, A.; Sosa, S.; Negri, R. *et al.* Comparative topical anti-inflammatory activity of cannabinoids and cannabivarins. **Fitoterapia**, 81, n. 7, p. 816-819, 2010/05 2010.

Turner, C.; Elsohly, M., 1976, **ANHYDROCANNABISATIVINE, A NEW ALKALOID ISOLATED FROM CANNABIS-SATIVA**. LLOYDIA 917 PLUM STREET, CINCINNATI, OH 45202. 474-474.

Turner, C. E.; Elsohly, M. A.; Boeren, E. G. Constituents of Cannabis sativa L. XVII. A review of the natural constituents. **Journal of Natural Products**, 43, n. 2, p. 169-234, 1980.

Van Cauwenberge, P.; Sys, L.; De Belder, T.; Watelet, J.-B. Anatomy and physiology of the nose and the paranasal sinuses. **Immunology and Allergy Clinics**, 24, n. 1, p. 1-17, 2004.

Van Hoogdalem, E. J.; De Boer, A. G.; Breimer, D. D. Pharmacokinetics of rectal drug administration, Part II. Clinical applications of peripherally acting drugs, and conclusions. **Clin Pharmacokinet**, 21, n. 2, p. 110-128, Aug 1991.

Vanhoenacker, G.; Van Rompaey, P.; De Keukeleire, D.; Sandra, P. Chemotaxonomic features associated with flavonoids of cannabinoid-free cannabis (Cannabis sativa subsp. sativa L.) in relation to hops (Humulus lupulus L.). **Natural product letters**, 2010.

Vicente-Valor, M.; Garcia-Llopis, P.; Mejia Andujar, L.; Antonino De La Camara, G. *et al.* Cannabis derivatives therapy for a seronegative stiff-person syndrome: a case report. **Journal of clinical pharmacy and therapeutics**, 38, n. 1, p. 71-73, 2013.

Vranka, J. A.; Kelley, M. J.; Acott, T. S.; Keller, K. E. Extracellular matrix in the trabecular meshwork: intraocular pressure regulation and dysregulation in glaucoma. **Exp Eye Res**, 133, p. 112-125, Apr 2015.

Wang, E. A.; Maverakis, E. M. The rapidly evolving lesions of ulcerative pyoderma gangrenosum: a timeline. **International journal of dermatology**, 2018.

Xu, D. H.; Cullen, B. D.; Tang, M.; Fang, Y. The Effectiveness of Topical Cannabidiol Oil in Symptomatic Relief of Peripheral Neuropathy of the Lower Extremities. **Current pharmaceutical biotechnology**, 21, n. 5, p. 390-402, 2020.

8. TABLES

Table 1: Methodological details of pre-clinical included studies concerning the capability of cannabinoids in IOP reduction (8) and corneal hyperalgesia (1).

Animals	Total N	Drug	Formulation	Exposure/Duration	Before treatment	Treatment	Drug Response	Authors
Male and Female Albino Rabbits	18	Δ^9 and Δ^8 -THC and its metabolites, cannabidiol, cannabinol and cannabinol acetate	Sesame oil or mineral oile	7 hours	Ganglionectomy surgery	**Control eye:** no treatment **Treated eye:** 1 drop containing the drug, and after 4 hours, a second drop was administrated	The drugs tested showed interesting results, leading to the need of clinical experimentation.	Green; Bowman and Wynn (1978)
Albino rabbits (gender not specified)	N/A	Δ^9 and Δ^8 -THC and its metabolites	N/A	N/A	N/A	**Control groups:** vehicle administration into intravenous and topical paths **Treated IV group:** cannabinoids derivates solution **Treated Topical group:** cannabinoids derivates in mineral oil **Treated Topical group:** cannabinoids derivates in water solution	Despite the IV rote showing better IOP reduction, the topical administration of 9α,10α-epoxy-HHC and 90-OH-HHC were also effective. Further, the water soluble derivates of Δ^9-THC and Δ^8-THC reduce IOP but may cause irritant activity.	Elsohly *et al.* (1984)
Male mice C57BL/6J (C57) strain except CB1 -/-	N/A	(-)-Δ 8 -THC-DMH with controlled deactivation	N/A	5hours	N/A	**Control eye:** received 5uL of Tricosolve **Treated eye:** received 5uL of the drug	The IOP reduction by cannabinoids is probably by stimulation of CB1 receptor as direct targeting with controlled-deactivation ligands.	Miller; Kulkarni*; et al.* (2018)
Male and Female mice C57BL/6J (C57) strain except CB1 -/-	N/A	THC and CBD	Tocrisolve*	8 hours	IOP measurement	**Control eye:** vehicle **Treated eye:** formulation	More CBD studies must be conducted, and THC showed to be a great strategy for IOP treatment.	Miller; Daily*; et al.* (2018)
Adult cats (gender not specified)	50	Δ^9-THC and cannabichromene	Mineral oil.	9 days	N/A	**Control group:** vehicle (PEG-400) **Δ^9-THC treated group:** Δ^9-THC through osmotic minipumps at 1uL per hour	From the cannabinoids studied, Δ^9-THC was the most effective in IOP reduction but with damaging side effects.	Colasanti; Powell and Craig (1984)

Adult cats (gender not specified)	N/A	Δ^9-THC and cannabigerol	Mineral oil and PEG-400, respectively	9 days	Ganglionectomy surgery (also removed small portions of the postganglionic fibers)	**Cannabichromene treated group:** cannabichromene through osmotic minipumps at 1uL per hour **Control group:** vehicle (PEG-400) **Δ^9-THC treated group:** Δ^9-THC through osmotic minipumps at 1uL per hour **Cannabigerol treated group:** cannabichromene through osmotic minipumps at 1uL per hour	The use of cannabigerol may be a promising drug for glaucoma treatment, considering its activity and also the potential increase of fluid drainage from the eye.	Colasanti (1990)
Albino, Pigmented Rabbits and Rhesus monkeys	N/A	Marijuana derived material (MDM)	N/A	24 hours	N/A	**Intravenous**: antagonist administration, phenoxybenzamine, phentolamine, sotalol, propranolol, timolol or indomethacin **Topical administration:** 50uL of MDM, from 1 to 10% concentration, unilaterally in the presence and absence of antimicrobial chemicals (benzalkonium chlorides or cetylpyridinium chloride)	MDM administrated topically did not showed effectiveness in the IOP changes when compared to the intravenous path.	Green; Zalkow and Deutsch (1981)
Male BALB/c and CB_2R knockout mice	N/A	Δ^8 - THC, CBD or HU-308	Soybean oil (0,2-5% w/v)	6 and 12 hours	Corneal injury was induced by cauterization with silver nitrate	**Treatment:** CBD or Δ^8 - THC or HU-308 solution was applied topically to cauterized corneas at 30min, 60min and 120 minutes post cauterization	There is evidence that the use o D8THC and CBD and other cannabinoid derivates (HU-308) reduce corneal hyperalgesia and neutrophil infiltration caused by corneal injury.	Thapa *et al.* (2018)
Female C57BL/6 mice	N/A	CBD	Nanoemulsions**	2 weeks	IOP measurement	**Control eye:** blank nanoemulsion **Treated eye**: nanoemulsion with CBD	CBD nanoemulsions decreased IOP after in a concentration of 0.4% and 1.6% after a few days of treatment.	Rebibo *et al.* (2022)

*Tocrisolve: water soluble emulsion, composed of 1:4 ratio of soya oil/water, emulsified with a block co-polymer Pluronic F68.
**Tween 80, castor oil, MCT, BHT, propyl gallate, solutol HS15 or glycerin
N/A: Not available

Table 2: Methodological details of pre-clinical included studies concerning the cannabinoids delivery studies in mucosal: oromucosal (1), intranasal (2) and rectal (1).

Animals	Total N	Drug	Formulation	Exposure/ Duration	Treatment	Drug Response	Author
Dogs, with different breed, age, body weight and gender, with spontaneous osteoarthritis	21	CBD	Oil (not specified)	12 weeks	(1) **Control:** without CBD treatment (2) **Treatment:** CBD oil (at 2mg/kg) every 12 hours for 12 weeks (period of blood collection) All the dogs, in both groups, received anti-inflammatory, gabapentin and amitriptyline by oral path as complementary therapy	In conclusion, CBD used with other anti-inflammatory drugs, may enhance quality of life, and reduce chronic pain in OA condition.	Brioschi *et al.* (2020)
Female mice (C57B1/6) with 6-7 weeks of age. Induced to present autoimmune encephalomyelitis (EAE)	~ 117	CBD	Nasal delivery system*	~ 28 days	(1) **GA (6,7mg/mL) Nasally acute (nasal delivery system):** 9 mices (5 immunized and 4 EAE) (2) **GA (6,7mg/mL) Subcutaneous injection (solution):** 10 mices (5 immunized and 5 EAE) (3) **GA (13,7mg/mL) Nasally acute (nasal delivery system):** 12 mices (6 immunized and 6 EAE) (4) **GA (13,7mg/mL) Subcutaneous injection (solution):** 12 mices (6 immunized and 6 EAE) (5) **GA (13,7mg/mL) Nasally acute (solution):** 13 mices (7 immunized and 6 EAE) (6) **CBD (6,7mg/mL) Nasally acute (nasal delivery system):** 10 mices (5 immunized and 5 EAE) (7) **CBD (6,7mg/mL) Subcutaneous injection (solution):** 9 mices (5 immunized and 4 EAE) (8) **CBD + GA (6,7+6,7mg/mL) Nasally acute (nasal delivery system):** 10 mices (5 immunized and 5 EAE) (9) **CBD + GA (6,7+6,7mg/mL) Subcutaneous injection (solution):** 9 mices (5 immunized and 4 EAE) (10) **Prednisolone (1,5 mg/mL) Nasally preventive (nasal delivery system):** 11 mices (6 immunized and 5 EAE) (11) **Prednisolone (1,5 mg/mL) Subcutaneous preventive (solution):** 12 mices (6 immunized and 6 EAE) (12), (13), (14) and (15) Control groups: no treatment	Besides, the combination of the CBD with GA in general, showed great efficiency suppression of the clinical effect of the EAE when compared to the drugs alone.	Duchi; Ovadia and Touitou (2013)
Male CD1 mices (5-9 weeks old). Colitis induction in mice	N/A	CBD	Canola oil	N/A	(1) **Control group:** received 100uL of 30% ethanol (2) **Control groups:** received only vehicles from the formulations of CBD intrarectal, intraperitoneal and intragastric	The use of CBD in treatment for colon inflammation is a great option and it should be further studied.	Schicho and Storr (2012)

with Sulfonic Acid (TNBS)				(3) **TNBS + CBD Intrarectal:** received TNBS 4mg in 100uL of 30% ethanol + CBD intrarectal with catheter daily (4) **TNBS + CBD Intraperitoneal:** received TNBS 4mg in 100uL of 30% ethanol + CBD intraperitoneal (10mg/Kg) (5) **TNBS + CBD Intragastric:** received TNBS 4mg in 100uL of 30% ethanol + CBD intragastric by gavage daily		
Male and female hairless rats IAF	12	CBD	PEG 400; 50:35:15 PEG:saline:ethanol solution (ES) with or without enhancers .(1% sodium glycocholate or 1% DM-β-CD)	4hours	(1) BD + PEG 400 (2) CBD + PEG 400 + ES (3) CBD + PEG 400 + ES + 1% glycochocolate (4) CBD + PEG 400 + ES + 1% DM--β-CD	Intranasal application of CBD showed rapid and significant absorption of CBD from the nasal cavity. The plasma concentration of CBD was significant after topical gel application in vivo. Further, there were differences in absorption in formulation with and without enhancers.	Paudel *et al.* (2010)

*Nasal system delivery: pressurized inhaler, using soy phospholipid with 94% phosphatidylcholine, lysophosphatidylcholine and Vitamin E.
N/A= Not available

Table 3: Methodological details of pre-clinical included studies concerning the cannabinoids transdermal delivery studies (7).

Animals	Total N	Drugs	Formulation	Exposure/ Duration	Treatment	Drug Response	Reference
Part 1: Male CD1 nude mice (8-9 weeks old) **Part 2 and 3:** Male ICR mice (20g)	**Part 1:** 6 mice **Part 2 and 3**: 30 mice	CBD ethosomes	Patches containing (with 3% w/w CBD and 40% w/w EtOH in carbomer gel)	**Part 1:** 24 hours **Part 2:** 73 hours **Part 3:** 19 hours	**Part 1:** patches containing CBD ethosomes **Part 2:** 200mg of CBD ethosomes covered with Hill-Top patch and fixed at certain time points (2; 4,25; 9; 13; 25; 37,5; 49,5 and 73 hours) **Part 3:** 100mg of CBD ethosomes covered with Hill-Top patch applied 19 hours before the carrageenan injection (treated group) and no treatment prior inflammation induction (control group)	CBD in ethosomes by transdermal patch, was delivered systemically to the inflamed organ and promote an anti-inflammatory effect.	Lodzki *et al.* (2003)
Male Sprague Dawley rats, with induced monoarthritis, by Freud's adjuvant (CFA) injection	54	CBD	1% and 10% CBD gel	4 days	(1) **Control (CFA +VEH):** vehicle gel administration on day 3 post-arthritis induction (2) **CFA + CBD:** 1% CBD gel: 0,62mg by 75 µL (3,5cm² of back skin), 3,1mg by 375 µL (17,5 cm² of back skin), 6,2mg by 750 µL (35 cm² of back skin 10% CBD gel: 62 mg by 750 µL	Topical CBD application showed great therapeutic potential for relief of pain in arthritis and inflammation without evident side-effects.	Hammell *et al.* (2016)
Six poupose-bred female research beagles	6 dogs	32 mg/ml CBD, 33 mg/ml CBDA, 1.3 mg/ml THC, and 1.0 mg/ml THCA	Ethanol extract with Pencream (HUMco)*	2 weeks	The dogs received the ointment 2x a day, every 12 hours	Transdermal application is a potential path to CBD administration and the superior absorption of CBDA suggests that acidic forms of cannabinoids may be a better focus for anti-inflammatory effect.	Hannon *et al.* (2020)
Male beagle dogs, healthy conditions and sexually intact	30	CBD	Transdermal cream	6 weeks	**CBD- infused transdermal cream (110mg/mL):** subgroup (a) with 5 dogs 75mg q12h and subgroup (b) with 5 dogs 150mg q12h **Oral microencapsulated oil beads (25mg- 50mg):** subgroup (a) with 5 dogs 75mg q12h and subgroup (b) with 5 dogs 150mg q12h **Oral CBD- infused oil (75mg/mL or 150mg/mL):** subgroup (a) with 5 dogs 75mg q12h and subgroup (b) with 5 dogs 150mg q12h	N/A	Bartner *et al.* (2018)

Male Wister rats, addiction induced with Ethyl Alcohol (10% w/v) orally	~ 24	2,5% CBD	Hydroalcoholic gel formulation with non-toxic skin biocompatible permeation enhancer.	138 days	**Control:** CBD vehicle each 24 hours for 7 days **Treatment:** CBD transdermal gel (15mg/Kg approximately) each 24 hours for 7 days	The authors provided information regarding CBD therapeutic profile for relapse prevention for substances use disorders.	Gonzalez-Cuevas *et al.* (2018)
Adult male Sprague Dawley rats with alcohol induced neurodegeneration with Ethanol	~ 86	CBD	1%, 2,5% and 5% CBD transdermal gels	4 days	**(1) (2) (3) Neurodegeneration induction (Ethanol) + CBD gel:** at 1%, 2,5% and 5% **(4)** Vehicle gel **(5)** Ethanol only **(6)** Ethanol + vehicle gel CBD gels and vehicles were first applied after the third dose of ethanol by gel application, daily at 11am and CBD solution were administrated by intraperitoneal injection in a concentration of 20mg/kg twice a day, every day	There is feasibility of the CBD transdermal delivery systems use, for the treatment of alcohol-induced neurodegeneration.	Liput *et al.* (2013)
Hartley guinea pigs	N/A	CBD	Scotch™ patches placed over CBD gel with or without enhancers (Transcutol HP**)	48 hours	**CBD gel formulation + patch + enhancer (Transcutol HP):** 500uL for 48 hours **CBD gel formulation + patch**: 500ul for 48 hours	View table 2	Paudel *et al.* (2010)

*Pancream: oil-in-water vanishing cream formulation.
**Transcutol HP: 2-(2-etoxietoxi) etanol, an organic solubilizer used in commercial formulation.
VEH= Vehicle
CFA= Freud's adjuvant
EtOH= Ethanol
N/A= Not available

Table 4: Methodological details of pre-clinical included studies concerning the cannabinoids topical delivery studies (6).

Animals	Total N	Drug	Formulations	Exposure/ Duration	Treatment	Drug Response	Reference
Male CD1 mice, with induce inflammation with Croton oil dissolved in acetone	~ 90	CBD	Solution	6 hours	(1) **Control group:** received irritant solution, only (2) **Control group for anti-inflammatory activity:** irritant solution + indomethacin (NSAID) (3) **Psychoactive cannabinoids:** Δ9 - tetraidrocannabinol (Δ9 -THC), Δ8 - tetraidrocannabinol (Δ8 -THC) or cannabinoid antagonist (Δ8 - tetraidrocannabivarin) (4) **Non-psychoactive cannabinoids:** cannabichromene (CBC), cannabidiol (CBD), and their lower homologues (CBCV and CBDV)	At topical anti-inflammatory activity the data showed differences in the terpenoid moiety of phytocannabinoids, which showed more importance when compared to alkyl residues.	Tubaro *et al.* (2010)
Wild- type and CB1/CB2 receptor-deficient (Cnrl/2-/-) C57B1/6J mice, with induced hypersensitivity by DNFB (1-Fluoro-2,4 dinitrobenzene)	N/A	THC	Acetone solution	**~19 days**	(1) THC mice treated immediately before DNFB sensibilization; 24- and 48-hours hours after DNFB sensibilization (2) DNFB sensibilization and vehicle treatment (3) Vehicle only.	In conclusion the authors presented enough data to demonstrate the anti-inflammatory activity of the THC not only by the CB1 and CB2 receptors.	Gaffal *et al.* (2013)
Standard bred horses, submitted to wounds formation or scars on their forelimbs, contaminated with fresh feces	6 horses	1% CBD extract	N/A	**Treatment:** 13 days **Study duration:** 42 days	<u>Leg with 2 wounds:</u> **Wound 1:** UMF 5 manuka honey (carrier), daily **Wound 2:** Treated with 1% CBD extract in UMF 5 manuka honey, daily <u>Leg with one wound:</u> **Wound 1:** Sterile saline 0,9% **Wound 2:** UMF 20 manuka honey (carrier) **Wound 3**: UMF 5 manuka honey (carrier) The treatment followed daily application for 13 days	The authors were unable to determine the effect of 1% CBD extract in a carrier UMF 5 manuka honey applied topically.	Mciver *et al.* (2020)

Male C57BL/6 mice, induced to encephalomyelitis (EAE)	40	CBD 1%	O/A cream	28 days	**Naïve:** did not received treatment (5 animals) **EAE:** EAE mices without CBD treatment (10 animals) **EAE + 1% CBD-cream:** EAE mices treated with topical 1% CBD- cream, every 24 hours (10 animals) **EAE + vehicle cream:** EAE mice treated with topical application os vehicle cream, every 24 hours (5 animals) **Control group + 1% CBD-cream:** healthy mice treated with 1% CBD-cream every 24 hours (5 animals) **Control group + vehicle cream:** healthy mices without treatment with CBD, only the vehicle cream for 24 hours (5 animals)	In conclusion, CBD treatment showed to protect neurons from the inflammatory cascade after EAE induction and its degeneration.	Giacoppo *et al.* (2015)
Nude male rats (RH-FOXN1RN)	25	CBD	Cream	4 weeks	**Control:** application of nontoxic hydrophilic petrolatum for 20 mi, for 4 weeks every 12hs **UVA exposure:** in 365 nm, increasing from 0,5 to 5J/cm² every 48 hours for 4 weeks **UVA + CBD:** UVA exposure (= group 2) + topical treatment for 20 min with 2,5g CBD in 100g petrolatum, every 12 h for4 weeks **UVB exposure:** 312nm in increasing doses for 0,02 to 2J/cm² every 48 hours for 4 weeks **UVB + CBD:** exposure (= group 4) + topical treatment 20 min with 2,5g CBD in 100g petrolatum, every 12 h for4 weeks	Side effects caused by UVA and UVB exposure could be revert with the use of CBD, since acts by normalizing the expressions of keratinocytes proteins and its metabolization and function.	Atalay *et al.* (2021)
Female outbred CD1 mice (UQBR-AIBN), with contaminaded back skin with bacterial inoculum in the concentration of 5 x 10^7 CFU	24	CBD	Gel	48 hours	**Control:** saline solution **Bactroban (2% mupirocin)**: topically, at time 0, 12, 24 and 32hours right after the infection **CBD (BTX 1503)**: orally, at time 0, 12, 24 and 32 hours right after the infection **CBD (BTX 1503 gel)**: topically, at time 0, 12, 24 and 32 hours right after the infection **CBD (BTX 1204 gel):** topically, at time 0, 12, 24 and 32 hours right after the infection	CBD and its analogs are a great choice for a new class of antibiotics.	Blaskovich *et al.* (2021)

N/A= Not available

Table 5: Methodological details of clinical studies included using eye mucosal as topical pathway.

Study Design	Population	Total N	Drug	Formulations	Exposure/ Duration	Treatment	Drug Response	Reference
Case- Control	Healthy volunteers	N/A	Δ^9-THC	Mineral oil	7 hours	In one of the eyes, the volunteers applied the vehicle and Δ9-THC, in the other eye, 1 drop of each	THC did not appear to reduce IOP, on the other hand, it could be possible that exists other cannabinoids that may produce this effect, requiring more studies.	Green and Roth (1982)

N/A= Not available

Table 6: Methodological details of clinical studies included using oromucosal as topical pathway.

Study Design	Population	Total N	Drug and Pharmaceutical form	Exposure/ Duration	Treatment	Drug Response	Reference
Case- reports	Patient with Stiff-person syndrome (SPS)	1	Sativex®	N/A	40-year-old man, with SPS with progressive muscle stiffness and intermittent spasm for 6 years. The patient treatment included: diazepam, levetiracetam, levomepromazine, corticosteroids, oral baclofen, mofetil mycophenolate and IV immunoglobulin infusion. The multidisciplinary team decided to introduce the off- label use of Sativex®, the daily dose for the patients was two- sprays for optimum symptom relief. The patients answered the Quality of Life (QoL) interview, before and during the 14 months of therapy.	THC: CDB (Sativex®) could be an alternative for SPS patient by further investigation.	Vicente-Valor *et al.* (2013)
	Patient with Multiple Sclerosis	1	Sativex®	4 weeks	53 years old man with relapsing rating multiple sclerosis diagnosed in 1999. The patient treatment included maximum doses of tizanidine, baclofen, benzodiazepines, and botulinum toxin. Sativex® was included in therapy managing to an optimal tolerance of 6 sprays applications/day after trying different dosages. 4 weeks later patient presented convulsive seizure, leading to the discontinuation of the treatment.	Tests proved the safety of Sativex® in multiple sclerosis patients. Also, was proven by statistical data that the convulsive seizure was more likely due to patients' previous conditions.	Aparicio Rosana and Polo Virginia (2013)
Prospective pilot Study	Patients with MS diagnosis	15	THC/CBD spray	4 weeks	Patient dependent	THC/CBD oromucosal spray has shown effectiveness in improving the overactive bladder symptoms and spasticity in MS patient.	Maniscalco *et al.* (2018)
Open Label Trial	Randomized, double-blind, placebo-controlled, parallel-group	Patients with central neuropathic pain syndrome due to MS	66	THC/CBD spray (2,7mg of THC and 2,5mg of CBD)	5 weeks	**Clinical Trial Phase** **Control group:** placebo (ethanol: propylene glycol 50:50) **Treatment group:** THC: CBD spray, not specific dose was set, although patients were informed to increase the atomization every day up to 48 sprays/day, but no passing 8 sprays within 3 hours interval and reduce if any signs of intoxication was noticed	**Open Label Phase** N/A	Cannabis-based medicine showed to be well tolerable and effective in pain reduction and sleep disturbance in patients with multiple sclerosis.	Rog *et al.* (2005)

ls					**Control group:** received placebo and analgesia previous prescribed was maintained			
	Randomized, double-blind, placebo-controlled parallel groups study	Patients with unilateral peripheral neuropathic pain and allodynia	125	Sativex®	36 weeks	**Treatment groups:** received Sativex® initially in a maximum of 8 sprays over 2 hours. Later, patients began home dose titration, in a maximum dose of 8 sprays per 3hours interval, no passing 48 sprays in 24 hours. Analgesia previous prescribed was maintained	After the trial, patients were invited to participate to the open- label phase where all the patients received Sativex®	With those results, is possible to affirm that Sativex is effective in the relief of peripheral neuropathic pain when used with existing analgesic medication.	Nurmikko *et al.* (2007)
	Randomized double-blind, placebo-controlled, parallel-group study	Patients with central neuropathic pain syndromes associated to MS	66	THC/ CBD spray	years	**Control group:** received placebo. **Treated group:** received THC/CBD (21.6 mg/20 mg), from 8 every 3 hours, or 48 administrations in 24 hours, not passing 50 sprays, for 5 weeks	After the treatment, patients entered open label if wanted to, where all of them received THC/CBD	THC/CBD treatment in patients with CNP and MS, after 2 years of treatment, may cause adverse events, in which the most common are dizziness and nausea, and others are characterized as mild to moderate severity.	Rog; Nurmikko and Young (2007)
	Double-blind, randomized, placebo-controlled, parallel-group study	Male and female subjects with (CNP) due to (MS)	393	THC/CBD spray	1 year	Lasted 14 weeks. Treatment was divided in placebo group and THC/CBD group The spray administration was limited to 12 sprays in 24-hour period	**Part 1:** 14- weeks: It was an open-plan treatment phase consisting of 2-week re-titration and 12-week stable dose phase with THC/CBD spray **Part 2:** 4- weeks: randomized- withdrawal phase, placebo- treated in double- blind manner. Treatment in re-titration period started and increased until 12 sprays a day: day 1 and 2: one spray; day 3 and 4: two sprays; day 5: three sprays; day 6: four sprays; day 7: five sprays, until reaches 12 sprays a day	The study presented useful results about the efficacy of THC/CBD spray in the treatment of CND caused by MS.	Langford *et al.* (2013)

Multicenter, double-blind, randomized, placebo-controlled, parallel-group, phase 2 trial	Volunteers aged from 18-80 years old, with possible, laboratory-supported probable, probable or definitive amyotrophic lateral sclerosis or primary lateral sclerosis	59	Nabiximol spray (THC/CBD-2,7mg/2,5mg)	13 weeks	**Control group**: received placebo **Treated group**: received nabiximol, 100ul of the solution was pumped, for 14 days in which they adjusted to reach the optimal dose, and them maintained the dose for 4 weeks	After 6-weeks patients were invited to participate to the open- label phase where all the patients received nabiximol	The nabiximol may be a great choice to control spasticity in patients with motor neuron diseases.	Riva *et al.* (2019)

N/A= Not available

Table 7: Methodological details of clinical included studies using transdermal (3) and topical skin as pathway (13).

Study Design	Population	Total N	Drug	Formulation	Exposure/ Duration	Treatment	Drug Response	Reference
Case Series	Male and Female patients with Pyoderma Gangrenosum (PG) wounds	3	Argyle™ and Bedrolite®	spray	N/A	**Patient 1** 1mL daily of Argyle (THC 5mg/mL and CBD 6mg/mL **Patient 2 and 3** 0,5 to 1,0mL Bedrolite (THC 7mg/mL and CBD 9mg/mL) twice daily; with 1 to 3 times daily for breakthrough pain.	Cannabinoids extracted: THC and CBD in union, showed to be a great choice to treat painful skin conditions.	Kiefer (2017)
	Male and Female patients with Pyoderma gangrenosum (PG)	3	Argyle™ and Bedrolite®	spray	N/A	**Patient 1** 1mL of Argyle® was applied to wound bed daily followed by application of inelastic compression bandaging. **Patients 2 and 3** 0,5mL-1,0mL of Bedrolite® to the wound bed 2x a day, and 1-3x daily for pain.	Case series demonstrated the potential of THC and CBD in provide effective analgesia in the PG.	Maida and Corban (2017)
	Children with epidermolysis bullosa (EB), using CBD by self-medication	3	CBD	spray, oil and cream.	N/A	**Patient 1** 6-month-old boy presenting recessive dystrophic EB. The treatment included: wound care by using petrolatum ointment, emu oil, and silicone-based dressing. Mupirocin for decolonization and diphenhydramine and morphine. No recovery was significant, so the parents switch to self-initiated CBD spray (tincture) 2 to 3 times a day. **Patient 2** 3-year-old girl presented erosions on the extremities and oral mucosa, which was determined to by caused by EB simplex, generalized severe with KRT5 mutation. The treatment included: petrolatum ointment with coconut oil and spot treatment with zinc oxide and allantoin 6% cream, and 10% urea in the keratodermas, topical bacitracin, and dilute bleach baths. By recommendation	The reports discussed demonstrated that the CBD use reduced pain and blistering and promoted rapid wound healing.	Chelliah *et al.* (2018)

					the mother start to be applied emu oil and CBD oil topically in the face, extremities, and trunk twice a day. **Patient 3** 10-year-old boy diagnosticated with EB simplex localized in the neckline since the first month of age. The wounds increased to posterior upper arms and soles. The keratodermas hinder the patient to walk. The treatment included: topical antibiotics and emollients, naproxen and gabapentin. The parents self-initiated CBD oil and cream topically.		
Patients with Epidermolysis bullosa (EB)	3	Cannabinoid based medicine containing tetrahydrocannabinol (THC) and cannabidiol (CBD)	N/A	N/A	**Patient 1** 64- year- old woman with junctional EB generalized intermediate, reporting pain scale of 9 /10. CBM oil (20mg/mL CBD and 13mg/mL THC) was administrated by sublingual path, in a dose of 2,5mg of CBD and 1,65mg THC, 4x daily. Further, after a few years the patient started using topical CBM oil with CBD concentration at 1mg and THC at 0,65mg, in daily applications. **Patient 2** 41- year- old man diagnosed with JEB-gen intermed, reporting pain scale of 9/10. CBM oil (20mg/mL CBD and 13mg/mL THC) was administrated by sublingual path. After 1 month treatment he reached a treatment in a dose of 3mg of CBD and 1,95mg THC, 4x daily. After 5 month an addition of the 5mg oxycodone-IR 3x daily was necessary. **Patient 3** 36- year- old man dystrophic EB generalized severe (RDEB-gen sav). CBM oil (20mg/mL CBD and 13mg/mL	There were some limitations considering the pharmacological activity of CBD, by suppression of CB1/2 receptors, which its presence is unknown in EB.	Schräder (2020)

						THC) was administrated by sublingual path and intrapulmonary, for two years.		
	Healthy male Asian, volunteers	11	3% Cannabis seeds extract	cream (BP3%CSE)	3 months	Each volunteer received two similar appearance products: base plus 3% Cannabis seeds extract and the other containing pure base. the bases were applied in the forearms of each volunteer, and a patch were placed with 1,0g base (left) and base plus 3% Cannabis seeds extract (right).	3% Cannabis seeds extract cream is safe to be use and showed great action against erythema and skin sebum	Ali and Akhtar (2015)
Case- control	Males and Females, healthy volunteers.	3	hemp oil	Cream	3 days	**Preview case report:** Male driver (43 years old) was stopped by police officers, which documented disorientation, deficiency in concentration and unsafe walk. The driver claimed non consumption or inhalation of cannabinoids drugs. The driver only assumed the use of cream containing hemp oil, the man was using the cream for at least 3 days for bruises in different body parts. **Case control:** Cream containing hemp oil was applied by the volunteers every 3-4 hours in an extensive area.	Considering the application of the oil by the driver, the amount of THC could not be enough to guarantee an absorption through the skin even with frequent application.	Hess; Krämer and Madea (2017)
	Healthmale Asians, with no dermatological conditions or allergy.	11	C. sativa seed 3% extract	dermocosmetic formulation (14% paraffin oil, 2% abil EM 90 and 1% fragrance)	3 months	A base and the dermocosmetic was given to each volunteer: 500mg of both, 2x/day in mornings and evenings, for 12 weeks.	Extract used in the dermocosmetic improved the skin surface and it could be useful to skin aging.	Ali *et al.* (2020)
Cross- sectional	Local inhabitants of North-West Frontier Province in Pakistan.	328	Crushed plant applied directly		N/A	Patient dependent.	In order to maintain the cultural heritage of the natives, is important to confirm the therapeutic uses of the plants, considering the scientific criteria.	Abbasi *et al.* (2010)

	Outpatient palliative patients.	58	According to the patients preview experiences, including topical		N/A	Patient dependent.	Results reported reinforce the importance of further investigation about the clinical efficacy and ideal dosing of the substances.	Highet *et al.* (2020)
Patient reported Statistical	Volunteers with diagnosed eczema.	16	CBD	N/A	2 weeks	The patients had the option of ordering a CBD topical for no charge or using their own CBD topical. Administration was patient dependent.	CBD showed to be a promising treatment to skin conditions.	Maghfour *et al.* (2020)
Longitudinal	Costumers reporting adverse events.	N/A	PlusCBD™	N/A	2 years (January 2018-December 2019)	N/A	Despite these two cases discussed, the authors affirmed that the article may contribute to future studies with CBD formulation.	Schmitz; Lopez and Marinotti (2020)
Open label	Male and female pediatric patients with Fragile X syndrome (FXM)	20	CBD (ZYN002)	Transdermal gel	12 weeks and 24 months	**Clinical Trial Phase:** • The ZYN002 application were done as an adjunct treatment to their existing treatment. Once daily 50mg dose; twice daily 50mg dose equivalent to a 100mg total dose; twice daily 125mg dose equivalent to a 250mg total dose. • The doses were increased to these three doses, according to the tolerability assessment, clinical response every 2 weeks for 6 weeks (titration period) and investigator judgment. **Open Label Phase:** Patients which showed great acceptance of the first dose in the period of 12 weeks, were invited to continue to the study for the next 24 months. CBD gel produced clinically and statistically significancy by reducing the anxiety, behavioral and emotional symptoms.	CBD gel produced clinically and statistically significancy by reducing the anxiety, behavioral and emotional symptoms.	Heussler *et al.* (2019)

Clinical Trial		Children with one or more year since epileptic encephalopathies diagnosis	48	42 % CBD	Transdermal gel	26 weeks	**Initial dose:** 250 mg/d for patients weighing 25 kg or less and 500 mg/d for patients weighing more than 25 kg. **1 month:** 500 mg/d for patients weighing 25 kg or less and 750 mg/d for patients weighing more than 25 kg **2.5 months:** 750 mg/d for patients weighing 25 kg or less and 1000 mg/d for those weighing more than 25 kg	CBD transdermal gel was well tolerated and reduced impaired awareness seizures and tonic-clonic seizures, as well as improved daily activities.	Scheffer *et al.* (2021)
	Prospective open label clinical trial	Patients with intractable integumentary wounds involving mucous membranes caused by Non-Uremic Calciphylaxis (NUC)	2	VS-12 and VS-14 (cannabis-based medicines)	N/A	N/A	Patients received daily topical applications, until wound closure (wound bed with 100% epithelization).	Cannabinoids-based medicines are a promising novel, non- invasive and safe treatment option for NUC leg ulcers.	Maida *et al.* (2020)
	Single- center, double-blind, randomized and placebo-controlled trial	Patients with symptomatic peripheral neuropathy	29	CBD oil cream	N/A	3 weeks	**Clinical trial** (1) **Control group:** received Emu oil cream as placebo. (2) **Treatment group:** received CBD oil cream (250mg/ 3fl). **Open Label** After 4 weeks the patients were invited to participate to the second phase of the study where all the patients received CBD oil.	Clinical results favored of CBD transdermal use, with that in mind, these medication by topic administration may be a great alternative for neuropathic pain treatment.	Xu *et al.* (2020)
	Randomized double-blinded placebo-controlled interventional pilot study	Male and female, health volunteers	66	CBD and aspartame	Cream	14 days	(1) CBD alone (2) Placebo group CBD and aspartame group	Comparing to placebo group, CBD with aspartame decreased atopic dermatitis symptoms such as erythema, papulation, and crusting. The CBD alone groups did show some reduction in dermatitis atopic symptoms, but not as much as the aspartame group.	Gao *et al.* (2022)

Single- center, randomized controlled trial	Male and female, health volunteers	10	CBD	Shea butter	1 week	**Phase 1: skin test** 1-mL syringes of 6.2 mg/mL CBD with shea butter applied in nondominant thumb basal joint 2 times daily. Symptom diary was provided for recording skin changes and other noticed adverse events or side effects **Phase 1: randomized trial** Control group: shea butter alone Treated group: 6.2 mg/mL CBD with shea butter Applied 1 mL of the cream to their symptomatic thumb basal joint 2 times daily. Symptom diary was provided for recording skin changes and other noticed adverse events or side effects	In this single-center, randomized controlled trial, twice-daily topical CBD application resulted in improvements in thumb basal joint arthritis-related pain and disability without adverse events. Larger, multicenter clinical trials are warranted to further investigate the safety, therapeutic potential, and dose response of CBD for musculoskeletal pain.	Heineman *et al.* (2022)
	Healthy subjects with symptomatic thumb basal joint arthritis	18			2 weeks			

N/A= Not available

Table 8: Adverse events reported by pre-clinical and clinical included articles.

Author	Administration pathway	Formulation	Adverse event reported
Colasanti; Powell and Craig (1984)	Eye	Δ^9-THC	Neurotoxicity
Riva *et al.* (2019)	Oromucosal	CBD: THC spray	Nausea and anxiety, disease progression, asthenia, dizziness, somnolence, vertigo, muscle spasticity or rigidity and dry mouth
Rog *et al.* (2005)	Oromucosal	CBD: THC spray	Nervous system disorders (dizziness)
Nurmikko *et al.* (2007)	Oromucosal	Sativex® spray	Gastrointestinal discomforts (nausea, vomit, diarrhea and constipation), nervous system (severe and mild- moderate psychiatric) and oral discomfort. Ischemic attack, a serious adverse event
Langford *et al.* (2013)	Oromucosal	CBD: THC spray	
Rog; Nurmikko and Young (2007)	Oromucosal	CBD: THC spray	
Hannon *et al.* (2020)	Transdermal	CBD: THC	Erythema
Scheffer *et al.* (2021)	Transdermal	CBD	Gastrointestinal discomforts
Giacoppo *et al.* (2015)	Topical skin	CBD	Allergic reaction in local or systemic

9. SUPLEMENTARY MATERIAL

Plataform	Subject vocabulary	Descriptors and terms used in the Search strategy					
		1	2	3	4	5	6
PUBMED	MeSH - Medical Subject Headings	Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"
PUBMED PMC	MeSH - Medical Subject Headings	Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"
BVS / BIREME	DeCS	Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"
		Cannabidiol	Cannabis	"Marihuana Medicinal"	"Administración Tópica"	"Administración Cutánea"	"Parche Transdérmico"
		Canabidiol	Cannabis	"Maconha Medicinal"	"Administração Tópica"	"Administração Cutânea"	"Adesivo Transdérmico"
Scopus		Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"
WEB OF SCIENCE		Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"
EMBASE	Emtree	Cannabidiol	Cannabis	medical marijuana *use preferred term:* medical cannabis	administration, topical *use preferred term:* topical drug administration	administration, cutaneous *use preferred term:* cutaneous drug administration	"Transdermal Patch"
Cochrane Library	MeSH - Medical Subject Headings	Cannabidiol	Cannabis	"Medical Marijuana"	"Administration, Topical"	"Administration, Cutaneous"	"Transdermal Patch"

5. CONCLUSÃO

Embora os resultados obtidos mostraram efeitos promissores após a administração do canabidiol tanto em estudo com animais quanto em humanos, a concentração do bioativo foi um obstáculo quando se tratava de comparação da formulação e as dosagens adotadas, entre os estudos.

A administração tópica mostrou eficiência na adesão ao tratamento e redução de efeitos adversos. No entanto, muitos deles apresentaram baixa eficácia no tratamento com CBD isolado em comparação com a combinação de CBD com THC. Esta, por sua vez, foi a formulação mais presente na literatura, visto que já existem produto farmacêuticos disponíveis para comercialização.

Mesmo assim, o presente estudo contribui para que novas hipóteses sejam levantadas a respeito da capacidade da formulação em afetar positivamente, ou negativamente, nas atividades dos canabinóides administrados pela via tópica/transdérmica bem como na eficácia do uso do canabidiol como ativo principal no tratamento de diversas condições de saúde.

6. REFERÊNCIAS BIBLIOGRÁFICAS

BABSON, K. A.; SOTTILE, J.; MORABITO, D. Cannabis, cannabinoids, and sleep: a review of the literature. **Current psychiatry reports**, 19, n. 4, p. 1-12, 2017.

CHESHER, G. B.; JACKSON, D. M. The quasi-morphine withdrawal syndrome: effect of cannabinol, cannabidiol and tetrahydrocannabinol. **Pharmacology Biochemistry and Behavior**, 23, n. 1, p. 13-15, 1985.

COLASANTI, B. K.; POWELL, S. R.; CRAIG, C. R. Intraocular pressure, ocular toxicity and neurotoxicity after administration of Δ9-Tetrahydrocannabinol or cannabichromene. **Experimental Eye Research**, 38, n. 1, p. 63-71, 1984. Article.

ELSOHLY, M. A.; HARLAND, E. C.; BENIGNI, D. A.; WALLER, C. W. Cannabinoids in glaucoma II: the effect of different cannabinoids on intraocular pressure of the rabbit. **Curr Eye Res**, 3, n. 6, p. 841-850, 1984/06 1984.

FARAG, S.; KAYSER, O. The cannabis plant: botanical aspects. In: **Handbook of Cannabis and Related Pathologies**: Elsevier, 2017. p. 3-12.

FIANI, B.; SARHADI, K. J.; SOULA, M.; ZAFAR, A. et al. Current application of cannabidiol (CBD) in the management and treatment of neurological disorders. **Neurological Sciences**, 41, n. 11, p. 3085-3098, 2020/11/01 2020.

HAVLICKOVA, B.; FRIEDRICH, M. The advantages of topical combination therapy in the treatment of inflammatory dermatomycoses. **Mycoses**, 51, n. s4, p. 16-26, 2008.

INSTITUTE, J. B. Joanna Briggs Institute reviewers' manual 2014. **Adelaide: The Joanna Briggs Institute**, 2014.

LEE, M. C.; PLONER, M.; WIECH, K.; BINGEL, U. et al. Amygdala activity contributes to the dissociative effect of cannabis on pain perception. **PAIN®**, 154, n. 1, p. 124-134, 2013.

MASSI, P.; SOLINAS, M.; CINQUINA, V.; PAROLARO, D. Cannabidiol as potential anticancer drug. **British Journal of Clinical Pharmacology**, 75, n. 2, p. 303-312, 2013.

MATOS, R. L.; SPINOLA, L. A.; BARBOZA, L. L.; GARCIA, D. R. et al. O uso do canabidiol no tratamento da epilepsia. **Revista Virtual de Química**, 9, n. 2, p. 786-814, 2017.

MECHOULAM, R.; SHANI, A.; EDERY, H.; GRUNFELD, Y. Chemical basis of hashish activity. **Science**, 169, n. 3945, p. 611-612, 1970.

MEIRI, E.; JHANGIANI, H.; VREDENBURGH, J. J.; BARBATO, L. M. et al. Efficacy of dronabinol alone and in combination with ondansetron versus ondansetron alone for delayed

chemotherapy-induced nausea and vomiting. **Current medical research and opinion**, 23, n. 3, p. 533-543, 2007.

RADWAN, M. M.; CHANDRA, S.; GUL, S.; ELSOHLY, M. A. Cannabinoids, Phenolics, Terpenes and Alkaloids of Cannabis. **Molecules**, 26, n. 9, May 8 2021.

RUSSO, E. B.; GUY, G. W.; ROBSON, P. J. Cannabis, pain, and sleep: lessons from therapeutic clinical trials of Sativex®, a cannabis-based medicine. **Chemistry & biodiversity**, 4, n. 8, p. 1729-1743, 2007.

7. ANEXO

DECLARAÇÃO DE ISENÇÃO DO PARECER DO COMITÊ DE ÉTICA EM PESQUISA

DECLARAÇÃO

Em observância ao **§5º do Artigo 1º da Informação CCPG-UNICAMP/001/15**, referente a Bioética e Biossegurança, declaro que o conteúdo de minha Dissertação de Mestrado, intitulada **"AVALIAÇÃO DA ATIVIDADE DA CANABIDIOL EM FORMULAÇÕES TÓPICAS",** desenvolvida no Programa de Pós-Graduação em Ciências Médicas da Faculdade de Ciências Médicas da Unicamp, não versa sobre pesquisa envolvendo seres humanos, animais ou temas afetos a Biossegurança.

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