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Faculdade de Farmácia

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Margarida Figueira Serrano Caldeira Barradas

Monografia orientada pelo
Professor Doutor Igor Locatelli, Prof. Associado na Faculdade de Farmácia da Universidade de Liubliana e coorientada pelo Professor Doutor Luís Pleno de Gouveia, Prof. Associado na Faculdade de Farmácia da Universidade de Lisboa

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

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2024

Declaro ter desenvolvido e elaborado o presente trabalho em consonância com o Código de Conduta e de Boas Práticas da Universidade de Lisboa. Mais concretamente, afirmo não ter incorrido em qualquer das variedades de fraude académica, que aqui declaro conhecer, e que atendi à exigida referenciação de frases, extratos, imagens e outras formas de trabalho intelectual, assumindo na íntegra as responsabilidades da autoria.

Resumo

O sildenafil é um medicamento utilizado na gestão e no tratamento da disfunção erétil e da hipertensão arterial pulmonar, sendo ocasionalmente utilizado *off-label* para o tratamento do fenómeno de *Raynaud* secundário, da perturbação da excitação sexual feminina e como adjuvante no tratamento da hipoxemia induzida pela altitude. Faz parte da classe de medicamentos inibidores da fosfodiesterase-5 (PDE5-I) e a *Food and Drug Administration* (FDA) aprovou o sildenafil como o primeiro PDE5-I em 1998. (1)

O mecanismo de ação do sildenafil envolve a sua capacidade de inibir a enzima PDE5, que é responsável pela decomposição do monofosfato de guanosina cíclico (GMPc), um mensageiro químico envolvido no relaxamento do músculo liso. Durante a estimulação sexual, o óxido nítrico é libertado, levando a um aumento dos níveis de GMPc no corpo cavernoso do pénis. Isto provoca o relaxamento do músculo liso, permitindo o aumento do fluxo sanguíneo para os tecidos do pénis, resultando numa ereção. Ao inibir a PDE5, o sildenafil aumenta os efeitos do GMPc, promovendo uma vasodilatação sustentada e um melhor fluxo sanguíneo para o pénis. No entanto, é importante notar que o sildenafil por si só não causa uma ereção; a estimulação sexual continua a ser necessária para iniciar a libertação de óxido nítrico. Além disso, o sildenafil tem algumas aplicações terapêuticas no tratamento da hipertensão arterial pulmonar, uma doença caracterizada por pressão arterial elevada nas artérias dos pulmões. Em resumo, o sildenafil atua inibindo a PDE5, aumentando os níveis de GMPc, promovendo o relaxamento do músculo liso e facilitando a ereção do pénis. (2)

O objetivo da revisão sistemática integrada na meta-análise foi comparar os parâmetros farmacocinéticos do sildenafil em humanos. A estratégia de pesquisa desta revisão sistemática foi realizada nas seguintes bases de dados electrónicas: PubMed, Web of Science e Clinical Trials Registry.

Esta revisão sistemática incluiu vinte e quatro estudos que incluíram estudos de farmacocinética populacional, modelação farmacocinética de base fisiológica e outras análises farmacocinéticas. Estes vinte e quatro estudos foram sujeitos a uma avaliação, tendo sido eleitos através de um fluxograma associado a revisões sistemáticas.

Palavras-chave: Hipertensão, Farmacocinética, Sildenafil

Abstract

Sildenafil is a medication used in the management and treatment of erectile dysfunction and pulmonary arterial hypertension, it is occasionally used off-label for the treatment of secondary Raynaud phenomenon, female sexual arousal disorder, and as an adjunct in the treatment of altitude-induced hypoxemia. It is in the phosphodiesterase-5 inhibitor (PDE5-I) class of medications and the Food and Drug Administration (FDA) approved sildenafil as the first PDE5-I in 1998. (1)

The mechanism of action of sildenafil involves its ability to inhibit the enzyme PDE5, which is responsible for breaking down cyclic guanosine monophosphate (cGMP), a chemical messenger involved in smooth muscle relaxation. During sexual stimulation, nitric oxide is released, leading to increased levels of cGMP in the corpus cavernosum of the penis. This causes smooth muscle relaxation, allowing increased blood flow into the penile tissues, resulting in an erection. By inhibiting PDE5, sildenafil enhances the effects of cGMP, promoting sustained vasodilation and improved blood flow to the penis. However, it is important to note that sildenafil alone does not cause an erection; sexual stimulation is still necessary to initiate the release of nitric oxide. Additionally, sildenafil has been found to have some therapeutic applications in the treatment of pulmonary arterial hypertension), a condition characterised by high blood pressure in the arteries of the lungs. In summary, sildenafil works by inhibiting PDE5, increasing cGMP levels, promoting smooth muscle relaxation, and facilitating penile erection. (2)

The aim of the systematic review integrated in the meta-analysis was to compare the pharmacokinetic parameters of sildenafil in humans. The search strategy in this systematic review was performed in the following electronic databases: PubMed, Web of Science and Clinical Trials Registry.

This systematic review included twenty-four studies, which comprised population pharmacokinetic studies, physiologically-based pharmacokinetic modelling, and other pharmacokinetic analyses. These twenty-four studies were subjected to an evaluation and were selected through a flowchart associated with systematic reviews.

Keywords: Hypertension, Pharmacokinetics, Sildenafil

Abbreviations

ADME - Absorption, Distribution, Metabolism, and Excretion
AST - Aspartate Transaminase
AUC - Area Under the Curve
BPD - Bronchopulmonary Dysplasia
CDH - Congenital Diaphragmatic Hernia
CHD - Congenital Heart Disease
cGMP - Cyclic Guanosine Monophosphate
CL - Clearance
CL/F - Apparent oral clearance
ClinPK - Clinical Pharmacokinetics
Cmax - Maximum Concentration
FDA - Food and Drug Administration
IV - Intravenous
NA - Not Applicable
NONMEM - Nonlinear Mixed-Effect Modeling (software)
PBPK - Physiologically-Based Pharmacokinetic Model
PDE5-I - Phosphodiesterase-5 Inhibitor
PD - Pharmacodynamics
PK - Pharmacokinetics
PK-Sim® - Physiologically-based pharmacokinetic simulation software (PK-Sim)
popPK - Population Pharmacokinetics
PPHN - Persistent Pulmonary Hypertension of the Newborn
Q - Inter-compartmental Clearance
tmax - Time to Maximum Concentration
V1, V2 - Volume of Distribution in Compartments 1 and 2 (used in multi-compartment modeling)
Vd - Volume of Distribution

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1. Introduction

Pharmacokinetics (PK) studies how the body interacts with administered substances for the entire duration of exposure (3). Sildenafil is a drug substance used in the management and treatment of erectile dysfunction and pulmonary arterial hypertension. It is in the phosphodiesterase-5 inhibitor (PDE5-I) class of medications. The pharmacokinetics of sildenafil have garnered significant attention since its introduction for their treatments. A thorough understanding of sildenafil's absorption, distribution, metabolism, and excretion (ADME) is essential for maximising its therapeutic effectiveness and reducing the risk of potential side effects. Despite numerous individual studies investigating various aspects of sildenafil pharmacokinetics, there is a need for a comprehensive synthesis of these findings to provide a clear and consolidated understanding of its pharmacokinetic profile in humans.

2. Basic concepts of Pharmacokinetics

2.1. Absorption (4)

Absorption is the process that brings a drug from the administration, into the systemic circulation. Absorption affects the speed and concentration at which a drug may arrive at its desired location of effect.

2.2. Distribution (4)

Distribution describes how a substance is spread throughout the body. This varies based on the biochemical properties of the drug as well as the physiology of the individual taking that medication.

2.3. Metabolism (4)

Metabolism is the processing of the drug by the body into subsequent compounds. This is often used to convert the drug into more water-soluble substances that will progress to renal clearance or, in the case of prodrug administration, metabolism may be required to convert the drug into active metabolites.

2.4. Excretion (4)

Excretion is the process by which the drug is eliminated from the body. The kidneys most commonly conduct excretion, but for certain drugs, it may be via the lungs, skin, or gastrointestinal tract.

2.5. Volume of distribution (4)

This metric is a common method of describing the dissemination of a drug. The volume of distribution is defined as the amount of drug in the body divided by the plasma drug concentration.

2.6. Clearance (4)

Clearance is an essential term when examining elimination; it is defined as the ratio of a drug's elimination rate to the plasma drug concentration. This is influenced by the drug and the patient's blood flow and organ status.

2.7. One compartment model (5)

A one-compartment model assumes that all the tissues in the body are contained in a single compartment called the “central compartment”. The drawback of this type of model is that, by its construction, the drug is assumed to be equally distributed throughout the whole body instantly, which is rarely true. For drugs that do not distribute throughout the body, the simplification that this type of model provides may make it the most appropriate option.

2.8. Two compartment model (5)

Two-compartment models account for the distribution parameter that a one-compartment model cannot by dividing the single compartment into two separate containers called the “central” and “peripheral” compartments. The central compartment represents plasma and highly perfused tissues, including the kidneys and the liver. The drug will only leave the body through the central compartment. The peripheral compartment represents poorly perfused tissues such as muscle. In this model, the drug is not assumed to be evenly distributed throughout the body but disbursed between these two different compartments.

2.9. Non Compartment Analysis (5)

In a Non Compartment Analysis, pharmacokinetic parameters are estimated directly from the data.

2.10. Covariate (6)

Covariates are used to describe predictable sources of variability. A useful covariate is expected to explain some of overall variability and should lead to a decrease in unpredictable variability.

2.11. Inter compartment clearance (7)

Inter-compartment clearance refers to the rate at which a drug moves between different compartments within the body in pharmacokinetic models. These compartments represent distinct areas or tissues where the drug is distributed, such as blood, organs, or other tissues.

2.12. Inter individual variability (8)

Inter-individual variability refers to the differences in drug response or pharmacokinetic parameters observed between individuals within a population. This variability is attributed to

factors such as genetic differences, age, weight, gender, health status, organ function, and environmental influences.

3. Methods

3.1. Create search profiles

On the 22nd and 24th of January 2024, comprehensive searches were conducted across the PubMed, Web of Science, and Clinical Trials Registry databases to investigate the pharmacokinetics of sildenafil. The search employed a combination of the terms "pharmacokinetics" and "sildenafil." Restrictions were applied concerning language and species to refine the results. The detailed search strategy is provided in the Supplementary Information (9).

Supplementary Information 1. Detailed search strategies
Pubmed Search date (22nd January of 2024)

Search: sildenafil AND pharmacokinetics Filters: Humans, English, Portuguese Sort by: Most Recent
(("sildenafil citrate"[MeSH Terms] OR ("sildenafil"[All Fields] AND "citrate"[All Fields]) OR "sildenafil citrate"[All Fields] OR "sildenafil"[All Fields] OR "sildenafil s"[All Fields]) AND ("pharmacokinetic"[All Fields] OR "pharmacokinetical"[All Fields] OR "pharmacokinetically"[All Fields] OR "pharmacokinetics"[MeSH Subheading] OR "pharmacokinetics"[All Fields] OR "pharmacokinetics"[MeSH Terms])) AND ((humans[Filter]) AND (english[Filter] OR portuguese[Filter]))
Translations
sildenafil: "sildenafil citrate"[MeSH Terms] OR ("sildenafil"[All Fields] AND "citrate"[All Fields]) OR "sildenafil citrate"[All Fields] OR "sildenafil"[All Fields] OR "sildenafil's"[All Fields]
pharmacokinetics: "pharmacokinetic"[All Fields] OR "pharmacokinetical"[All Fields] OR "pharmacokinetically"[All Fields] OR "pharmacokinetics"[Subheading] OR "pharmacokinetics"[All Fields] OR "pharmacokinetics"[MeSH Terms]

Web of Science Search date (24th January of 2024)

ALL=(sildenafil) AND ALL=(pharmacokinetics) AND ALL =("human" or "women" or "men" or "adult" or "adults" or "patient" or "infant" or "infants"or "newborn" or "newborns" or "humans" or "subject" or "subjects")

Clinical Trials registry (24th January of 2024)

Condition/disease
Sildenafil

Other terms
Pharmacokinetics

3.2. Data selection

This review primarily encompassed studies involving paediatric populations, including neonates (0–28 days), infants (29 days–1 year), and children (>1–12 years). Additionally, studies involving adult participants were also included.

Studies were selected based on the following inclusion criteria: original research articles, reviews, and studies published in the English language. The selected articles were categorised according to the following themes: bioequivalence, drug interactions, the influence of sildenafil on other medications, pharmacodynamics, and review articles.

Exclusion criteria comprised ex vivo studies, in vitro studies, animal research, and studies not directly related to sildenafil. Abstracts, conference proceedings, and corrigenda were also excluded. Duplicate articles were removed to avoid redundancy, as several studies were retrieved from both PubMed and Web of Science databases. Clinical trials registered in databases were included when indexed in PubMed or Web of Science

3.3. Data extraction and synthesis

The search commenced with the databases PubMed and Web of Science, utilising the previously specified terms. The resulting articles that met the search criteria were documented in an Excel format. In total, three distinct Excel workbooks were created, one for each platform: Web of Science, PubMed, and Clinical Trials Registry.

The articles were organised by publication date, commencing with the most recent and progressing to the oldest. They were further categorised according to the predefined classifications, following an examination of the titles and abstracts.

Duplicate entries were removed using the VLOOKUP function in Excel, specifically eliminating any PubMed reference numbers (PMID) that appeared more than once. Additionally, the Mendeley platform was employed as a bibliographic reference manager to facilitate the organisation of citations.

4. Results

4.1.1. Data processing according with the Prisma Flow-chart

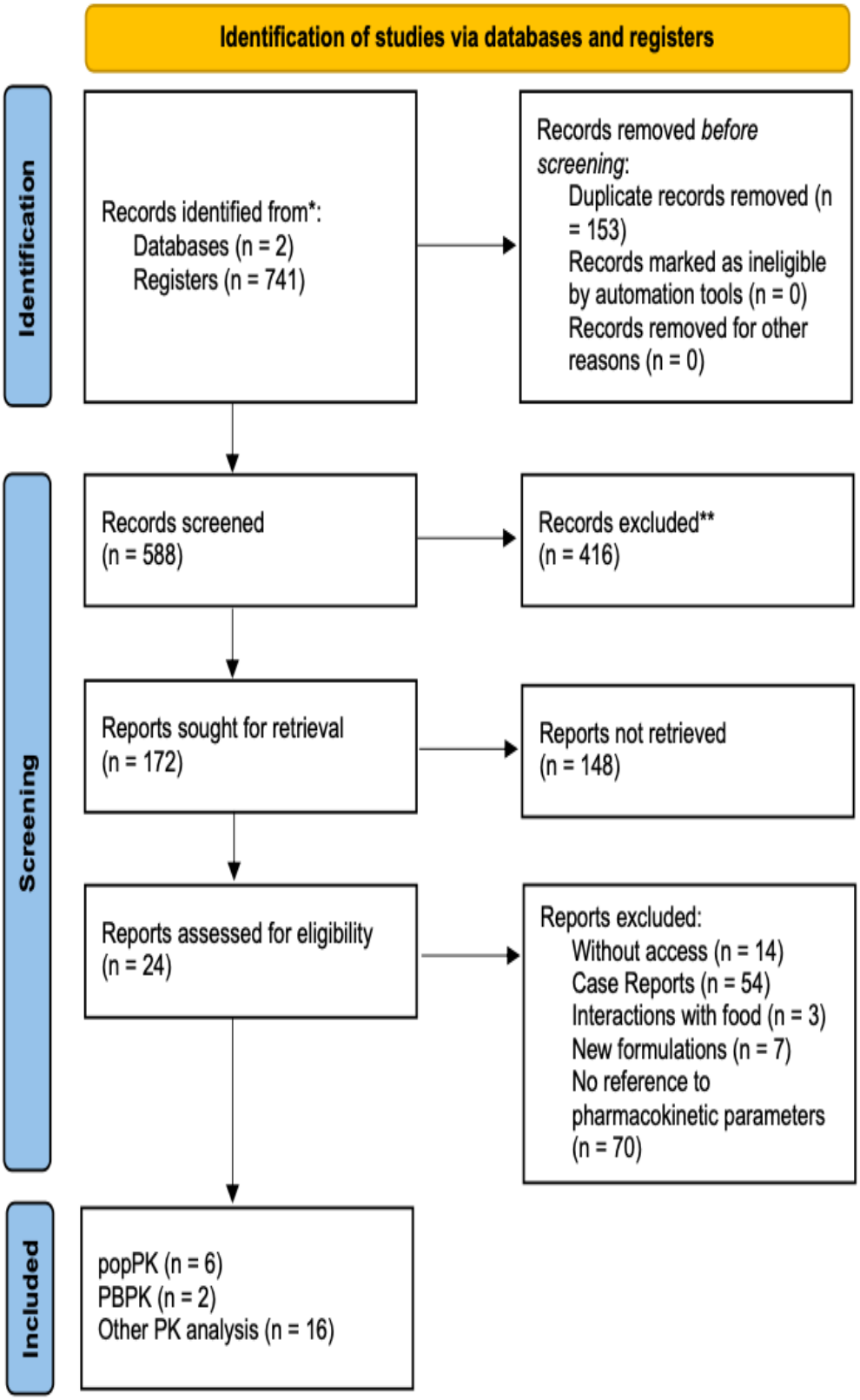

Figure 1: Prisma Flow Chart

4.1.2. Compliance with the PK Checklist

Each of the studies was meticulously evaluated and included in this systematic review according to the ClinPK checklist (10), a 24-item tool developed by Kanji et al. 2015 to ensure transparent and consistent reporting of clinical pharmacokinetic studies. Following a thorough analysis, it was determined that the primary topics pertaining to both the methodology and results evaluation would be included, resulting in a total of fourteen parameters. These parameters were specifically selected for their relevance to the pharmacokinetic aspects of the studies; those pertaining to the structural elements of the articles were excluded.

The checklist encompassed essential criteria such as the eligibility criteria for study participants, co-administration of study drugs with potentially interacting substances, validation of quantitative bioanalytical methods, pharmacokinetic modelling approaches, and the reporting of results using appropriate statistical measures.

Table 1: Clinical Pharmacokinetic checklist according to Kanji et al. 2015 (10)

		Cochius- den Otter, 2019 (11)	Milligan, 2002 (12)	Salerno, 2021 (13)	Rhee, 2022 (14)	Gonzalez, 2019 (15)	Mukherjee, 2008 (16)	Ghonem, 2020 (17)	Ahsman, 2009 (18)
Methods	Eligibility criteria of study participants is described	✓	✓	x	✓	✓	✓	✓	NA
	Co-administration (or lack thereof) of study drug(s) with other potentially interacting drugs or food within this study is described	✓	✓	✓	✓	✓	x	✓	✓
	Drug preparation and administration characteristics including dose, route, formulation, infusion duration (if applicable) and frequency are described	✓	✓	✓	✓	✓	✓	✓	✓
	Body fluid or tissue sampling (timing, frequency and storage) for quantitative drug measurement is described	✓	✓	✓	x	✓	✓	✓	✓
	Validation of quantitative bioanalytical methods used in the study is described in detail or described briefly and referenced	✓	x	✓	x	✓	x	x	✓
	Pharmacokinetic modelling methods, observed and derived parameters along with the formulas, and software used are described	✓	✓	✓	✓	✓	x	x	✓
	Formulas for calculated variables (such as creatinine clearance, body surface area, AUC, and adjusted body weight) are provided or referenced	✓	NA	NA	✓	NA	NA	NA	✓
	The specific body weight used in drug dosing and pharmacokinetic calculations are reported (i.e ideal body weight vs. Actual body weight vs. Adjusted body weight)	NA	NA	NA	NA	NA	NA	NA	NA
	Statistical methods including software used are described	✓	✓	✓	✓	✓	✓	x	✓
	Study withdrawals or subjects lost-to-follow up (or lack thereof) are reported	x	x	x	x	x	✓	x	x
	All relevant variables that may explain inter- and intra-patient pharmacokinetic variability (including: age, sex, end-organ function, ethnicity, weight or BMI, health status or severity of illness and pertinent co-morbidities) are provided with appropriate measures of variability	✓	✓	x	✓	✓	✓	x	✓

Results	Results of pharmacokinetic analyses with appropriate measures of variability and precision (such as range, standart deviation, 95% confidence interval, etc.)	✓	x	✓	✓	✓	✓	x	✓
	Studies in patients receiving extracorporeal drug removal (i.e., dialysis) should report the mode of drug removal, type of filters used, duration of therapy and relevant flow rates	NA	NA	NA	NA	NA	✓	NA	✓
	In studies of drug bioavailability comparing two formulations of the same drug, F (bioavailability), Area Under the Curve (AUC), Cmax (maximum concentration) and tmax (time to maximum concentration) should be reported	x	x	✓	x	x	x	x	x

NA: Not applicable

x: Not included

✓: Included

4.2. Study Characteristics

4.2.1. Population Pharmacokinetic (popPK) Studies

Population pharmacokinetics investigates pharmacokinetic parameters at the population level, where data from all individuals within a population are collectively analysed using a nonlinear mixed-effects model. The term “nonlinear” signifies that the dependent variable (e.g., concentration) exhibits a nonlinear relationship with the model parameters and independent variables. The designation “mixed-effects” refers to the parameterisation: parameters that remain constant across individuals are termed “fixed effects,” while those that vary among individuals are classified as “random effects” (19).

The six articles included in this review each have distinct objectives. Ahsman et al. (2009) aim to elucidate the pharmacokinetics and exposure of oral sildenafil in neonates diagnosed with pulmonary arterial hypertension (PAH). Gonzalez et al. (2019) seek to characterise the population pharmacokinetics of sildenafil and its metabolites in premature infants. Milligan et al. (2002) focus on analysing the pharmacokinetics of sildenafil citrate in patients with erectile dysfunction. Cochius-den Otter et al. (2019) aim to achieve a specific target plasma concentration of the drug. Finally, both Mukherjee et al. (2008) and Rhee et al. (2022) seek to characterise the pharmacokinetics of sildenafil in term and preterm neonates with persistent pulmonary hypertension of the newborn (PPHN) and PAH, respectively.

Table 2: Characteristics of popPK Studies

	Population				
	Number of patients	Postnatal age	Gestational age	Body weight	Disease
1. Mukherjee A et al. , 2008 (16)	36	11-71 hours	36-42 weeks	2.5 - 4.4 kg	Persistent pulmonary hypertension of the newborn (PPHN)
2. Cochius - den Otter S. et al., 2019 (11)	23	0.3 - 0.9 days	35.8 - 39.2 weeks	2.15 - 3.75 kg	Congenital Diaphragmatc hernia (CDH) with pulmonary hypertension
3. Gonzalez D. et al. , 2019 (15)	34	7 - 279 days	22 - 28 weeks	0.59 - 8.06 kg	Pulmonary hypertension associated with Bronchopulmonary dysplasia (BPD)
4. Rhee S. et al., 2022 (14)	19	5 - 98 days	24 - 41 weeks	0.79 - 4.09 kg	PPHN Congenital Heart Disease (CHD) BPD

					Others
5. Ahsman M. et al., 2014 (18)	11	2 - 121 days	/	2 - 5.1kg	Pulmonary hypertension
6. Milligan P. et al., 2002 (12)	1335	19 - 87 years	/	49 - 159 kg	Erectile Dysfunction

	Sildenafil			Sample time	Metabolite
	Dose	Single dose	Multiple dose		
1. Mukherjee A et al. , 2008 (16)	Group 1 (i.v.): 0,0282 mg (loading dose); 0,629 mg (maintenance dose) Group 2 (i.v.): 0,0357 mg (loading dose); 0,879 mg (maintenance dose) Group 3 (i.v.): 0,09 mg (loading dose); 2,16 mg (maintenance dose) Group 4 (i.v.): 0,185 mg (loading dose); 4,326 mg (maintenance dose) Group 5 (i.v.): 0,43 mg (loading dose); 6,804 mg (maintenance dose) Group 6 (i.v.): 0,8909 mg (loading dose); 13,288 mg (maintenance dose) Group 7 (i.v.): 18,128 (maintenance dose) Group 8 (i.v.): 1,44 mg(loading dose); 19,822 mg (maintenance dose)	X	✓	1, 4, 8, 12, 24, 48 and 72h	UK - 103320
2. Cochius - den Otter S. et al., 2019	Loading dose (i.v.): 0, 35 mg/kg Continuous infusion	X	✓	Different times between 1 and 385 hours	UK - 103320

(11)	(i.v.): 1,5 mg/kg/day				
3. Gonzalez D. et al. , 2019 (15)	0,13 mg/kg - 2,09 mg/kg (i.v. or enteral)	X	✓	Cohort 1: 1-2; 3-4; 0,5h before the next dose (enteral) 0,25; 3-4 and 0,5h before the next dose (i.v.) Cohort 2: 15 min; 1-2; 3-4; 7-8; 12-14; 24-30 and 48-56 h	UK - 103320
4. Rhee S. et al., 2022 (14)	1,58 mg -9,8 mg per day during 2 weeks (per os) 2,37 mg - 12,27 mg per day if there was no clinical improvement (per os)	X	✓	/	UK - 103320
5. Ahsman M. et al., 2014 (18)	0,99 mg - 15,3 mg (nasogastric tube)	X	✓	0, 1, 2, 4 and 6 h	UK - 103320
6. Milligan P. et al., 2002 (12)	25 mg 50 mg 100 mg 200 mg	X	✓	1-3 h, 3-5 h, 5-7 h and >7 h postdosing	/

X: Not administered

✓: Administered

Table 3: Pharmacokinetic Methodology in popPK Studies

	PK Methodology				
	Program	Base Model	Inter Individual Variability	Covariates	Number of compartments
1. Mukherjee A et al. , 2008 (16)	NONMEM	V1, CL	V1, CL	Age, Weight, Sex	two-compartments
2. Cochius - den Otter S. et al., 2019 (11)	NONMEM	V1, V2, CL	V1, CL	Age	two-compartments
3. Gonzalez D. et al. , 2019 (15)	NONMEM	Ka, CL, V1, V2	V1, CL	/	two-compartments
4. Rhee S. et al., 2022 (14)	NONMEM	Ka, CL/F, V	CLI, V	Age, Weight, Sex,	one-compartment

5. Ahsman M. et al., 2014 (18)	NONMEM	Ka, CL/F, V	CL, V	/	one-compartment
6. Milligan P. et al., 2002 (12)	NONMEN	Ka, CL/F, V	/	Age, Aspartate transaminase (AST) concentration, co-administration of CYP3A4 potential inhibitors, co-administration of noncardioselective β blockers and body weight	one-compartment

4.2.2. Physiologically based pharmacokinetic modelling (PBPK) Studies

Physiologically-based pharmacokinetic (PBPK) models are mathematical frameworks that incorporate multiple compartments, integrating physiological, anatomical, biochemical, and physicochemical parameters to comprehensively describe absorption, distribution, metabolism, and excretion (ADME). These models can vary in complexity, ranging from empirical and semi-mechanistic to compartmental approaches, with compartmental models closely mirroring human anatomy and physiology. Currently, PBPK models are widely utilised in pharmaceutical and environmental sciences to predict the pharmacokinetic behaviour of drugs or chemicals, taking into account factors such as dose, route of administration, and species (20).

Physiological pharmacokinetic modelling was conducted using two distinct software programs. Ghoneim et al. (2020) employed the Simcyp Simulator, which accurately predicts drug behaviour within the human body, thereby supporting various stages of drug development. This includes determining optimal dosing strategies for first-in-human trials, optimising clinical study designs, evaluating new drug formulations, predicting drug-drug interactions, and conducting virtual bioequivalence analyses. The Simcyp Simulator is applicable to a wide range of drug types, including small molecules, biologics, antibody-drug conjugates, bispecifics, and oligonucleotides (21).

Salerno et al. (2022) utilised PK-Sim, an innovative whole-body physiology-based pharmacokinetic simulation software. PK-Sim® facilitates the modelling of the fate of xenobiotics, particularly pharmaceuticals, within the mammalian body through a mathematical representation of key physical and physiological processes. Its modular architecture is designed to meet the diverse needs of drug research and development companies, encompassing all phases from early research and lead optimisation to preclinical and clinical development stages (22).

Table 4: Characteristics of PBPK Studies

	Population				
	Number of patients	Postnatal age	Gestational age	Body weight	Disease
1. Ghoneim A. et al., 2020 (17)	100	20 - 45 years	/	/	/
2. Salerno S. et al., 2021 (13)	9	3 - 42 days	< 32 weeks	849 g	/

	Sildenafil			Sample time	Metabolite
	Dose	Single dose	Multiple dose		
1. Ghoneim A. et al., 2020 (17)	100 mg (per os)	✓	X	/	NA
2. Salerno S. et al., 2021 (13)	0,125 mg/kg or 0,25 mg/kg (i.v.)	✓	X	15 min, 1-2 h, 3-4 h, 7-8 h, 12 - 14 h, 24 - 30 h and 48 - 56 h	NA

NA: Not applicable
X: Not administered
✓: Administered

4.2.3. Other Pharmacokinetic analysis

All eleven studies employed non-compartmental analysis; consequently, systematic reviews and meta-analyses were not included in this evaluation.

Table 5: Characteristics of other Pharmacokinetic Studies

	Population			
	Number of patients	Postnatal age	Body weight	Disease
1. Olguín H.		1.4 - 15	7.5 - 26 kg	Pulmonary Arterial Hypertension (PAH)

et al., 2017 (23)	12	years		
2. Gao X. et al., 2014 (24)	18	18 - 55 years	> 50 kg	/
3. Muirhead G. et al., 2022 (25)	78	19 - 81 years	57 - 90 kg	First Study: Healthy Second Study: Renal impairment Third study: Chronic hepatic cirrhosis
4. Roh H. et al., 2013 (26)	120	20 - 50 years	47 - 87.1 kg	/
5. Bloemers J. et al., 2016 (27)	12	18 - 35 years	/	/
6. Loprete L. et al., 2018 (28)	12	18 - 45 years	/	/
7. Taylor-Cousar JL. et al., 2014 (29)	36	20 - 32 years	/	Pancreatic insufficient
8. Lv Y. et al., 2020 (30)	36	18 - 40 years	> 50 kg	/
9. Dadey E., 2015 (31)	18	25 - 60 years	59.9 - 100.1 kg	/
10. Lee S. et al., 2020 (32)	32	19 - 50 years	63.1 - 80.5 kg	/
11. Murtadha M. et al., 2021 (33)	36	18 - 55 years	56.91 - 88.12 kg	/

	Sildenafil			Sample time	Metabolite
	Dose	Single dose	Multiple dose		
1. Olguín H. et al., 2017 (23)	1 mg/kg (per os)	✓	X	0.25, 0.5, 1, 2, 4, 8 and 12h	NA
2. Gao X. et al., 2014 (24)	20 mg (per os)	✓	X	0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12 and 14h	NA
	50 mg (per os)	✓	X	First Study: 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 10, 12,	UK - 103320

3. Muirhead G. et al., 2022 (25)				16, 20, 24, 32, 40 and 48h Second Study: 0.5, 1, 1.5, 2, 4, 6, 8, 10, 12, 18, 24, 36 and 48h Third study: 0.25, 0.5, 1, 1.5, 2, 6, 10, 18, 24, 36 and 48h	
4. Roh H. et al., 2013 (26)	First Study: 50 mg (per os) Second Study: 100 mg (per os)	✓	X	0.017, 0.05, 0.117, 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 3, 4, 6, 8, 10, 12, and 24 h	UK - 103320
5. Bloemers J. et al., 2016 (27)	50 mg (per os)	✓	X	5, 10, 15, 20, 25, 30, 60, 90, 120, 135, 145, 165, 180, 195, 210, 225, 240, 270, 300, 330, 360, 390, 450, 570, 690, 810, 930 and 1590 min	UK - 103320
6. Loprete L. et al., 2018 (28)	50 mg (per os)	✓	X	1, 1.25, 1.5, 2, 2.5, 3, 4, 6, 8 and 12h	UK - 103320
7. Taylor-Cousar JL. et al., 2014 (29)	20 mg (per os)	X	✓	0.5, 1, 2, 4 and 6h	NA
8. Lv Y. et al., 2020 (30)	50 mg (per os)	✓	X	1, 1.5, 2, 3, 4, 6, 8, 10, 12 and 14h	NA
9. Dadey E., 2015 (31)	100 mg (per os)	✓	X	0.167, 0.333, 0.5, 0.75, 1, 1.25, 1.5, 1.75, 2, 2.5, 3, 4, 6, 8, 10, 12, 18 and 24h	NA
10. Lee S. et al., 2020 (32)	25 mg (per os)	✓	X	0.17, 0.33, 0.5, 0.75, 1, 2, 3, 4, 6, 8, 12 and 24h	UK - 103320
11. Murtadha M. et al., 2021 (33)	50 mg (per os)	✓	X	10, 20, 30 and 45 min and 1, 1.5, 2, 3, 4, 6, 8, 10, 12 and 24h	NA

NA: Not applicable
X: Not administered
✓: Administered

3.4. Model results - PopPK Studies

3.4.1. Paediatric population

The two studies outlined below utilise a one-compartment model, with sildenafil administered via the oral route.

Table 6: Pharmacokinetic parameters in paediatric population in one compartment models

	Ahsman M. et al., 2009 (18)	Rhee S. et al., 2022 (14)
CL	7,3 L/h	10,1 L/h
Vd	34 L	19,8 L

In the study by Rhee et al. (2022) (9), the equation for clearance per bioavailability (CL/F) is expressed as CL/F = TV $(BW/3,14)^{0,899}$, where the typical value (TV) is 10,1 L/h and the median body weight is 3,18 kg. The covariate identified is body weight, resulting in a CL/F value of 10,22 L/h for a child with a median body weight.

Conversely, the study by Ahsman et al. (2009) (18) did not consider any covariates.

The three studies discussed below employ a two-compartment model, with sildenafil administered intravenously (i.v.).

Table 7: Pharmacokinetic parameters in paediatric population in two compartment models

	Gonzalez D. et al., 2019 (15)	Cochius - den Otter S. et al., 2019 (11)	Mukherjee A. et al., 2008 (16)
TV (CL)	27,8 L/h	5,2 L/h	1,72 L/h
Vd	116 L	115 L	10,4 L
V2	28,1 L	10 L	12 L
Q	1,67 L/h	221 L/h	0,19 L/h

In the study by Gonzalez et al. (2019) (15), the equation for clearance (CL) is defined as CL = TV$(BW/70\ kg)^{0,96}$, where the typical value is 27,8 L/h and the median body weight is 3,41 kg. The identified covariate is body weight, leading to a CL value of 1,53 L/h for a child with a median body weight.

In the study by Cochius-den Otter et al. (2019) (11), the equation for CL is expressed as CL =TV $(BW/70)^{0,75}$ x $(Age/2,4)^{0,58}$, with a typical value of 5,2 L/h. The median body weight in this study is 2,95 kg, and the median postnatal age is 0,1 days. The covariate in this case is postnatal age, resulting in a CL value of 0,08 L/h for a child with a median postnatal age and body weight.

In Mukherjee et al. (2008) (16), the equation for CL is presented as CL = TV $(Age/3,5)^{0,58}$, with a typical value of 1,72 L/h. The median body weight is 3,4 kg, and the median postnatal age is 34,3 hours, approximately 1,4 days. The covariate identified is postnatal age, yielding a CL value of 1,02 L/h for a child with a median body weight.

To facilitate comparison between the CL values reported by Mukherjee et al. (2008) (11) and Cochius-den Otter et al. (2019) (13), we established median values for the parameters of postnatal age and body weight, specifically 2 days and 3,4 kg, respectively.

Based on these median values, the calculated CL for Cochius-den Otter et al. (2019) (6) is 0,48 L/h, while for Mukherjee et al. (2008) (16), it is 1,24 L/h.

3.4.2. Adult population

Table 8: Pharmacokinetic parameters in adult population in one compartment model

	Milligan P. et al., 2002 (12)
CL	58,5 L/h
Vd	310 L

One compartment equations

Table 9: Equation and interindividual variability of one compartment model

Study	CL (L/h)	Vd (L)	Interindividual variability	
	Equation	Equation	CL	Vd
1. Rhee S. et al., 2022 (14)	$$CL_{Sil}/F\ (L/h) = \theta_{CL(Sil)} \times (body\ weight/3.14)^{\theta weight} \times \theta_{female} \times (1 - e^{-\theta maturation \times postnatal\ age})$$	NA	23,4	37,7

NA: Not applicable

Two compartment equations

Table 10: Equation and interindividual variability of two compartment models

Study	CL (L/h)	Vd (L)	Interindividual variability	
	Equation	Equation	CL	Vd
1. Mukherjee A. et al., 2008 (16)	$CL_{par}(l/h) = \theta_{CL(par)} \cdot (Age/3.5)^{\theta age} \cdot (Weight/3.44)^{\theta weight} \cdot (1 + \theta male)$	$V1_{par}(l) = \theta_{V1(par)} \cdot (Weight/3.44)^{\theta weight} \cdot (1 + \theta male)$	54,6	43,7
2. Cochius-den Otter S. et al., 2002 (11)	NA	NA	50,3	32,3
3. Gonzalez D. et al., 2019 (15)	$CL = CL_{std} * \left(\frac{WT_i}{70\ kg}\right)^{\theta WT,CL}$	$V = CL_{std} * \left(\frac{WT_i}{70\ kg}\right)^{\theta WT,V}$	45,5	98,6
4. Ashman M. et al., 2014 (18)	NA	NA	42	94

NA: Not applicable

5. Discussion

This meta-analysis was conducted across three principal databases, focusing on the terms "pharmacokinetic" and "sildenafil," with a particular emphasis on the paediatric population.

The selection process for the articles began with the elimination of duplicates across the databases, followed by the categorization of each article based on their titles and abstracts into the following categories: bioequivalence, drug interactions, the influence of sildenafil on other medications, pharmacodynamics, and review articles. A total of one hundred and forty-eight articles were subsequently excluded to ensure that only the most relevant studies were included in the meta-analysis. These exclusions were based on the articles exploring themes that fell outside the scope of this investigation.

To elucidate the reasons for exclusion, those articles categorised as "Without access" were deemed potentially relevant based on their titles; however, full texts could not be accessed for review. Articles classified as “Case Reports” contained information pertaining to real patients treated with varying dosages of sildenafil for erectile dysfunction rather than pulmonary hypertension and did not address pharmacokinetic parameters. Additionally, some articles referenced interactions with other medications as well as dietary components. Finally, articles investigating new formulations of this active principle were also assessed, alongside those that did not address pharmacokinetic parameters or the ADME (Absorption, Distribution, Metabolism, and Excretion) process.

It is important to note the phenomenon of "over-rejection" in the elimination of articles from a meta-analysis. This practice involves the excessive discarding of studies or data, often due to overly stringent criteria. Such over-rejection can have multiple implications, including a reduction in data breadth, which may diminish the representativeness of the findings; publication bias, which may exacerbate the exclusion of studies with negative results, thus skewing the perceived effectiveness and presenting only positive outcomes; loss of valuable information that could enhance the analysis; and challenges to reproducibility, where a meta-analysis lacking a representative range of studies may struggle to replicate findings in future research, thereby compromising the robustness of the conclusions.

Moreover, over-rejection may introduce bias, ultimately limiting the external validity and applicability of the results. It is, therefore, essential that the inclusion and exclusion criteria are clearly defined and rigorously justified to mitigate these risks.

In total, six pharmacokinetic studies were included, three of which were one-compartment models, while the remaining three employed two-compartment models. Additionally, two physiologically based pharmacokinetic (PBPK) studies were included, conducted using different software programs. Of the six studies, five focused on the paediatric population, the majority of whom presented with pulmonary hypertension. Furthermore, eleven additional studies involving adult populations were identified, all of which were non-compartmental studies.

For the paediatric population, the key covariates identified were body weight and postnatal age, with average weights ranging from 2,95 kg to 3,41 kg. Notably, in the study by Mukherjee et al. (2008), the total volume of distribution of sildenafil in neonates was observed to be higher

than previously estimated in adults, potentially attributable to lower plasma protein binding in neonates compared to adults.

6. Conclusion

The pharmacokinetics of sildenafil was explored, a drug widely used in the treatment of erectile dysfunction and pulmonary arterial hypertension, with emerging applications in other conditions, such as secondary Raynaud's phenomenon and altitude-induced hypoxaemia. The systematic review encompassed population-based studies and physiologically based pharmacokinetic (PBPK) models, highlighting the different methodological approaches employed to assess the absorption, distribution, metabolism and excretion (ADME) parameters of this compound in both paediatric and adult populations.

The results indicated that factors such as body weight and postnatal age are important covariates in determining clearance and volume of distribution in neonates and children, with these values differing significantly from those observed in adults. Sildenafil exhibits a unique pharmacokinetic profile in neonates, particularly due to the reduced plasma protein binding in this population, which may account for its greater distribution compared to adults.

Despite advances in pharmacokinetic modelling and simulation techniques, gaps remain in the comprehensive understanding of sildenafil's long-term effects and drug interactions in various populations, particularly in paediatric patients. Future studies should focus on expanding the evidence base, incorporating more robust clinical trial methodologies and exploring new therapeutic indications for sildenafil.

In conclusion, sildenafil remains a drug of considerable clinical importance, and a deeper understanding of its pharmacokinetic parameters is essential for optimising its therapeutic use, ensuring both safety and efficacy across all patient populations.

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