Universidade de Lisboa

Faculdade de Farmácia

Cyclodextrins as Medicine and Therapeutic Adjuvants in Drug Delivery Systems

Bárbara Claro Ferreira

Monografia orientada pela Professora Doutora Helena Maria Cabral Marques, Professora Associada com Agregação da Faculdade de Farmácia da Universidade de Lisboa, e coorientada pelo Doutor Oluwatomide Adeoye, Formulation specialist, VectorB2B-Drug Development Association, Lisboa

Mestrado Integrado em Ciências Farmacêuticas

2021

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Trabalho Final de Mestrado Integrado em Ciências Farmacêuticas apresentada à Universidade de Lisboa através da Faculdade de Farmácia

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Resumo

Ao longo de várias décadas desde a sua descoberta, as ciclodextrinas foram sendo usadas como excipientes e veículos de muitas substâncias ativas para aumentar a solubilidade aquosa, estabilidade físico-química e fisiológica e a veiculação destes fármacos. Atualmente, as ciclodextrinas estão presentes nas mais diversas formulações de fármacos, como comprimidos, pomadas, gotas oftálmicas, entre outras. Adicionalmente a esta função de excipientes, recentemente as ciclodextrinas também são encontradas como substância ativa no tratamento de algumas patologias.

Esta monografia foca-se em avanços recentes e encorajantes sobre a aplicação de ciclodextrinas como fármacos e adjuvantes terapêuticos em sistemas de veiculação de fármacos. As primeiras três secções fornecem uma perspetiva histórica e descrição das ciclodextrinas e ciclodextrinas modificadas, enquanto que a quarta secção fornece uma revisão dos avanços atuais no uso terapêutico das ciclodextrinas. O potencial terapêutico das ciclodextrinas é maioritariamente derivado da sua habilidade em formar complexos com colesterol e outras moléculas. Deste modo, estas moléculas estão a ser estudadas como novo agente terapêutico em muitas patologias conhecidas onde outras classes terapêuticas não conseguem reduzir a severidade da doença e/ou controlar a mesma, e até como entidades químicas para algumas patologias sem terapia conhecida. Na maioria das situações, as ciclodextrinas modificadas são frequentemente usadas devido ao facto de terem uma maior afinidade para o alvo terapêutico, colesterol, quando comparadas às ciclodextrinas "convencionais", tendo também uma menor taxa de efeitos adversos e toxicidade.

Em algumas patologias, o mecanismo exato da atividade das ciclodextrinas é desconhecido. Assim é necessário continuar a investigação, incluindo a iniciação e conclusão de muitos estudos clínicos antes que estas moléculas cheguem a farmácias, hospitais e doentes como fármaco. A produção destes dados, incluindo considerações toxicológicas, são relevantes para a sua aprovação e uso posterior ou comparticipação dos diversos sistemas nacionais de saúde. Assim, num futuro próximo, poderemos ter novas estratégias disponíveis com diferentes ações benéficas no organismo humano, com a máxima segurança e benefício para o doente.

Palavras-chave: ciclodextrina, terapêutica, colesterol, cancro, antivíricos

Abstract

Over several decades since its discovery, cyclodextrins have been used as excipients and vehicles of many active pharmaceutical ingredients to enhance aqueous solubility, physicochemical and physiological stability, and the deliverability of these drugs. Currently, cyclodextrins are present in diverse drug formulations, such as pills, ointments, ophthalmic drops, among other formulations. In addition to the function of pharmaceutical excipients, cyclodextrins have also found recent use as active substances in the treatment of some pathologies.

This monograph focuses on some recent and encouraging advances in the application of cyclodextrin as medicine and therapeutic adjuvants in drug delivery systems. The first three sections provides an historical overview and description of cyclodextrins and modified cyclodextrins while the fourth section reviews the current advances in the therapeutic use of cyclodextrin molecules. Cyclodextrin's therapeutic potential is mainly derived from their ability to form complexes with cholesterol and other molecules. As such they are being studied as new therapeutic agents in many known pathologies where other therapeutic classes are not able to reduce the severity of the disease and/or even control it, and as new chemical entities for some pathologies with no known therapy. In most situations, modified cyclodextrins are more frequently used as they will have greater affinity for the therapeutic target, cholesterol, when compared to "conventional" cyclodextrins, also having a lower rate of adverse effects and toxicity.

In some pathologies, the exact mechanism of cyclodextrins activity is unknown. Further research activities, including the initiation and completion of many clinical trials are required before these molecules reach pharmacies, hospitals and patients as active substance. The production of these data, including toxicity consideration, will be relevant for its approval and later use or reimbursement by multiple national health systems. Thus, in a near future, we could have new strategies available with different beneficial actions in the human body, with maximum safety and benefit for the patient.

Keywords: cyclodextrin, therapeutic, cholesterol, cancer, antivirals

Agradecimentos

Em primeiro lugar, agradeço aos meus pais por todo o suporte ao longo destes anos, à minha irmã por todas as dicas e pormenores que me foi dando, e por me animarem em todos os dias menos bons.

Em segundo, a todos os amigos de sempre, aos que fui fazendo na faculdade ao longo destes últimos anos e que sempre estiveram ao meu lado para todas as sessões de estudo e descontração.

Por último, mas não menos importante, à minha orientadora, Professora Doutora Helena Marques, e co-orientador, Doutor Oluwatomide Adeoye por me terem orientado no conteúdo e revisão da escrita desta monografia.

Abreviaturas

CD – Cyclodextrin

EMA – European Medicine Agency

HP-β-CD - Hydroxypropyl-β—cyclodextrin

NPC - Niemann-Pick disease type C

LXR - Liver X-receptor

HIV – Human Immunodeficiency Virus

M-β-CD – Methyl-β-Cyclodextrin

FA-M-β-CD – Folate-appended Methyl-β-Cyclodextrin

HA/M-β-CD – Hyaluronic Acid Methyl-β-Cyclodextrin

PBC – Poly-β-Cyclodextrin

KT cells – Keratin-forming Tumor Cell Line

TBW - Total Body Weight

IBW - Ideal Body Weight

Aβ protein - Amyloid β protein

AD - Alzheimer Disease

CNS - Central Nervous System

BBB – Blood-Brain Barrier

HSV - Herpes Simplex Virus

HMPV - Human Metapneumovirus

PIV 3 - Parainfluenza Virus Type 3

HBV – Hepatitis B Virus

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1 Cyclodextrins: Brief History background and description

Cyclodextrins (CDs) are cyclic oligosaccharides made up of repeating units of α(1→4)glucopyranosides. The most common commercially available ones have 6, 7 and 8 (α, β, γ) units. They were first discovered by Villiers in the last decade of the 19th century, and have since found utility in different areas, such as food, cosmetics, toiletries, catalysis, textile industry, biotechnology, nanotechnology and pharmaceutical industry. In pharmaceutical area, the production of industrial quantities cyclodextrins, the discovery of their reasonably good toxicological profile led to their application as excipients or inert components of medicines. However, recent studies have shown that CDs are not as inert as initially though, and could be used to treat many illness (1–3).

While their pharmaceutical industry utility as excipients, is mainly derived from their ability to form complexes with many drug molecules thereby modifying their physicochemical properties such as aqueous solubility, physicochemical and physiological stability, and ability to control drug delivery, their applications as medicines is often a consequence of their ability to interact with and extract cholesterol from cell membranes and tissues. In addition, CDs can protect several molecules from oxidation and volatilization and a solubilizing power in nonpolar vehicles (3,4).

2 Structure and Characteristics of Cyclodextrins

CDs can be described as a truncated cone, with a hydrophobic internal cavity (it has the presence of oxygen atoms ring), while the exterior cavity is hydrophilic due to the presence of primary and secondary hydroxyl functional groups. This structure enables to form complexes with a vast number of compounds through non-covalent interaction (5–7).

The 3 natural CD, α, β and γ CD have 6, 7 and 8 glucose units respectively and differ in ring size and solubility. Smaller CD, that is those with less than 6 glucose units, were initially hard to produce due to steric barriers and their small size, but group of scientists have recently demonstrated that it can be obtained by introducing a bridge with Suzuki glycosylation. On the other hand, bigger CD (> 8 glucose units) are expensive due to

difficulties on purifying methods. When in solid state, CD are as durable as sucrose or starch, and can be kept for many years at room temperature without any identified deterioration, although temperature can be a deterioration factor. In addition, CDs have a large number of hydrogen donors and receptors, they cannot be absorbed by gastrointestinal tract in their normal form, they are not hydrolysable by human saliva and pancreatic amylase, and they are only fermented by intestinal microflora (1,5,8–11).

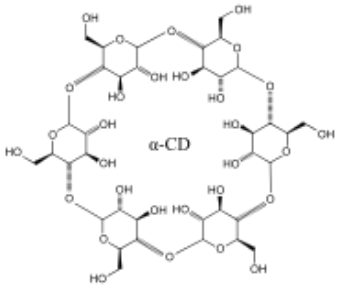

FIGURE 1 - ALPHA-CYCLODEXTRIN

The hydroxyl groups on the outside surface of CDs are able to create hydrogen bonds with various molecules, providing the capability to form complexes with lipophilic molecules insoluble in water. In addition, it is possible to change or add a particular ligand to CD surface in order to modify/enhance their physicochemical properties or as a strategy controlled targeted drug delivery (1,5,8).

Beta-Cyclodextrins (β-CDs) have been used since the beginning CD's pharmaceutical applications due to ease of production and ability to form complexes with a lot of drugs used in therapeutics. Nevertheless, β-CD has poor aqueous solubility, being a potentially nephrotoxic molecule. Since the structure and cytotoxic characteristics of CD are important considerations in drug design and drug product development, the important question of CD substituents have been studied to evaluate the potential to conferring higher toxicity (because of a higher affinity to cholesterol, like methyl substituent of β-CD) or higher complexation ability (9,12,13).

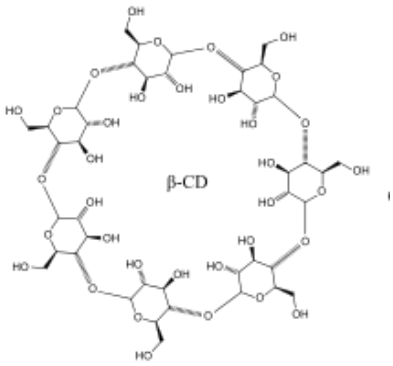

FIGURE 2 - BETA-CYCLODEXTRIN

The complexation power of CDs is dependent on various factors such as adequate guest molecule size, guest chemical structure and physiochemical properties, type of CD, temperature and thermodynamic interactions, complex preparation methods, presence of ternary compounds or polymers. Also, the driving force for the observed molecular interactions between CD and drug molecule have been reported to include electrostatic, van der Waals' and hydrophobic interactions, implying a dynamic interaction between both molecules (5,9,14).

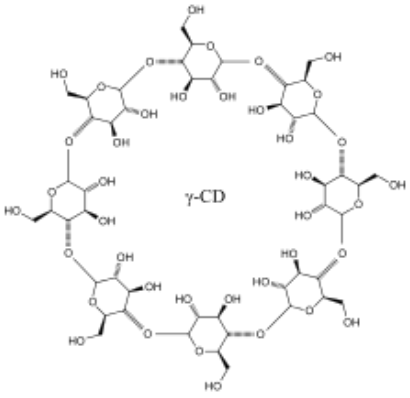

FIGURE 3 - GAMMA-CYCLODEXTRIN

3 Modified Cyclodextrins

Chemically modified CDs derivatives are typically prepared with an objective – to extend physicochemical properties of a certain molecule and inclusion capability. CDs have the characteristic of having a longer time to form stable inclusive complexes with hydrophobic drugs, and then to have prolonged drug release properties. Inserting ionic and non-ionic substituents on CDs can influence their ring adjustment and reduce toxicity. Modified CDs have a higher water solubility, can act as enzyme mimics, and can also be important for molecular recognition (12,13,15–17) .

Efficient synthetic methodologies that can be easily scaled up need to be created to manufacture small molecule CD derivates with high purity and desirable efficiency, to increase CD-containing polymers with controlled topology, structure and molecular weight as well as distribution. Most of CD polymers fail in systemic biocompatibility and toxicological evaluation, with exception of few commercially available products (18).

There are three types of modifications: C-2, C-3 and C-6, where can be added a hydroxyl group – the C-6 modification is the one that produces minor effect on other hydroxyl groups, being mainly used in chromatographic analysis to separatee rigid compounds. Typically, C-2 and C-3 modifications stay directed to the bigger opening, while C-6 modification is directed to the smaller opening of CD (19).

When a specific CD is not capable to perform a certain objective, it is important to modify it and make them accomplish their purpose. After making the substitution, this alteration can have 3 potential isomers, and scientists provided a method to select the isomer that is important to archive the main goal by making all enantiomers pass by a magnetic selection with Fe_3O_4 (iron oxide) nanoparticles. This magnetic selection and immobilization also gives surface diversity linked with an enhanced stability of CD. It was seen that hydrophilic β-CD are easily oxidized and lead to a weak gastrointestinal absorption when complexed with hormones (19).

Around late 1990, only hydroxypropyl and sulfobutylated β-CD had satisfactory toxicological data to act as drug carriers. Also, at this time, β-CD had very low solubility and a high tendency to be nephrotoxic, and some imperative points at that time are seen as impossible such as being inert and not interfering with cholesterol. Scientists verified that when upgrading CDs in their substituents, they had a slower enzymatic metabolism

in comparison with a non-substituted CD. As the number of methyl groups in CD increases, water solubilization also increases at a maximum related to 14 methyl groups. The addition of anionic (carboxymethyl) or cationic (amino, thiolate) groups to methyl substituents leads to a decrease in affinity to cholesterol and lower hemolytic activity (12,13).

Thiolate CD are recent CDs that have the capacity to link with mucus layer, being the best CD to provide local delivery when the principal objective is delivering drugs on gastrointestinal mucosa – there is an improvement on therapeutical efficacy by increasing the time that the drug can stay on specific tissues. This way, modified CDs can also be a great mimic of enzymes, as they can quite act like molecular recognition (15,16).

As β-CD has a lot of inconveniences when used in a non-gastrointestinal route like hemolysis, high cytotoxicity due to membrane disruption though destruction of lipid rafts, irritation and nephrotoxicity, usually it is used a modified β-CD to overcome them, adding pharmaceutical improvement on the new medicine. Although modified β-CD do not demonstrate such disadvantages, some of them are still capable of removing cholesterol from lipid rafts, induce cell unviability and even apoptosis, as M-β-CD and DM-β-CD (20).

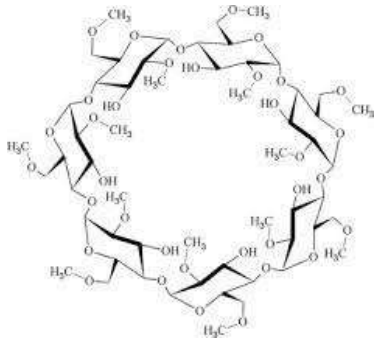

FIGURE 4 - METHYL-BETA-CYCLODEXTRIN

4 Cyclodextrins as Medicine and Therapeutic Adjuvants

4.1 Cyclodextrins and Cholesterol Depletion

CDs are able to complex cholesterol and have thus been evaluated as therapeutic agents for cholesterol depletion in diseases conditions mediated by cholesterol accumulation. CD cholesterol depletion is dependent on the type of CD used, its concentration, incubation time, temperature and cell type. β-CD has been reported to be the most effective CD to "receive" cholesterol since its internal cavity size matches with cholesterol size i.e – cholesterol can enter with no effort inside CD hydrophobic cavity, avoiding aqueous phase. When experimenting cholesterol removal with β-CD of a monolayer membrane, it was seen that β-CD is efficient on cholesterol removal when in presence of β-CD dimers perpendicularly oriented to membrane surface (favored at high concentrations). The great affinity of methylated-β-CD to cholesterol turned obvious making it usable as a cholesterol retention agent to reshape lipid rafts from membrane cells (21–25).

4.1.1 Niemann-Pick Disease type C

Niemann-Pick Disease type C (NPC) is a rare autosomal recessive condition where cells are not able to make intracellular cholesterol transportation, inducing an atypical cholesterol accumulation inside endosomal compartments. The major symptoms are neurodegeneration, lung and liver degeneration, and almost all treatments available for this disease are directed for symptoms, adding the fact that the majority of them are not able to cross blood-brain barrier (26,27).

Due to the ability of hydroxypropyl-β-cyclodextrin (HP-β-CD) to complex cholesterol, it has been evaluated as a candidate for the treatment of NPC and has shown to facilitate life prolongation, lysosomal volume decrease and improved cholesterol trafficking. When compared with miglustat, the most used treatment drug for NPC, HP-β-CD showed better results in decreasing cholesterol levels. Despite the good advantages of HP-β-CD in treating NPC, it is unable to cross brain-blood barrier. Thus, an alternative treatment approach is a direct injection into the brain with a combined treatment of subcutaneous injections. This combination led to a normal brain histology and elimination of neurodegeneration, without interfering with cholesterol homeostasis. There is differences between low and high HP-β-CD doses, both demonstrating therapeutic use, but high doses showed to be toxic for brain cells and changed astrocyte morphology (26–29).

While HP-β-CD is being clinically evaluated to treat this disease, there are some concerns about lung injury and ototoxicity when is used for a long period of time. It was found that 2-HP-β-CD is a larger CD that also changes the abnormal cholesterol homeostasis in NPC, being more toxic than HP-β-CD because of the higher cholesterol attachment. HP-β-CD is capable to transfer cholesterol from limiting lysosome membranes, at an equilibrium that favors efflux of cholesterol. Normally, CD and cholesterol are at 1:1 proportion, but when a 2:1 HP-β-CD:cholesterol was used, 2 CD molecules will encapsulate both cholesterol endings, making a greater solubilization. HP-γ-CD is capable of removing the excess of cholesterol in NPC as HP-γ-CD, with a lower proportion (1:1 proportion), also being a better option when doctors are seeking for homeostasis and biocompatibility – so, HP-γ-CD can be used in a wider therapeutic use than HP-β-CD. Some studies point out a 4000 mg/kg, subcutaneously injected once a week, as a usual HP-β-CD dosage as NPC treatment (30,31).

4.1.2 Atherosclerosis treatment

CDs have been proposed for the treatment of atherosclerosis, as they reduce cholesterol blood levels by increasing cholesterol solubility and facilitates cholesterol removal from cells. This has been observed to lead to a reduction of heart attack, stroke, and peripheral vascular disease. Although statins, fibrate and PCSK-9 inhibitors contributes to a 80% reduction in cholesterol levels, there is still no reduction of atherosclerotic plaques with these drugs, so there is a need for a new drug capable of reducing this plaques (32,33).

A subcutaneous CD injection can reduce atherogenesis and induce regression of established atherosclerosis by increasing cholesterol metabolism and liver X-receptor (LXR)-dependent cellular reprograming, which promotes an effective reverse cholesterol transport as well as reduction in pro-inflammatory gene expression. This injection does not affect plasma cholesterol, weight gain, blood pressure or heart rate, only affects reactive oxygen species production and pro-inflammatory cytokines plasma levels (32).

Treatment with CD for atherosclerotic plaque regression demonstrated to be effective even without lifestyle changes, even though it didn't reduce blood cholesterol levels. CDs demonstrated to be like a pro-drug, not being metabolized itself, but promoting cholesterol metabolism into active pharmacologically metabolites. The CD-mediated atheroprotection could be multifactorial because CD is able to improve cholesterol

solubilization and promote cholesterol metabolism, mechanisms that cannot be isolated (32).

From the three types of CDs, α-CD seems to be the perfect one to improve the remotion of cholesterol from atherosclerotic plaques and reduction of cholesterol crystals, also reducing cholesterol phagocytosis. In one hand, 2-HP-β-CD demonstrated to reduce and prevent new formation of atherosclerotic plaques even with a diet enriched with cholesterol in mices. In the other hand, α-CD demonstrated to lowing proatherogenic lipoproteins levels in mices with the same cholesterol enriched diet. It was seen that oral ingestion of α-CD reduced not only cholesterol levels (including LDL values in hypertriglyceridemia) in serum, but also increases insulin sensitivity in overweight and obese patients with diabetes type II. These high reductions were not seen in healthy population, leading to a conclusion that this CD is more effective in specific population (dyslipidemic and obese population) (33).

As cholesterol is the main "problem" to atherosclerosis, there are some considerations saying that 2-HP-β-CD and α-CD can extract and dissolve cholesterol crystals from inside and outside of the cells. Also, this mechanism is extremely important to reduce cholesterol from macrophages and reducing the formation of foaming cells, maintaining and reducing atherosclerotic plaques and subsequent lesions (25).

TABLE 1 - SUMMARY OF THERAPEUTIC USES OF CYCLODEXTRINS

Indication	Description	References
Cardiovascular Disease	There are being developed clinical trials to use α-CD in cardiovascular disease, as they will connect to dietary fats and, consequently, they will reduce the amount of absorbed fats. This therapy is about taking 1 tablet with 2 mg, needing to be taken 3 times a day, 1 hour before meal.	(34)
Obesity and Dietary Supplement	It is considered that a diet with α-CD can improve fecal cholesterol elimination in a high saturated fat diet, reducing the amount of absorbable fat, LDL, and weight.	(35,36)
Niemann-Pick Disease	All these studies have the objective to provide information that CDs can be an effective treatment for patients having Niemann-Pick disease, as they can extract cholesterol from brain cells and central nervous system, with the maximum tolerable, safe, and effective dose.	(37–43)
Sars-Cov-2 Detection	The objective is to use a mouth rinse with β-CD and an antiviral to prevent Sars-CoV-2 transmission though saliva, directly and/or indirectly. The combination of CDs and antiviral or antimicrobial agents (like chlorhexidine) will have a reduction in Sars-CoV-2 infection and oral microbiota (44).	(45)
Fertilization	Methyl-β-CD shows to be capable to increase sperm fertility after cryopreservation by interfering on cholesterol from lipid rafts. Exists a patent in this regard	(46)
Cancer	CD can be a weapon against cancer due to its affinity to cholesterol, disrupting cell membranes by linking specifically to tumor cells.	(47,48)
Antivirals	CD could be important as antivirals because they can interact with cholesterol present in viral particles, and interact with lipid rafts in targeted cells that are essential for viral particles to attach.	(49,50)
Muscular Relaxant	Sugammadex is becoming an important alternative for a faster surgery recovery in many patients, as this CD promotes a recovery within minutes.	(51–53)
Alzheimer Disease	It is observed that β-CD act as an adjuvant therapeutic in this disease, controlling cholesterol quantity and indirectly interfering in Aβ protein deposition in brain cells.	(54,55)

4.1.3 Cardiovascular Disease

High quantity of LDL is a problem that can lead to a cardiovascular disease, as hypertension, stroke, cardiac insufficiency, especially in diabetic population, younger people with very high LDL values and older people with other risk factors. So, as soon as this problem is identified it need to be reduced and controlled with a pharmacological option, dietary control, and exercise. When we talk about the pharmacological option, it is interesting to think about CDs, specifically α-CD, as a novel soluble dietary fiber, for reducing cholesterol levels (56).

Some advantages that this CD is that it has a higher lipid affinity than others soluble dietary fibers requiring a lower dose to accomplish the same reduction. It is also well tolerated at gastrointestinal lumen causing minimum trouble. Although it mainly has the capability to lower serum cholesterol, it also promotes insulin sensibility when given to type 2 diabetic patients and possibly facilitates some weight reduction. This CD, as it is non-absorbable and hydrolyzed by gut bacteria, can be considered as a pre-biotic. When it was measured all biochemic and lab parameters there was no changes between groups during clinical trials, even with lipophilic vitamins. It is also seen that α-CD perform a better LDL reduction in dyslipidemic and obese or overweight people (56).

4.1.4 Obesity, Fecal Elimination and Dietary Supplement

When α-CD is given to healthy people, it was seen that with a meal with low carbohydrates, glucose response was low while insulin response did not suffer changes. As α-CD has lipid binding properties and glycemic reduction properties, it potentially can be used in type 2 diabetic population, metabolic syndrome, and dyslipidemia. The ingestion of α-CD for 1 month led to a weight reduction in overweight population without changes in food intake, and it was seen that in type 2 diabetic population weight could be maintained. It was also observed that a 5g or 10g of α-CD can reduce post-prandial glucose levels while a 2g dose did not change these levels. Despite these facts, a 2g dose of α-CD taken 3 times a day, without diet or lifestyle changes did reduce body weight, atherogenic lipoproteins and increased insulin sensitivity in overweight population (57,58).

As dietary supplement, the objective is to reduce and control weight and lipid blood levels in overweight but not obese and not diabetic population. The ingestion of α-CD will help

patients to maintain or lose weight, to reduce triglycerides and cholesterol levels, and even increase insulin sensitivity. The reduction of food intake is difficult for many, as we see that junk food is tasty and much more accessible than healthy food, then we can conclude that taking a pill that could reduce fat ingestion is a lot easier. The ingestion of α-CD makes a complexation with fat when in stomach, is resistant to salivary amylases and pancreatic lipases and does not promote an explosive elimination of feces, always associated with lipase inhibitors. α-CD is fermented by intestinal microbiota promoting stool regularity, cholesterol lowering effect and having a prebiotic result. After fermentation, physiological energy taken from α-CD is lower than starch usually taken (57–60).

4.1.5 Infertility

One of the initial steps to capacitate human sperm and enhance fertilization is by reducing its cell membrane cholesterol. Methyl-β-cyclodextrin (M-β-CD) can improve fertilization of sperm by altering cholesterol present on membrane surfaces, changing lipid rafts location and thereafter increase spontaneous acrosome reaction, making it more capable to penetrate oocyte. Also, when sperm is preserved with milk containing casein, this component protects sperm from losing cholesterol, increasing sperm cryo-conservation lifetime. Although this need to reduce cholesterol to capacitate sperm for fertilization, it was seen that sperm is sensible to temperature changes created by cryofreezing, and cholesterol when freeze creates a gel phase with restricted rotation moves. When scientists introduce CDs loaded with cholesterol to sperm, it creates a good proportion of cholesterol when freezing, making possible to achieve a better lifetime after unfreezing, as it provides enough cholesterol to suffer temperature and osmotic pressures instead of sperm cholesterol. This introduction of CD with cholesterol seems to preserve sperm from cryopreservation, but at the same time can be an obstacle to capacitation, and then fertilization (61,62).

When talking about freezing oocytes, the most used method is vitrification, that will freeze at a -196º with liquid nitrogen the wanted oocyte for future fertilization. This method does not create crystals inside oocytes, preserving them as it was “fresh”. The main disadvantage is that vitrification creates damage to membrane cells, due to rapid temperature and osmotic differences, reducing the capacity of fertilization (63).

TABLE 2 - CLINICAL TRIALS

Disease	Clinical Trial Phase	Reference
Cardiovascular Disease	2	(64)
Obesity	Not applicable	(65)
NPC	2 and 3	(66)(67)
NPC - Infants	2	(68)
NPC – Neurological Manifestations	3	(69)
Breast Neoplasm	2	(70)

4.2 Cyclodextrin in Cancer Therapeutics

The basis of this disease is DNA mutations in various tissues. Mutations can be hereditary, caused by environmental changes, metabolic processes or some error on DNA replication. There are various treatments for cancer, like surgery, radiotherapy and chemotherapy, and the choice of one treatment over another is based on patient age, cancer evolution at the moment of decision, patient general health, among others (5,71).

Most of early-staged cancers are removed by surgery, but some of them, because of their aggressive presentation, can only be reduced and treated with chemotherapy, which is the administration of one or more anticancer drugs by intravenous administration to suppress and eradicate tumor growth, delivered in frequent cycles so organism have time to recover from inherent toxic effects. Nevertheless, chemotherapy is associated with many side effects as vomiting, diarrhea, anorexia, hair loss, poor patient compliance – this problems can be prevailed with systemic drug delivery systems as nanoparticles inside intravenous injection (72,73).

The usage of nanoparticles for designing effective delivery systems can reduce the disadvantages of traditional anticancer treatments, making possible to orientate anticancer drugs to affected tissues with success and safety. As they reach tumor cells, drug delivery systems discharge anticancer drugs to prosecute therapeutic effect while toxic effects on non-tumor cells are minimized. Still, this nanoparticles have to face a lot of adverse conditions, like pH and salt conditions, contact with various plasma proteins that can disturb nanoparticles with thermodynamic and kinetic equilibrium changes, so the

knowledge of this barriers, it can be formulated a local drug delivery to reduce the obstacles of systemic drug delivery systems (72,73).

4.2.1 Cyclodextrins as anticancer medicines

The development of original anticancer agents for clinical use is an important step that needs to be done, making oral chemotherapy a possible and desirable option, reducing a lot of costs associated with IV administration and all inherent complications. The oral chemotherapy isn't an actual way of treatment because of anticancer agents low water solubility, high gastrointestinal degradation, low permeability and toxic side effects, even though it is a desirable way when doctors want a prolonged exposure (5,14).

As seen in cardiovascular diseases, CDs ability to extract cholesterol from cell membranes, interfere with lipid rafts and receptors for signaling, cell proliferation and angiogenesis can play a role in inhibiting tumor growth. There are some cancer types that ate more sensible to cholesterol variations, like breast cancer, prostate cancer, and myeloid leukemia, so it is important to think about an anticancer drug thar can interfere with cholesterol levels. HP-β-CD can interact with membrane cholesterol and disrupt cells (leading to apoptosis), being well tolerated among many species with low toxicity accordingly with administration route and dose. M-β-CD also inhibits phosphorylation of a specific kinase (Akt) that prevent apoptosis, leading to a disruption of all normal cell functioning (49,74–77).

In vivo, intraperitoneal injections with M-β-CD with an 800 mg/kg dosage, once a week, suppressed breast cancer with significantly good results. Although M-β-CD has a very good antitumor effect when directly injected at tumor site, when it is an intravenous injection, these results are not as effective perhaps because of the lack of specificity to tumor cells and a great renal clearance. Intravenous injection leads to changes in some values, such as LDH and BUN (urea nitrogen), that can inform if there is any problem at kidney – normally this high values are associated with high hemolysis (77,78).

For instance, M-β-CD has been shown to have cytotoxic activity and an antitumor agent by efflux of cholesterol from plasma membrane by inducing apoptosis. However, they have the disadvantage of low antitumor activity and low tumor selectivity. In many cancer types (brain, kidney, breast, lung, myeloid cells) we see an increase of folate receptors in

membrane tumor cells, that is important for DNA synthesis, so the folate-M-β-CD can be an interesting option to "attack" tumor cells – low interaction with serum proteins and high physiochemical properties. Thus, folate-appended Me-β-CD (FA-M-β-CD) demonstrated a reduction in tumor growth as tumor cells express folate receptors in a high quantity which were used for tumor targeting (75,76).

The FA-M-β-CD is recognized by overexpressed folate receptors at cancer cells, leading to mitophagy-induced cell death instead of apoptosis by upregulating mitochondrial transmembrane potential via mitochondrial folate transporter on mitochondria. Many cancer cells also overexpress a hyaluronic acid ligand on their surfaces, making possible to use HA/M-β-CD to induce apoptotic activity (even though this apoptotic event is lower than the one promoted by M-β-CD) (79).

When in presence of folic acid, FA-M-β-CD was not capable of having antitumor activity, so we can admit that folic acid is a FA-M-β-CD inhibitor. The CD cited also has a beneficial effect on recovery from cancer, as there are no relapses on mices and no body weight loss after intravenous injections. Although FA-M-β-CD is able to reduce cholesterol from membrane cells, like M-β-CD, it don't disturb DNA and mitochondrial content, so we can think that the cell death induction is not related with depletion of cholesterol, but with autophagy by increasing LC3-II expression (autophagosome marker) (75,76).

Cationic β-CD is another class of CD with potential utility in cancer therapeutics. They have shown a great ability to penetrate and accumulate into dense tumor structure, as they interact with the negative charged cell membrane. This cationic CD were shown to be non-toxic to non-tumor cell lines, while having consistent toxicity to tumor cell lines by promoting apoptosis associated with p53 gene (tumor suppression), increasing p53 expression and patients survival (80).

4.2.2 Cyclodextrins as potential Adjuvants in Cancer Therapeutics

CD's ability to complex cholesterol have also been exploited in cancer therapeutics. Some scientists have developed 2DG-ABT combination therapy, molecules that will promote apoptosis in tumor cell lines and block glycolysis, leading to a slow cell growth and cell death. 2-DG-ABT is taken up by cancer cells by glucose transporter, activating several

signal transduction cascades leading to formation of a “death” apotosome complex. The addition of β-CD to reduce membrane cholesterol when co-administered as 2DG-ABT-β-CD, in the right sequence, was found to promote tumor cells apoptosis, causing no significant harm to healthy cells. However this association only caused a significant weight loss in mices that was not regained (81,82).

4.3.3 Sulforadex

This compound was being studied as an anticancer drug directed to reverse breast cancer cells resistance against many anticancer drugs, demonstrating to be successful and currently being on studies as adjuvant in endocrine therapy (83,84).

Sulforaphane is an indirect antioxidant and a natural modulator of phase II xenobiotic-metabolizing enzymes, upregulating factor Nrf2 transcription (antioxidant response) and downregulating factor NF-kB transcription (anti-inflammatory response; when in high concentration is associated with the progress of various diseases, like rheumatoid disease, atherosclerosis, inflammatory bowel disease, multiple sclerosis and tumor initiation and progression). It is seen in many vegetables, principally in broccoli when attacked by insects. Sulforaphane is absorbed very fast, promoting oxidative stress protection in many tissues such as kidney, retinal pigment epithelial cells, pancreatic islet β-cells, skeletal muscle, vascular muscle cells, cardiomyocytes and neurons. Sulforaphane also demonstrates to be effective in cell protection against apoptotic, angiogenic and anti-inflammatory actions. As Sulforaphane is unstable, a combination with α-CD (SFX-01, or Sulforadex) gives it the stability needed to produce its therapeutic use. In cartilages, SFX-01 did not demonstrate any beneficial use neither influencing articular cartilage lesions severity, but in osteoporosis was seen changes in bone architecture, pain reduction and gait improvement SFX-01 can also be used as anti-inflammatory compound in acute respiratory syndromes and inhibiting STAT3 malfunctioning (that conducts to many cancers due to errors during DNA to mRNA transcription) (85–88).

When Sulforadex were given to mice, it passed in toxicological and safety tests, having a good bioavailability, not causing any major side effect associated to a 700mg/day, administrated at a 300mg twice a day. Sulforadex also is associated with preventing oxidative stress occurring in subarachnoid hemorrhage, and in cell stress promoted by inflammation in Multiple Sclerosis (88).

4.3 Modified Cyclodextrins as Antivirals

Viruses are a real problem all around the globe, with many endemic outbreaks and many other pandemic outbreaks with a relatively controlled propagation. Even if viruses are controlled, there are always infections between individuals or even infections though vehicles (like mosquitos) that need treatment and a higher control on these transmissions.

As virus are always mutating, it is very difficult to produce a medicine that will inhibit virus proliferation inside the host. There are many tries in affecting specific targets, like inhibiting adsorption, RNA or DNA replication, and release of virus particles, always leading to a weakened cell or even cell death. Therefore, it is highly recommended to search for a new kind of drugs that can step in virus replication process with the minimum cell harm.

CDs have been shown to possess antiviral activity against HIV, herpes simplex, influenza, RSV and Zika viruses. This antiviral activity has been attributed to the ability of CDs to interacting with cholesterol present on viral membrane, leading to its removal and consequently inhibition of the pathogens ability to bind to cells. The cholesterol removal is associated to lipid rafts interference, resulting in at least 3 mechanisms of action: inhibition of viral entry, inhibition of viral replication and cholesterol sequestration and virucidal activity. Scientists verified that Influenza and Hepatitis B viruses are susceptible to viral membrane cholesterol depletion, while leukemia and Ebola viruses are susceptible to cellular membrane cholesterol depletion (49,50).

Methyl-CD and sulphate derivates are one of the most used CD for this goal, while β-CD are also used for this purpose, their hemolytic properties and renal toxicity limits their application. As such, the HP-β-CD is preferred against viruses as it has no restriction in its usage. Then *in vitro* testing, they show a good sustained antiviral activity, but when *in vivo* tests, they resist to show sustained results against viral exposure (89–91).

Biocompatible sulfonated CD shown broad-spectrum virucidal, irreversible mechanism of action, biocompatible, large impediment to viral resistance and being only virustatic. It has been demonstrated that they have both preventive and therapeutic activities, thus CD are a strong weapon to struggle against several viral infections. Any viral particle that

has exogenous cholesterol can be a target to CD, and this ability has been proven against many common viruses, such as HSV-1, HMPV, and PIV 3 (92–95).

4.3.1 HIV Management

HIV sexual transmission occurs at the mucosal surface and female genital tract, being the primary route of heterosexual transmission, and rectal transmission is also a general problem because of its susceptible physiology to infection. HIV-1 can link itself selectively with cell areas that have a high lipid rafts proportion. This lipid rafts are considered essential to many functions, such as vesicle transport, endocytosis, T cell receptor signaling and apoptosis. HIV infection is also characterized by an elevated atherogenic risk and cardiovascular disease, that can be associated with cholesterol uptake, synthesis and efflux changes (96–99).

For anti-HIV drugs to be efficient is required to have both intra and extracellular adequate distribution during sufficient time of action at target sites. Drug delivery systems can distribute antiretrovirals to subepithelial layers, as well as penetrate target cells to release the drug, increasing the eradication of the virus from infected cells. This type of system is recommended as they improve antiretrovirals efficiency by delivering high drug concentrations at the site of action – they can cross many barriers that stop antiretrovirals to pass, like blood-brain barrier and blood-cerebrospinal fluid barrier. CD complexes are capable of increasing solubility of lipophilic antiretroviral drugs, as protection from external degradation (100).

In this chronic disease there is the problem of drug resistance, as HIV can overcome antiviral properties of many existent drugs. This problem led to the investigation of sulphated CDs, as they inhibited HIV surface absorption *in vitro* tests. The only disadvantage of sulphated CDs is that they have a low intestine absorption and a high metabolization when in the blood – it has a half-time of 4 hours, not being interesting for clinical oral route (101).

Some drug delivery systems like CDs have the power to disrupt HIV lipidic envelope and to synergically act with antivirals, as they are capable to destroy lipid membrane and disturb RNA stability. All these changes will lead to a decrease on CD4 cells infection. It is known that M-β-CD and HP-β-CD have a higher cytotoxic effect, and when

formulating drugs, it's always important to balance therapeutic effect and possible/potential side or toxic effect. M-β-CD have shown a great cholesterol caption even when encapsulating anti-HIV medicines, and could be a path to reduction of antiretroviral drug dose (102).

For all this considerations, 2-HP-β-CD can be a potential anti-HIV drug to be used in chronic disease, also demonstrating to reduce inflammatory response of immune cells when cultured, low production of IL-10 and cytokines, independent from raft disruption. 2-HP-β-CD can remove some cholesterol of the membrane from host cells (making them less susceptible to viral infection without cell disruption), but also causes cholesterol removal from viral particles leading to disruption of HIV integrity. Methyl-β-CD lead to a reduction of virion-associated cholesterol and almost a serious elimination of virus infectivity by blocking fusion to target cells. 2-HP-CD demonstrated to reduce cholesterol on viral particles that were produced from treated cells (97–99,103).

When β-CD was applied in mice vaginal mucosa it prevented around 90% virus transmission, but when transported to a monkey specie similar to humans, the same vaginal application didn't show relevant outcomes. It was also seen that β-CD only confer protection on the first application and contact with HIV, not being effective on second contact and conducting to a normal HIV infection (89).

4.3.2 Influenza

As Influenza virus particles are mainly constituted by cholesterol, they are an easy target to β-CD, so it will create holes all around virus membrane, disrupting it and dropping viral charge. Influenza is one of the deadliest viruses in circulation, being controlled by vaccination, but there is a lack in therapeutics when we are talking about hospitalization. One of the most recent therapeutic research is focused in a sulfonated β-CD – this CD showed to be effective against an Influenza infection *in vitro* and *in vivo*, mostly at late stages by acting on virus replication phase. Sulfonated β-CD have a great virucidal activity, causing almost total loss of infectivity of Influenza virus. A study also showed that a intranasal treatment with this CD reduced deaths around two thirds, even with a reduced dose of sulfonated β-CD (89,104).

4.3.3 Dengue

Dengue is a viral disease transmitted by mosquitos, mostly seen in tropical and subtropical countries, being a serious disease and life-threatening due to hemorrhagic fever and shock syndrome. Infection prevention of Dengue is controversial – the virus incubation with CD lead to a low infectivity, but when CD is given to cells as a pre-treatment it does not have any result as a preventive treatment. Thus, as there is no bond between cell cholesterol membrane and the first step of virus entry, it can be assumed that CDs are not able to be a prophylaxis treatment. CD are capable to reduce infection by removing viral cholesterol, leading to a rigid virus envelope that could not bind to cell membrane and initiate infection. Therefore, virus particles previously treated with CDs cannot have a normal infectivity, and then can't have their RNA and protein synthesis and expression (50).

β-CD can be used in infected patients to control virus replication and to prevent further cell infections, but this can be risky as β-CD also has a strong connection to membrane cholesterol, leading to cell and renal toxicity. Another way to control Dengue transmission is to introduce β-CD in mosquitos (initial transmission stage), as it was seen that it interferes with a non-structural protein 1 (NS1) (essential to viral replication and excretion), but this approach seem to be non-effective as it doesn't produce a needed viral particles reduction (89).

4.3.4 Herpes Simplex Virus

When HP-β-CD was applied in patients with relapses during the last half year, it was seen that CD reduced relapses of HSV for 6 months. Scientists also discovered that a new CD, sulfoundecylthiomether-β-CD, is active in both HSV-1 and HSV-2 and is time-dependent – within 5 minutes reduces viral amount, and within 15 minutes inactivates HSV, so we can assume that this new CD is virucidal (89).

4.3.5 Ebola

Ebola virus is a very problematic virus, causing lethal hemorrhagic fever and around 90% of deaths. Until the last research for this monography, there was no available treatment for this virus, neither vaccine nor a medicine, being an endemic virus in North Africa.

Like other viruses, Ebola virus has to entry into a cell to initiate all process of infectivity, having a strong interference from lipid rafts present on cell membrane surfaces. Previous Ebola infections “leave” signaling proteins in lipid rafts to facilitate new infections from new virus particles. This way, enriched and non-enriched lipid rafts can be the therapeutic solution for Ebola treatment, using CDs as medicine (105).

4.3.6 Hepatitis B Virus

One of the most important steps for a viral infection is the first step of entry in targeted cell, being important the amount of cholesterol present on the cell membrane for this infection. Although the pointed before, many viruses are not as dependent of membrane cholesterol as others (as influenza virus) (106).

In the case of HBV, the most important cholesterol for first step of infection is cholesterol from virion. When M-β-CD is introduced in human plasma, it led to a cholesterol reduction in virion particles, consequently promoting irreversible inactivation of the virus. M-β-CD only demonstrated to reduce the size of HBV, not disintegration of viral particle. This partially disintegration may be a consequence of morphology of the HBV as it has a highly dense packed protein all cross linked with very low amount of lipids. When hepatocyte membranes are treated with M-β-CD, it is seen a lower infection of HBV, not needing lipid rafts or domains enriched with cholesterol (106).

4.3.6 Sars-Cov-2

So, as we previous have seen, we know that CDs can change membrane cells, having a big impact on lipid rafts. These lipid rafts in most cases are very important in many cell functions as cancer treatment, and they are also important to help the entry of viruses inside the cell. Therefore, we can assume that if CD capture cholesterol from cell membrane, the virus entry can be compromised. Also, when CD are continuously used, they can change how ACE2 receptors are distributed along the membrane, making Sars-Cov-2 entry impossible. Some scientists believe that if CD are used with some oral rinses can reduce the number of viruses present along mouth, throat and nose, and this way an infected person does not expel contaminated particles to the air and other person. We can use CD molecules to capture cholesterol from Sars-Cov-2 particles and disrupt them, this way viral particles are not able to “survive” and infect cells (107,108).

4.4 Other Cyclodextrin Therapy Indications

4.4.1 Muscular Relaxant

Sugammadex (modified γ-CD) has the therapeutical objective of selectively encapsulate steroidal muscular blockers such as rocuronium or vecuronium (muscular relaxant mainly used in general surgeries and emergency operations), therefore it returns the excitation capacity of neurotransmitters. It is given to patients after a surgery that needs to go under general anesthesia and makes possible a faster recovery when compared with neostigmine, and the dose given to the patient depends on how much rocuronium was administered. The usual intravenous dose is 2.0 mg/kg, and recovery is not interfered by maintaining anesthesia. If it is given a low dosage of Sugammadex to patient, there will not be enough CD to encapsulate rocuronium, so the patient is at a serious risk of having postoperative pulmonary complications. Sugammadex has no interference in any biological process, such as metabolization, linking to plasma proteins, blood-brain barrier and placenta translocation, and doesn´t have any interference in reproductive processes. It is also well tolerated in pediatrics and elder population, but in obese population it is recommended to use TBW to estimate the right Sugammadex dose, instead of using IBW, although when calculating the dose of rocuronium it is used IBW. When dealing with patients with renal failure, there has to be caution, as Sugammadex and Sugammadex-Rocuronium are eliminated by kidneys – these complexes can be eliminated by hemodialysis (51,52,109).

The amount of Sugammadex used can change with the objective: 2 mg/kg is used to reverse a moderate neuromuscular blockage, 4 mg/kg is used when patient has a deep blockage, and 16 mg/kg is used to reverse immediately rocuronium. As this CD gives the opportunity of a faster recovery, doctors observe their patients starting to move and cough after Sugammadex administration. Some of the most side effects of Sugammadex are nausea, vomiting, headache and itching. There is also an anaphylactic risk associated with high dosage, but not related to repeated administration. It is considered that when Sugammadex is given to women taking birth control, Sugammadex creates the same effect as these women missed one birth control dose, as it has a high affinity to progesterone (53).

4.4.2 Alzheimer Disease

Although there is not an effective treatment for controlling and "block" Alzheimer Disease, it is still important to prevent it from a continuous progression. It is observed that cholesterol is an important molecule to produce Aβ protein, also being seen a higher cholesterol quantity in AD. The administration of HP-β-CD in these patients can be useful to reduce cholesterol outside CNS, specifically outside brain, control Aβ protein production and may reduce AD incidence. It was also seen that HP-β-CD has a positive impact on ABCA1 gene (gene that code information about cholesterol efflux regulatory protein), increasing ABCA1 expression and consequently reducing Aβ deposition. The way HP-β-CD act on cholesterol is different from the way statins act, when only seeing AD, so HP-β-CD can be an adjuvant in AD treatment. It was stated that when CD was a vehicle for many drugs (as testosterone, estradiol and morphine) it had a great passage through BBB and a good nervous cell targeting. Also, when HP-β-CD is subcutaneous administered in mice, it shows some brain improvements such as spatial learning and memory deficit, also eliminating Aβ plaque deposition in a specific concentration (Aβ:HP-β-CD molar ratio of 1:2). HP-β-CD showed a preference to block hydrophobic residues of Aβ, inhibiting elongation (54,55).

CDs can be administrated in 500-3000 mg/kg via parental administration, or 100-750 mg directly in CNS, and these amounts can be given to patients weekly, twice a month or once a month, accordingly with the effective necessary amount of CD (110,111).

4.4.3 Dermatological Disorders

Substituted CDs like HP-β-CD are important vehicles for some medicine used in skin conditions as they can form complexation units like micelles. There are also toxicological considerations about CD when in contact with epidermis, but there is some studies that show that natural CD are well tolerated as they not disrupt stratum corneum. Some other studies showed HP-β-CD improve permeability in epidermis, most likely because of remotion of cholesterol. Usually, CDs are used as skin penetration enhancers and due to their big weight they do not disrupt skin barrier system, but some studies pointed out CD can disrupt stratum corneum (by extracting lipid components) and facilitate the entrance of medicine though disrupted skin (112).

Modified and non-modified CDs can encapsulate gas and many compounds related to bad odor in population and are also capable to be a facial exfoliating component. When HP-β-CD is associated with salicylic acid it is improved salicylic acid properties of disinfectant and bacteriostatic. CDs are capable to form a complex with insulin (preventing insulin degradation), and this complex can be used to heal pressure ulcers and promote re-epithelization (112).

5 Toxicity

Around 1950 decade, scientists thought that β-CD ingestion was toxic because led to rats death, later it was discovered that the cause of death was not CDs but impurities relative to production process. Studies performed about CD toxicity demonstrated that they have few toxicological concerns, derived from low amount of absorption in gastrointestinal tract. As β-CD have a 10% amount of absorption, only a small amount can be incorporated in formulations, not being suitable for parental formulations due to toxicological concerns (low water solubility) – for parental formulations it is advisable to use α-CD (113).

The intravenous LD_{50} of α-CD in mice is 750-1000 mg/kg , while in rats this LD_{50} is 500-750 mg/kg. Intraperitoneal injections have limited toxicity, showing tolerance until a dosage of 2086.5 mg/kg for 5 consecutive days, during 13 weeks in mice, and the maximum tolerated dose is 10000 mg/kg. When α-CD is given to dogs within their diet, it is seen that some of them have transient diarrhea (normal situation when poor digestible but fermentable carbohydrates are ingested), and urinary pH was lower in males when in comparison to control group. There is a α-CD LD_{50} value for humans that is around 12.5 g/kg, but this value is not verified, as it does not have an official source of information. There are some differences between α-CD and β-CD: the first one is that β-CD demonstrate to be toxic to liver and kidneys (in contrast, α-CD did not show any toxicity to the organism), the second difference is that α-CD does not perfectly incorporate cholesterol in its cavity (59,60,78).

From the three main types of CDs, it is observed that β-CD is the most toxic to cells, leading to hemolysis, disruption of membrane cells though removing lipid rafts, and they can be nephrotoxic due to interfering with lipids from tubular cells. This indicates that

reabsorption and secretion processes can be compromised, leading to a critical situation for the patient (20).

When we are talking about substituted CDs, it is seen a higher toxicity among CDs that have more substituents (translated in a higher hemolytic effect) and the position of the substituents is not important for this toxicity. The toxicity associated to substituted CDs can be related to cholesterol removal and the higher affinity for cholesterol than non-substituted CDs. It is also seen that concentrations of methyl-β-CD above 5% are toxic to many tissues, such as corneal, nasal and digestive epithelium (13,114).

In humans, when is given orally 4 to 8 grams of HP-β-CD for 2 weeks there is no alteration seen in volunteers, while higher doses of 16 to 24 grams for 2 weeks were associated to soft stools and diarrhea. It is also referred that there are differences between oral and intravenous route, as the second route has a higher toxicological probability – HP-β-CD affects kidney, lungs, liver, spleen and urinary tract. As in kidney is a lack of cholesterol, when HP-β-CD complexed with cholesterol arrives, this complexation is dissociated, leading to an increase of HP-β-CD and cholesterol in kidney – these two compounds, mostly cholesterol, are able to produce alterations in kidney structure, being reversible and do not cause renal function impairment (115).

A study demonstrated hepatic injury and CNS dysfunction in two patients with NPC disease. Also, chronic HP-β-CD administration led to lung toxicity and pneumonia in healthy pigs. Mice that had wild type gene and heterozygotic gene for NPC disease showed several organs injury (lungs, kidney and liver). About homozygotic NPC gene, it was seen that these mice were resistant to multiple organs injury induced by HP-β-CD. The sublethal dose of HP-β-CD is 20000 mg/kg in mice, through subcutaneous injection, corresponding to a 50% lethal dose in wild type (healthy population) gene and 80% lethal dose in heterozygotic gene, that result in hepatic necrosis, vacuolization of renal tubular epithelium, serum creatinine level increasing, pulmonal hemorrhage and inflammation 8 hours of injection. All this cellular injury seems to be related to a lower cholesterol quantity present in cellular membranes, as HP-β-CD is able to extract it. HP-γ-CD is as capable as HP-β-CD in removing cholesterol in NPC disease, in a lower proportion, so it is less harmful (no ototoxicity and no lung injury) for patients when they are under a prolonged treatment (30,31).

Conclusions

Using CDs only for drug encapsulation and drug delivery is reductive even if CDs are very useful for this purpose. So, CDs are molecules endowed with many possible treatments based in one fundament: being able to encapsulate cholesterol from various sites, such as cell membranes.

Based on the cholesterol remotion mechanism, CDs can be a new therapeutic option in atherosclerosis, cardiovascular diseases associated with high cholesterol levels, Niemann-Pick Disease, in sequestering cholesterol in gastrointestinal tube, possible control of viral infections and even act in cancer. These diseases and specific therapeutic targets were being studied during last years and are now well known in medical field, but there is some lack in finding new medicines due to the difficulty to create therapeutic molecules that bind specifically to the target. Not only conventional CDs can be used as a therapeutic option but also modified CDs, as some of these CDs can create a disruption in targeted cells, viruses or in a specific target, and this way, these molecules are able to control or even reduce an identified disease.

CDs and modified CDs acting as medicine have space to grow in diverse areas, like caring cholesterol to cells in cholesterol deficit, restoring cell damage from a necrotic area and many more specific cases. In this perspective, CDs could be a brand-new therapeutic treatment or even a therapeutic adjuvant in various therapeutic schemes, controlling and even reduce a certain disease that without a CD could be life threatening.

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