FACULDADE DE CIÊNCIAS

DEPARTAMENTO DE CIÊNCIAS BIOLÓGICAS

PROGRAMA DE PÓS-GRADUAÇÃO EM BIOLOGIA E ECOLOGIA DE CONSERVAÇÃO

DISSERTAÇÃO DE MESTRADO

AGRICULTURA EM ÁREAS INDUSTRIAIS E CONTAMINAÇÃO POR METAIS PESADOS: UMA ANÁLISE AO RISCO ECOLÓGICO E TOXICOLÓGICO DE *ARACHIS HYPOGAEA*, *VIGNA UNGUICULATA* E *ZEA MAYS* CULTIVADAS EM SOLO CONTAMINADO POR CRÓMIO (CR)

MÁRIO MACHUNGUENE JÚNIOR

Maputo, 2024

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Dissertação apresentada ao programa de Pós-graduação em Biologia e Ecologia de Conservação da Universidade Eduardo Mondlane, como parte dos requisitos para obtenção do grau de Mestre em Biologia e Ecologia de Conservação.

Supervisor: Prof. Doutor Orlando António Quilambo
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Agradecimentos

Em primeiro lugar agradeço a Deus pela graça que me concedeu até o dia de hoje, pela oportunidade de mais uma vez poder me identificar como membro da Família UEM, por me ter dado forças e perseverança durante esta etapa da minha carreira académica e acima de tudo pela fé.

À minha mãe Maria Fernando Novela que me incentiva sempre a dar o meu melhor em tudo quanto me for confiado, ao meu pai Mário Machunguene e aos meus irmãos Verónica, Celina e Armando Mário Machunguene pelo amor, carinho, encorajamento e incansável apoio ao longo da minha carreira académica.

Aos meus supervisores a Prof.ª Doutora Célia Martins, o Prof. Doutor Orlando Quilambo e em especial a Mestre Sónia Ventura Guilundo pela compreensão, orientação, apoio e acompanhamento durante todo meu percurso.

A Universidade Eduardo Mondlane pela Bolsa de Estudos (Projecto SIDA 2.1.6).

Dedicatória

Aos meus pais,
E aos meus irmãos,
Dedico

Declaração

Declaro, por minha honra, que os dados apresentados neste trabalho refletem a verdade encontrada em campo e que este trabalho nunca foi apresentado, na sua essência ou parte dele, para obtenção de qualquer grau, e constitui o resultado da minha investigação, estando indicados na bibliografia as fontes utilizadas para o suporte científico.

(Mário Machunguene Júnior)

Resumo

A Agência Internacional de Pesquisa de Cancro (IARC) indica que o crómio é o principal agente cancerígeno que suscita sérias preocupações para a saúde humana e segundo Agência de Proteção Ambiental dos Estados Unidos (USEPA) o crómio está entre as 14 substâncias mais perigosas. Esse estudo teve com principal objectivo avaliar o risco ecológico e toxicológico do Cr no solo e nas plantas de amendoim (*Arachis hypogaea*), feijão (*Vigna unguiculata*) e no milho (*Zea mays*). Para tal foi montado um ensaio experimental na estufa, onde foi contaminado o solo com 10 e 30mg.kg^{-1} de Cr. Foi o usado o método de ICP-OES para quantificação dos metais no solo e nos tecidos das plantas, os factores de bioconcentração e translocação para analisar o movimento do metal nas plantas e foram calculados os índices de poluição (IP), potencial risco ecológico (E_i) e o risco incremental de contrair cancro ao longo da vida (RICCLV). Os resultados deste estudo demostraram haver um movimento significativo dos metais pesados do solo para as raízes das plantas e das raízes para as partes áreas. Os resultados do factor de translocação foram > 1 em todos os tratamentos para as três espécies. Nenhum risco ecológico ou toxicológico foi registado. O índice do potencial risco ecológico mostrou valores < 40 e o índice incremental de contrair cancro ao longo da vida mostrou valores abaixo e entre 1×10^{6}-1×10^{-4}. Esse estudo traz uma contribuição científica na área de ecotoxicologia em Moçambique e também fornece dados que poderão servir de base para a formulação de instrumentos de gestão ambiental e socio-ecológica em Moçambique.

Palavras chaves: Crómio, plantas alimentares, risco ecológico, risco toxicológico

Abstract

The International Agency for Research on Cancer (IARC) indicates that chromium is the main carcinogen that raises serious concerns for human health and according to the United States Environmental Protection Agency (USEPA) chromium is among the 14 most dangerous substances. This study's main objective was to evaluate the ecological and toxicological risk of Cr in the soil and in peanut (*Arachis hypogaea*), bean (*Vigna unguiculata*) and corn (*Zea mays*) plants. To this, an experimental test was set up in the greenhouse, where the soil was contaminated with 10 and 30mg.kg^{-1} of Cr. The ICP-OES method was used to quantify metals in soil and plant tissues, bioconcentration and translocation factors were used to analyze metal movement in plants, and pollution indices (PI) and potential ecological risk were calculated (Ei) and the incremental lifetime risk of contracting cancer (ILRCC). The results of this study demonstrated that there is a significant movement of heavy metals from the soil to the plant roots and from the roots to the aerial parts. Translocation factor results were > 1 in all treatments for the three species. No ecological or toxicological risks were recorded. The ecological potential risk index showed values < 40 and the incremental index of contracting cancer throughout life showed values below and between 1×10^{-6}-1×10^{-4}. This study makes a scientific contribution to the area of ecotoxicology in Mozambique and also provides data that can serve as a basis for the formulation of environmental and socio-ecological management instruments in Mozambique.

Keywords: Chromium, food plants, ecological risk, toxicological risk

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Capítulo I

1. Introdução

Nos últimos anos, a contaminação do solo por metais pesados tornou-se um problema primário e grave em muitas regiões do mundo (Adimalla *et al.*, 2019). Estudos relacionados à contaminação do solo agrícola e urbano receberam atenção especial devido ao aumento dos metais pesados causado pelas actividades antropogénicas (Al-Taani *et al.*, 2021). Metal pesado é qualquer elemento químico que apresenta propriedades metálicas, alta densidade (massa específica igual ou superior a $5g/cm^3$) e número atómico igual ou superior a 21, englobando os metais, semimetais e não metais como o selénio e o arsénio (Maronezi *et al.*, 2019; Zhu *et al.*, 2018).

Com o rápido crescimento populacional e expansão agro-industrial no mundo, a contaminação por metais pesados agravou-se devido a emissão de efluentes industriais, resíduos residenciais, uso de fertilizantes e emissões de gases contendo metais pesados para a atmosfera (Adimalla, 2019). A contaminação por metais pesados é preocupante por estes passarem através da cadeia alimentar por ingestão directa de vários produtos agrícolas, tais como vegetais, frutas e cereais, o que pode causar riscos para a saúde humana. Outro aspecto é que os metais pesados acumulados no solo infiltram-se através dos espaços porosos no solo e entram no sistema de águas subterrâneas deteriorando consequentemente a sua qualidade (Adimalla, 2019; Adimalla *et al.*, 2019; Zhu *et al.*, 2018).

Outra grande preocupação em relação aos metais pesados é o facto de serem potencialmente tóxicos, persistentes, não serem biodegradáveis, possuírem uma densidade que é cerca de 5 vezes maior do que a densidade da água, e ainda pelo facto de que uma pequena quantidade de metal pesado poder ser muito tóxica para o ser humano (Adewoyin *et al.*, 2023; Orosun *et al.*, 2023; Zhu *et al.*, 2018). Os metais pesados acumulam-se nos solos devido à deposição atmosférica proveniente de várias fontes e são assimilados pelas plantas, (Al-Taani *et al.*, 2021; Zhu *et al.*, 2018; Li *et al.*, 2015). A principal fonte está relacionada às emissões causadas pelas indústrias de mineração, metalurgia, construção, fundição, transformação, transporte e deposição de resíduos (Adewoyin *et al.*, 2023; Li *et al.*, 2015; Weissmannová e Pavlovský 2017).

O crescimento industrial causa uma progressiva escassez de terras para a prática da agricultura principalmente em áreas urbanas e periurbanas (Costa, 2015; Hu *et al.*, 2013). Vários estudos sobre a contaminação por metais pesados nos solos agrícolas em regiões urbanas e periurbanas têm sido relatados na literatura (Machunguene Jr *et al*., 2024; Sousa, 2021; Zhu *et al.,* 2018; Xiao *et al*., 2017; Costa, 2015; Li *et al.*, 2014; Atafar *et al*., 2010). No mundo inteiro, estima-se que cerca de 1.5 bilhões de pessoas, tanto em países desenvolvidos como em países em desenvolvimento praticam algum tipo de agricultura em áreas urbano-industrializadas, o que contribui com cerca de 15% dos alimentos (Fuller *et al*., 2022; FAO, 2018; FAO, IFAD, UNICEF, 2019). Em Moçambique a agricultura constitui a atividade económica de maior importância, por ser aquela que garante a subsistência de mais de 80% da população, com cerca de 88% dos agregados familiares a praticar esta actividade dentro e fora das áreas urbanas, com 99% da produção sendo destinada ao autoconsumo (Carrilho *et al.*, 2023; Chibuzor *et al.,* 2018; World Bank, 2022; Virga e*t al.,* 2007; WHO 2002).

Arachis hypogaea L. é a leguminosa mais importante em Moçambique e é a terceira cultura mais importante depois da *Zea mays* e da *Manihot esculenta*, sendo as províncias de Nampula e Cabo Delgado as principais produtoras (Bila *et al*., 2022; Chabite *et al*., 2020). A nível da Africa Austral, Moçambique é o maior produtor de *Arachis hypogea*, com alguns dos seus produtos derivados sendo consumidos internamente e outros exportados para Africa do Sul, India, China e Europa (Chabite *et al.,* 2020).

A *Vigna unguiculata* está entre as cinco principais leguminosas mais cultivadas em todo o mundo (Surukite *et al.*, 2023a). O seu elevado teor de proteínas (25%, com base no peso seco) é fundamental para aliviar a desnutrição e a pobreza, especialmente na África Subsaariana. Em particular, nos países africanos de língua portuguesa como Angola, a *V. unguiculata* é a cultura mais importante para a subsistência das comunidades rurais (Guimarães *et al.*, 2023). Em Moçambique, é, actualmente, a leguminosa mais cultivada e consumida. Tal como *A. hypogea*, sua produção é feita predominantemente por pequenos agricultores durante a estacão chuvosa (Guimarães *et al.*, 2023).

A *Zea mays L.* é um dos cereais mais produzidos no mundo, caracterizada pelas diversas formas de utilização, desde a alimentação humana e animal à indústrias de transformação (Boa *et al.*, 2023; Marcos *et al.*, 2023). Em Moçambique é a cultura mais extensivamente cultivada na agricultura de pequena escala e de sequeiro, e ocupa cerca de 1/3 da área total cultivada no país, com rendimentos de cerca de 1t/ha (Magaia *et al.*, 2016). É o cereal mais preferido na alimentação, e cerca de 46% das famílias rurais usam farinha de milho em sua dieta (Marcos *et al.*, 2023).

A exposição excessiva a metais pesados, especialmente quando estão em altas concentrações nas plantas alimentares ou contidos em pequenas proporções, pode representar grandes riscos à saúde humana (Machunguene Jr *et al.*, 2024; Pavlíková *et al.*, 2023; Zhu *et al.*, 2018; Xiao *et al.*, 2017). Podendo causar vários tipos de cancro (Mohammadi *et al*., 2019). A forma química de um metal pode influenciar a sua toxicidade e acumulação no corpo humano (USEPA, 2012). O crómio (VI) por exemplo, é muito mais tóxico que o crómio (III), tanto para exposições agudas quanto crônicas, e está ligado aos problemas de pele, dos sistemas respiratório e imunológico, hepática e renal, aos danos no ADN e stress oxidativo, cancro de estômago, cancro de pulmão e doença de Hodgkin (Georgaki *et al*., 2023; Emmanuel *et al.*, 2022; Sharma *et al.*, 2020; USEPA, 2012).

A Organização Mundial da Saúde (OMS) e Agência Internacional de Pesquisa de Cancro (IARC) classificam o Cr como o principal agente cancerígeno que suscita sérias preocupações para a saúde humana e segundo a Agência de Proteção Ambiental dos Estados Unidos (USEPA) o Cr está entre as 14 substâncias mais perigosas que podem causar sérios problemas de saúde aos organismos vivos (Georgaki *et al*., 2023; Emmanuel *et al.*, 2022; Sharma *et al.*, 2020; USEPA, 2012).

Um estudo realizado no Parque Industrial de Beluluane e Parque Industrial da Matola, demonstrou que as concentrações do alumínio (Al), arsénio (As), cobalto (Co), cádmio (Cd), crómio (Cr), chumbo (Pb), níquel (Ni) e zinco (Zn), estão acima dos padrões internacionais permitidos para as plantas alimentares (Machunguene Jr *et al.*, 2024), ademais, os metais podem causar toxicidade nas plantas (Pavlíková *et al.*, 2023; Sousa, 2021; Zhu *et al.*, 2018; Xiao *et al.*, 2017). Contudo, pouco se sabe sobre os mecanismos que permitem estas plantas fazer frente aos elevados níveis de metais pesados e se esses metais representam risco ecológico e toxicológico.

1.1 Problema

O risco ecológico causado pela contaminação por Cr ganhou muita atenção na última década devido aos seus elevados níveis na água e no solo (Ramos-Miras *et al*., 2020). A quantidade de resíduos contendo Cr descarregada pelas indústrias só no início desta década na África Subsaariana foi superior a 6 milhões de toneladas, com cerca 50.000L de água contaminada por tonelada de resíduo (Souza *et al.,* 2018; Wang *et al.,* 2021). Nesses países ainda existe uma deficiência no descarte de resíduos industriais que contêm Cr (Oruko *et al.*, 2021; Wang *et al.*, 2021). Sendo que este é considerado o segundo maior poluente metálico do solo, das águas subterrâneas e sedimentos, o que representa um sério risco ambiental (Ali *et al.,* 2023).

Na China por exemplo, é relatado que cerca de 12.500 mil toneladas de solo estão contaminadas por Cr, e esse valor tende a aumentar devido lixiviação (Orosun *et al*., 2023; Souza *et al.*, 2018; Wang *et al*., 2021). O Cr e suas espécies móveis e tóxicas quando são lixiviados a partir de resíduos industriais, são retidos no solo por um longo período de tempo, podendo ser absorvidos pela vegetação, contaminando o ecossistema terrestre, fontes de água superficiais e subterrâneas (Vardhan *et al.*, 2019; Xu *et al.*, 2023).

Embora em níveis vestigiais seja considerado elemento essencial por regular o metabolismo e atividades no corpo humano, quantidades excessivas deste elemento no ambiente são altamente perigosas e estão potencialmente relacionadas com o surgimento de cancro no homem, e redução do crescimento em plantas e animais (Oruko *et al.,* 2021). Segundo a Agência Internacional de Pesquisa de Câncer (IARC) o Cr é o principal agente cancerígeno para o Homem e segundo a Agência de Proteção Ambiental dos Estados Unidos (USEPA) está entre as 14 substâncias mais perigosas para os organismos vivos (Sharma *et al.*, 2020; USEPA, 2012).

A contaminação por crómio não é facilmente detectável, e ao exceder a tolerância ambiental, pode causar grandes danos ecológicos, para além de ser bioacumulável e passivo de adentrar ao longo da cadeia alimentar (Vardhan *et al.,* 2019). Um estudo feito por Machunguene Jr *et al*., (2024), nos parques industrias de Beluluane e Matola constatou que concentrações de crómio e outros metais pesados estão acima dos padrões internacionais de qualidade, daí a necessidade de ser avaliado o risco ecológico e toxicológico do crómio em plantas alimentares cultivadas em solos contaminados.

1.2 Justificativa

Na última década, tem havido uma enorme pressão sobre os países subsaarianos por parte das instituições de investigação, sociedade civil e de vários órgãos de controlo de poluição para regular e minimizar as descargas de diferentes indústrias, sob visão das Nações Unidas para os Objectivos de Desenvolvimento Sustentável (ODS) (Joston 2016; Oruko *et al*., 2021). Por outro lado, a corrida pelo desenvolvimento industrial em África, particularmente em Moçambique, causa a libertação para a atmosfera de resíduos que contêm material particulado, óxidos, compostos orgânicos e/ou voláteis e ainda metais pesados, e à semelhança do que acontece no resto do mundo, traz grandes desafios para a saúde do homem e do meio ambiente (Maronezi *et al.,* 2019; Souza *et al.,* 2018). Em muitos países, tendo como bases pesquisas quantitativas e qualitativas foram elaborados regulamentos e políticas ambientais para controlar a emissão de resíduos industriais dentro dos limites-padrão da OMS e da EPA (Oruko *et al.*, 2021). Devido ao possível potencial risco ecológico nos ecossistemas e o impacto directo na população humana, foi essencial pesquisar e compreender o comportamento do Cr nas plantas e o risco que este representa, de modo a fornecer dados que poderão servir de base para a formulação de instrumentos de gestão ambiental e socio-ecológica em Moçambique.

2. Objectivos

2.1. Geral

- Avaliar o risco ecológico e toxicológico de *Arachis hypogaea*, *Vigna unguiculata* e *Zea mays* cultivadas em solo contaminado por crómio (Cr).

2.2. Específicos

- Determinar os parâmetros de crescimento das plantas cultivadas em solo contaminado por Cr.
- Determinar a acumulação e translocação do Cr nas plantas cultivadas em solo contaminado por Cr.
- Determinar o potencial risco ecológico da contaminação do solo por Cr.
- Determinar o risco toxicológico resultante do consumo das plantas contaminadas por Cr.

2.3. Hipóteses

O Cr e suas espécies móveis e tóxicas quando são lixiviados a partir resíduos industriais, são retidos no solo por um longo período de tempo, podendo ser absorvidos pela vegetação, contaminando o ecossistema terrestre, fontes de água superficiais e subterrâneas (Vardhan *et al*., 2019; Xu *et al.*, 2023). Embora sejam conhecidas suas funções no metabolismo dos animais, quantidades excessivas no ambiente e a exposição a este, podem representar grandes riscos à saúde e ao ecossistema (Pavlíková *et al.*, 2023; Oruko *et al*., 2021; Zhu *et al.*, 2018; Xiao *et al.*, 2017).

H_0 – A contaminação do solo por Cr não representa risco ecológico e toxicológico.

H_1 – A presença do Cr no solo acima dos padrões de qualidade representa risco ecológico para os ecossistemas.

H_2 – O consumo de plantas cultivadas em solo contaminado por Cr representa risco toxicológico para o homem.

3. Revisão bibliográfica

3.1. Contaminação por metais pesados

A contaminação do meio ambiente por metais pesados pode ser definida como o aumento na concentração natural de metais pesados no solo causado pelas actividades antropogénicas (Mohamed *et al*., 2020). Em ambientes urbanos a contaminação por metais pesados, dá-se pela transferência deste do solo para o corpo humano através da inalação de poeiras, absorção pela pele ou consumo de alimentos contaminados (Mohamed *et al*., 2020; Mohammadi *et al*., 2019; Souza *et al*., 2018).

3.2. Toxicidade dos metais pesados

A toxidade dos metais pesados é definida como o efeito nocivo aos organismos vivos, podendo ser aguda se a dose for elevada durante o período de exposição, ou crónica se a exposição perdurar longos períodos de tempo (Maronezi *et al.*, 2019; Zhu *et al.,* 2018).

3.3. Fontes de contaminação

É qualquer atividade ou produto através do qual se originou a contaminação. Representa uma localização através da qual, por meio de algum transporte, o contaminante chega ao meio ambiente (Ramos-Miras *et al*., 2020).

As fontes de contaminação por Cr são maioritariamente provenientes das industriais (Mohamed *et al*., 2020). As fontes industriais incluem produção de cromato, produção de ferrocrómio e pigmentos de Cr, processamento de metais, curtumes e soldagem de aço inoxidável (Ramos-Miras *et al*., 2020). O crómio libertado no ar e nas águas residuais é proveniente das indústrias metalúrgica, química e refratária (Mohamed *et al*., 2020).

3.4. Risco ecológico

É a probabilidade do meio ambiente ser impactado como resultado da exposição a um ou mais estressores ambientais, como produtos químicos, mudanças no uso da terra, doenças e espécies invasoras (Park e Lek, 2015).

3.4.1. Avaliação de risco ecológico

É um processo científico que determina a probabilidade de ocorrência de danos as plantas e animais que possam ser expostos a substâncias perigosas de um local contaminado (EPA, 2017). É usado para: estudar como uma planta ou animal pode ser exposto a um contaminante e para determinar se o ecossistema ou parte dele será afetado negativamente (EPA, 2017; Park e Lek, 2015).

3.5. Risco toxicológico

Mede a probabilidade de um efeito adverso ser causado por um determinado composto. Isto abrange efeitos como irritações cutâneas causada por exposição atípica até riscos mais graves, como a probabilidade de um material ser cancerígeno ou fatal ao ser ingerido (USEPA, 2004; 2014)

3.5.1. Avaliação de Risco Toxicológico

É uma avaliação abrangente da segurança de um produto com base em sua composição, materiais e usos pretendidos (Park e Lek, 2015).

3.6. Crómio (Cr)

O Cr é considerado o 21º elemento mais abundante da crosta terrestre, com número atómico igual a 24 (24 prótons e 24 elétrons) e massa atômica igual 52µ (Sousa e Santos, 2018; Costa *et al*., 2021). É o 6º elemento em sua escala de metais de transição da tabela periódica (Costa *et al*., 2021). Sua forma metálica é obtida mediante o processamento da cromita ($FeO.Cr_2O_3$), mineral com maior aproveitamento econômico (Maronezi *et al.*, 2019). É encontrado em todas as fases no ambiente, incluindo ar, água e solo, e suas diversas formas químicas são poluentes com graves implicações para o meio ambiente e para a saúde humana (Sousa e Santos, 2018). Geralmente, o Cr total presente nos solos não está disponível, por se encontrar em compostos insolúveis, tais como óxidos de ferro e alumínio, ou fortemente fixados pela argila e pela matéria orgânica (Maronezi *et al*., 2019). Por origem antropogénica pode surgir associado a emissões industriais devido a produção do aço inoxidável, sendo a principal causa de contaminação dos solos os resíduos do curtimento do couro (Costa, 2015).

3.7. Biodisponibilidade do Cr

O Cr é encontrado na natureza sob duas formas Cr (III) e Cr (VI), distintas quanto à mobilidade, toxicidade e características físico-químicas (Chen *et al.*, 2022; Srivastava *et al.*, 2021). No geral, a espécie trivalente pode ser encontrada naturalmente na forma de minerais e sob ambientes redutores. É relativamente estável e apresenta uma baixa solubilidade em água sendo, consequentemente, pouco móvel (Srivastava *et al*., 2021; Maronezi *et al*., 2019). As espécies que predominam estão sob a forma de Cr(III), Cr(OH)(II), Cr(OH)(II), $Cr(OH)_3$ e suas frações solúveis podem ser facilmente adsorvidas pelos constituintes do solo e, em concentrações traço, são consideradas elementos essenciais para a nutrição animais e do homem (Costa *et al*., 2021; Srivastava *et al*., 2021; Maronezi *et al*., 2019). Por outro lado, a espécie hexavalente ocorre em condições oxidantes, sendo na maioria dos casos, de origem antropogénica (Srivastava *et al.*, 2021; Chibuzor *et al.*, 2018). Devido a sua forma aniónica como dicromato e cromato por exemplo, o Cr(VI) é mal adsorvido pelas componentes do solo e apresenta alta reatividade e solubilidade em água e é portanto, um importante contaminante de águas subterrâneas (Santos *et al.*, 2018; Sousa e Santos, 2018; WHO, 2017).

3.8. Bioacumulação do Cr

A bioacumulação é o termo geral que descreve o processo pelo qual uma substância (ou elemento químico) é absorvida pelos organismos vivos (Costa, 2020). O processo pode ocorrer de forma directa, quando no caso do Cr, este é assimilada a partir do meio ambiente (solo, sedimento, água) ou de forma indirecta quando é pela ingestão de alimentos contaminados (Chen *et al.*, 2022). O processo de bioacumulação do Cr pode desencadear um processo denominado de biomagnificação. A biomagnificação consiste na transferência deste para pelo menos dois níveis tróficos em uma teia alimentar (Costa, 2020).

3.9. Factor de bioconcentração e translocação

O factor de bioconcentração é uma razão da concentração de metal nos tecidos específicos (raiz, caule, folha e fruto) para a concentração no ambiente circundante (solo ou rizosfera) (Khalid *et al*., 2017). O factor de translocação é a capacidade da planta em captar e distribuir o metal pelos seus tecidos (Souza *et al*., 2019).

3.10. Toxicidade do Cr nas plantas

Até o momento, o Cr não tem nenhum papel biológico conhecido na fisiologia das plantas (Sharma *et al.,* 2020). Apesar de apresentar baixa acumulação na rizosfera e consequentemente baixa translocação para a parte aérea das plantas, quando absorvido e translocado, o resultado é a restrição do crescimento e do desenvolvimento das plantas, afetando negativamente os processos metabólicos essenciais (Bashri *et al.,* 2016; Costa *et al.,* 2021; Reis. 2019; Srivastava *et al.,* 2021). Vários estudos observaram que o estresse por Cr impõe um efeito prejudicial às células radiculares e o amarelecimento das folhas jovens prejudica a taxa fotossintética e a redução da biomassa (Chen *et al.*, 2022; Balali-Mood *et al.,* 2021; Srivastava *et al.*, 2021; Sinha *et al.*, 2018; Shanker *et al.,* 2005). A toxicidade do Cr induz alterações ultraestruturais, modificações da membrana celular e dos cloroplastos, provoca clorose nas folhas, danifica as células das raízes, reduz o conteúdo de pigmentos, perturba as relações hídricas e a nutrição mineral, afeta a transpiração e assimilação de nitrogênio e altera diferentes atividades enzimáticas (Bashri *et al.,* 2016; Saleem *et al.,* 2022; Srivastava *et al.,* 2021).

Postula-se que todos os efeitos tóxicos de Cr podem ser devido à produção excessiva de espécies reativas de oxigênio (ROS), que acabam por perturbar o equilíbrio redox nas plantas (Srivastava *et al.,* 2021). Sob condições naturais, as ROS estão envolvidas em vários aspectos essenciais ao metabolismo das plantas, como regulação da condutância estomática, transdução de sinal para célula programada para morte, quebra de dormência das sementes, senescência, regulação do crescimento, amadurecimento dos frutos e início da defesa metabólica sob estresse (Saleem *et al.*, 2022; Srivastava *et al.,* 2021).

3.10.1. Toxicidade do Cr no homem

Em baixas concentrações, o Cr está envolvido no metabolismo dos lípidos e proteínas, de modo que em pequenas quantidades é necessário para as funções vitais (Khan *et al*., 2019). A ingestão diária do Cr, normalmente é de cerca de 100µg proveniente de alimentos como grãos, frutas e vegetais (Khan *et al*., 2019; Achmad *et al.,* 2017). Porém, quantidades acima do recomendado podem desencadear efeitos adversos que incluem irritação e obstrução de vias respiratórias, asma, bronquite crônica e faringite (Georgaki *et al.,* 2023; Sharma *et al.*, 2020; USEPA, 2012). O contacto dérmico é responsável por desencadear alergias como dermatites, eritema e pápulas (Achmad *et al.,* 2017). O Cr(VI) está ligado ao cancro de estômago, cancro de pulmão e doença de Hodgkin (Georgaki *et al.*, 2023). Pode prejudicar a pele, os olhos, os sistemas respiratório e imunológico, hepático, renal, e causar danos ao ADN, levando ao crescimento de tumores (Georgaki *et al.*, 2023; Yang *et al.*, 2019). A exposição a longo prazo tem o potencial de impactar negativamente à saúde e depende de vários factores, como sexo, idade e peso corporal, mesmo em níveis baixos (Georgaki *et al.*, 2023; Yang *et al.*, 2019).

3.11. Plantas alimentares

3.11.1. *Arachis* hypogaea L

A *Arachis hypogaea* L. é uma cultura anual pertencente à família Fabaceae (Leguminosae). O gênero *Arachis* contém cerca de 80 espécies. A *A. hypogaea* é amplamente cultivada e consumida globalmente (Bila *et al.*, 2022; Santos *et al.*, 2021). É uma cultura que se desenvolve bem em diferentes condições de temperatura, clima e solo, por isso é considerada uma planta rústica (Santos *et al.*, 2021). É de grande importância econômica para Moçambique e para o mundo todo, por estar directamente ligada à indústria, pois possui alto valor nutricional, com composição rica em óleo e proteínas (Chabite *et al.*, 2020; Santos *et al.*, 2021).

Como a maioria das outras leguminosas, ela abriga bactérias simbióticas fixadoras de nitrogênio nos nódulos das raízes (Njoroge, 2018). A capacidade de fixar nitrogénio significa que ela requer menos fertilizantes contendo nitrogénio e melhora a fertilidade do solo, tornando-a valiosas nas rotações de culturas (Njoroge, 2018). De acordo com as estatísticas da FAO, só em 2019 a produção global da *A. hypogaea* foi de cerca de 48,76 milhões de toneladas (Ding *et al*., 2023). *A. hypogaea* é suscetível à contaminação por metais pesados, e seu cultivo em solos contaminados levanta preocupações sobre o seu consumo e a dos seus produtos derivados (Ding *et al*., 2023). É relatado que a *A. hypogaea* tem alta capacidade de hiperacumular metais pesados, o que pode afetar significativamente seu rendimento e qualidade, entretanto os efeitos das alterações fisiológicas e bioquímicas induzidas pela exposição aos metais nesta planta ainda não foram estudados (Ding *et al*., 2023; Nareshkumar *et al.*, 2015).

3.11.2. *Vigna unguiculata*

A *Vigna unguiculata* é uma planta anual, trepadeira, rastejante ou ereta, crescendo de 15 a 90 cm de altura ou de 2 a 3 m para formas trepadeiras (Singh *et al.*, 2014). Pertencente a família *Fabaceae* (Leguminosae) subfamília: *Fabaideae* tribo: *Phaseoleae* subtribo: *Phaseolinae* subgênero: *Vigna*. (Singh *et al.*, 2014). Está entre as cinco principais leguminosas mais cultivadas em todo o mundo (Surukite *et al.*, 2023a). É uma espécie polivalente, utilizada para consumo humano e animal com elevado teor de proteínas. É uma das principais fontes dietéticas de carbohidratos e proteínas à base de plantas nas regiões tropicais e subtropicais, onde é amplamente cultivado, consumido e usada para controlar a desnutrição proteico-energética principalmente em Moçambique e Angola. É rica em vitamina A, tiamina, ácido fólico, riboflavina, biotina, ferro e cálcio (Guimarães *et al*., 2023; Chibuzor *et al.*, 2018; Anitha *et al.*, 2012). É utilizada para manutenção e melhoramento do solo, sendo uma das leguminosas mais utilizadas nos países tropicais na rotação de culturas (Guimarães *et al*., 2023; Ailenokhuoria e Omolekan, 2019). É conhecida por emitir compostos químicos na rizosfera que atraem bactérias fixadoras de nitrogênio, as rizóbios (Surukite *et al.*, 2023b). Os nódulos formados nas raízes, protegem as rizóbios e fornecem uma fonte de carbono e em troca, elas recebem uma forma útil e estável de nitrogênio (Surukite *et al.*, 2023b). É descrita como sendo uma planta com potencial fitoremediador de solos contaminados por metais pesados (M. D. M. Silva et al., 2013; Surukite et al., 2023b).

3.11.3. *Zea mays*

O gênero *Zea* é membro da tribo Andropogoneae da família das gramíneas (Poaceae). Existem 5 espécies conhecidas do gênero *Zea*, das quais uma é a *Z. mays* que por sua vez é subdividido em 4 subespécies (Noman *et al.*, 2015). A *Z. mays* é uma gramínea anual ereta que normalmente atinge 2 a 3m de altura, possui um único colmo principal com nós e entrenós e, dependendo da base genética e da densidade populacional da planta, pode possuir 1 a 2 ramos laterais conhecidos como perfilhos nas axilas inferiores das folhas (Noman *et al.*, 2015). É uma planta economicamente importante, sendo um dos cereais mais produzidos no mundo, principalmente em regiões tropicais e subtropicais, com elevado valor nutricional (Boa *et al*., 2023; Rosas-Castor *et al.*, 2014). Em Moçambique é a cultura mais cultivada e mais consumida principalmente palas famílias rurais (Marcos *et al*., 2023; Magaia *et al*., 2016). É uma planta com alta capacidade de sobreviver sob escassez de água e estresse oxidativo (Noman *et al*., 2015). Sua produção só é reduzida devido aos impactos negativos de factores abióticos, como aumento da concentração de sais, alteração do regime de temperatura, escassez de água e toxicidade de metais. É conhecida como sendo uma planta proficiente em acúmulo de metais pesados e com a melhor taxa de fitoextração (Razzaq *et al*., 2024; Noman *et al*., 2015; Shanker *et al*., 2005).

3.12. Solo

O solo é um componente do ecossistema terrestre biologicamente ativo que se desenvolveu na camada superior da crosta terrestre. É um dos principais substratos da vida na Terra (Sposito, 2024). Serve como reservatório de água e nutrientes, como meio de filtração e decomposição de resíduos prejudiciais e participa na ciclagem do carbono e de outros elementos através do ecossistema. Encontra-se em contacto com a atmosfera, a litosfera, a hidrosfera e com a biosfera (Sposito, 2024; Costa, 2015). É composto por três fases, a sólida, a gasosa e a líquida. A primeira é constituída pela matriz do solo e contém os minerais e matéria orgânica. A fase gasosa corresponde à atmosfera do solo e a fase líquida representa a água do solo com substâncias dissolvidas, também conhecida como solução do solo (Disale *et al.*, 2020).

O solo apresenta quatro funções essenciais: suporte ao crescimento das plantas, fornecendo o ambiente para o desenvolvimento das raízes, água e nutrientes; recicla resíduos e tecidos mortos de seres vivos, ficando os elementos destes materiais novamente disponíveis no solo; fornece nichos ecológicos onde vivem milhões de organismos, desde pequenos mamíferos a fungos e bactérias; controla a infiltração da água e, assim, a sua qualidade como água de recarga dos aquíferos (Sposito, 2024; Costa, 2015).

3.12.1. pH do solo

O pH do solo se refere ao grau de acidez ou alcalinidade do solo. A acidez do solo define a concentração dos iões H^+ livres existentes na solução do solo, enquanto a alcalinidade representa o grau saturação do complexo de trocas, devido à ausência de arrastamento de bases por águas de infiltração (Adamczyk-Szabela e Wolf 2022). A escala de pH varia de 0 a 14; um pH de 7 é considerado neutro, se os valores forem maior que 7, a solução é considerada alcalina e se forem menores que 7 é considera básica (Alderman *et al.*, 2018).

Tabela 1: Escala de Pratolongo (modelo de Varennes, 2003)

pH do solo	Designação
<4.5	Hiperácido
4.6 – 5.5	Ácido

5.6 – 6.5	Subácido
6.6 – 7.5	Neutro
7.6 – 8.5	Subalcalino
8.6 – 9.5	Alcalino
> 9.6	Hiperalcalino

3.12.2. Capacidade de troca catiónica do solo

A capacidade de troca catiónica (CTC) de um solo é uma medida que representa a quantidade total de cations retidos à superfície do solo em condição permutável (Ca^{2+}, Mg^{2+}, K^{+}, Na^{+}, H^{+}, Al^{3+}) (Adamczyk-Szabela e Wolf 2022). Os primeiros quatro catiões correspondem às bases de troca, enquanto os últimos dois correspondem à acidez de troca e podem ser classificadas de forma qualitativa numa escala de muito baixa a muito alta segundo o modelo (Varennes, 2003).

Tabela 2: Classificação das bases de troca (modelo de Varennes, 2003)

Classificação	**Bases de troca (kg^{-1})**			
	Ca^{2+}	**Mg^{2+}**	**K^{+}**	**Na^{+}**
Muito baixa	<2.0	<0.5	<0.1	<0.1
Baixa	2.0-5.0	0.5-1.0	0.1-0.25	0.1-0.25
Média	5.1-10.0	1.1-2.5	0.26-0.50	0.26-0.50
Alta	10.1-20.0	2.6-5.0	0.51-1.0	0.51-1.0
Muito alta	>20.0	>5.0	>1.0	>1.0

3.12.3. Matéria Orgânica do solo

A matéria orgânica do solo e definida como o conjunto de compostos ou materiais orgânicos de origem vegetal ou animal, liteira, fragmentos de resíduos, biomassa microbiana, compostos solúveis e a matéria líquida ligados aos argilominerias do solo (Cunha *et al.,* 2016). A matéria orgânica pode ser classificada como baixa, media ou alta em relação a sua percentagem na solução do solo:

Tabela 3: Classificação da matéria orgânica

Classificação	**Matéria orgânica (%)**
Baixa	<2.5
Media	2.5 – 5
Alta	>5

3.12.4. Textura do solo

O termo textura do solo refere-se à proporção de partículas de diferentes dimensões na terra. A terra fina corresponde ao material com diâmetro inferior a 2 mm, sendo a principal responsável pelas propriedades químicas do solo. As partículas de terra fina podem ser subdivididas em: areia grossa (2 – 0,2 mm), areia fina (0,2 – 0,02 mm), limo (0,02 – 0,002 mm) e argila (<0,002 mm). Por outro lado, as partículas de dimensões superiores a 2 mm são designadas por elementos grossos (Costa, 2015; Embrapa, 2010).

Capítulo II

Assessment of the ecological and toxicological risk of *Arachis hypogaea, Vigna unguiculata* and *Zea mays* cultivated in soil contaminated by chromium (Cr)

Abstract

The International Agency for Research on Cancer (IARC) indicates that chromium is the main carcinogen that raises serious concerns for human health and according to the United States Environmental Protection Agency (USEPA) chromium is among the 14 most dangerous substances. This study's main objective was to evaluate the ecological and toxicological risk of Cr in the soil and in peanut (*Arachis hypogaea*), bean (*Vigna unguiculata*) and corn (*Zea mays*) plants. To this end, an experimental test was set up in the greenhouse, where the soil was contaminated with 10 and 30mg.kg $^{-1}$. The ICP-OES method was used to quantify metals in soil and plant tissues, bioconcentration and translocation factors were used to analyze metal movement in plants, pollution indices (PI), the potential ecological risk were calculated (E*i*) as well as the Lifetime incremental cancer risk (ILCR). The results showed a significant translocation of heavy metals from the soil to the plant roots and from the roots to the aerial parts. Translocation factor results were higher than 1 in all treatments for the three species. No ecological or toxicological risks were recorded. The ecological potential risk showed index values lower than 40 and the incremental index of developing cancer throughout life showed values between 1×10^{-6} and 1×10^{-4}. This study contributes to the area of ecotoxicology in Mozambique and also provides data that can serve as a basis for the formulation of environmental and socio-ecological management instruments in Mozambique.

Keywords: Carcinogenic risk, crops, greenhouse, heavy metal, industrial soil.

1. Introduction

In recent years, soil contamination by heavy metals has become a primary and serious problem in many regions of the world (Adimalla *et al.*, 2019)**.** Studies related to agricultural and urban soil contamination have received special attention due to the increase in heavy metals concentration caused by anthropogenic activities (Al-Taani et al., 2021a)**.** Heavy metal is any chemical element that has metallic properties, high density (specific mass equal to or greater than 5g/cm^3) and atomic number equal to or greater than 21, including metals, semimetals and non-metals such as selenium and arsenic (Maronezi *et al*., 2019; Zhu *et al*., 2018)**.**

With a rapid population growth and agro-industrial expansion in the world, contamination by heavy metals has worsened due to the emission of industrial effluents, residential waste, use of fertilizers and emissions of gases containing heavy metals into the atmosphere (Adimalla, 2019). Contamination by heavy metals is a concern because they pass through the food chain through direct ingestion of various agricultural products, such as vegetables, fruits and cereals, which can cause risks to human health. Another aspect is that heavy metals accumulated in the soil infiltrate through the porous spaces in the soil and enter the groundwater system, consequently deteriorating its quality (Adimalla, 2019; Adimalla *et al*., 2019; Zhu *et al*., 2018)**.** Another major concern regarding heavy metals is the fact that they are potentially toxic, persistent, non-biodegradable, have a density that is approximately 5 times greater than the density of water, and also the fact that a small amount of metal heavy can be very toxic to a humans (Adewoyin *et al*., 2023; Costa, 2015; Orosun *et al*., 2023; Zhu *et al*., 2018).

Industrial growth causes a progressive scarcity of land for agricultural practice, mainly in urban and peri-urban areas (Costa, 2015; Hu *et al.*, 2013)**.** Several studies on heavy metal contamination in agricultural soils in urban and peri-urban regions have been reported in the literature (Atafar *et al.*, 2010; Costa, 2015; Z. Li *et al*., 2014; Machunguene Jr *et al*., 2024; Sousa and Santos, 2018; Xiao *et al*., 2018; Zhu *et al*., 2018)**.** Worldwide, it is estimated that around 1.5 billion people, both in developed and developing countries, practice some type of agriculture in urban-industrialized areas, which contributes with at least 15% of food (FAO, IFAD, UNICEF, 2019; FAO, 2018; Fuller *et al*., 2022)**.** In Mozambique, agriculture is the most important economic activity, as it guarantees the subsistence of more than 80% of the population, with around 88% of households practicing this activity inside and outside urban areas (Chibuzor *et al*., 2018; Ramos-Miras *et al*., 2020; The World Bank, 2022; Virga *et al*., 2007; WHO, 2002).

The ecological risk caused by Cr contamination has gained much attention in the last decade due to its high levels in water and soil (Ramos-Miras *et al*., 2020). The amount of Cr-containing waste discharged by industries at the beginning of this decade in Sub-Saharan Africa alone was more than 6 million tons, with around 50,000L of contaminated water per ton of waste (Souza *et al*., 2018; Wang *et al*., 2021). In these countries there is still a lack disposal of industrial waste containing Cr (Al-Taani *et al*., 2021b; Oruko *et al*., 2021). This is considered the second largest metallic pollutant in soil, groundwater and sediments, which represents a serious environmental risk (Ali *et al*., 2023).

Although at trace levels Cr is considered an essential element for regulating metabolism and activities in the human body, excessive amounts of this heavy metal in the environment are highly dangerous and are potentially related to the emergence of cancer in humans, and also reduced growth in plants and animals (Ali *et al*., 2023). Cr contamination is not easily detectable, and when exceeding environmental tolerance, it can cause great ecological damage, in addition to being bioaccumulative and prone to entering along the food chain (Ali *et al*., 2023; Vardhan *et al*., 2019).

In Mozambique *Arachis hypogaea* L. is the most important legume and is the third most important crop after *Zea mays* and *Manihot esculenta* (Bila *et al*., 2022; Ibraimo Teleha Chabite *et al*., 2020)**.** At the Southern Africa, Mozambique is the largest producer of *Arachis hypogea*, with some of its derivatives being consumed domestically and others exported to South Africa, India, China and Europe (Chabite *et al*., 2020)**.**

On other hands, *Vigna unguiculata* is among the top five most cultivated legumes worldwide (Surukite *et al*., 2023a)**.** Its high protein content (25%, based on dry weight) is key to alleviating malnutrition, especially in sub-Saharan Africa. In particular, in Portuguese-speaking African countries, such as Angola, *V. unguiculata* is the most important crop for the subsistence of rural communities (Guimarães *et al*., 2023)**.** In Mozambique, it is currently the most cultivated and consumed legume. Like *A. hypogea*, its production is predominantly carried out by small farmers during the rainy season (Marcos *et al*., 2023).

Zea *mays L.* is one of the most produced cereals in the world, characterized by different forms of use, from human and animal food to processing industries.(Boa *et al*., 2023; Marcos *et al*., 2023) In Mozambique it is the most extensively cultivated crop in small-scale and rainfed agriculture, and occupies around 1/3 of the total cultivated area in the country, with yields of around 1t/ha (Pavlíková *et al*., 2023). It is also the most preferred cereal in the diet, and around 46% of rural families use corn flour in their diet (Boa et al., 2023; Magaia et al., 2016; Pavlíková et al., 2023).

Excessive exposure to heavy metals, especially when they are in high concentrations in food plants or contained in small proportions, can pose major risks to human health (Machunguene Jr *et al*., 2024; Pavlíková *et al*., 2023; Xiao *et al*., 2018; Zhu *et al*., 2018)**.** It can cause various types of cancer (Sharma *et al.*, 2020). According to the International Agency for Research on Cancer (IARC), Cr is the main carcinogen for humans and according to the United States Environmental Protection Agency (USEPA) it is among the 14 most dangerous substances for living organisms (USEPA, 2012; Weissmannová and Pavlovský, 2017). A study carried out by Machunguene Jr *et al***,** (2024) in the industrial parks of Beluluane and Matola found that concentrations of Cr and other heavy metals are above international quality standards, hence this study aimed to assess the ecological and toxicological risk of *Arachis hypogaea* , *Vigna unguiculata* and *Zea mays* cultivated in soil contaminated by chromium (Cr) in order to provide data that could serve as a basis for the formulation of environmental and socio-ecological management instruments in Mozambique.

2. Methodology

The study was carried out in the greenhouse. To set up the experiment, the soil was collected from the Eduardo Mondlane University main campus, located 17km from the industrial areas of Beluluane and Matola, an area without industrial influence. The soil was collected at a depth of 20cm and processed for the presence of any roots and debris and was filled into polyethylene pots with a capacity of 2kg^{-1} (Bila *et al*., 2022; Nyika *et al*., 2019). Subsequently, a concentrated Cr solution was prepared, labeled and stored for later use (Li *et al*., 2015)**.** The control set of pots was maintained with tap water while the experimental set of pots was contaminated with Cr solutions at 10mg.kg^{-1} and 30mg.kg^{-1} and then each pot was labeled appropriately and the soil contamination by Cr was carried out seven (7) days after planting the previously germinated seedlings (Cardoso, 2018; Costa, 2015).

2.1. Plant Material

Seeds of *Arachis hypogaea*, *Vigna unguiculata* and *Zea mays* were germinated in *Petri* dishes and when the root appeared three (3) seedlings were planted per pot. Each treatment had seven (7) replications, and a causal experimental design was followed, 60 days later, the plants were harvested (Costa, 2015; Zakir, 2017). The plants were separated into roots and leaves, weighed and dried in a forced circulation oven at 65 ^{o} C, until constant mass. Then the dry weight was recorded and crushed in a Willey, SMC- Hammer sample mill. Mill, 220Hz50 and stored in ziplocks (Cardoso, 2018)**.**

2.1.1. Soil physicochemical analysis

After 60 days of experiment, the soil was harvested, stored in paper envelopes and dried in a forced circulation oven at 65ºC until constant mass. After drying, it was sieved with a 2mm sieve, then weighed on an analytical balance (0.0001g precision) and the weights were expressed in grams. For physicochemical analyzes the following methods were used: to determine the pH, 10g of soil were weighed in an Erlenmeyer flask and 25mL of KCl were added. The solution was shaken for 60s in an orbital shaker and left to rest for 1h. Then the pH of the solution was determined using the multiparameter meter-AK151-AKSO (Adimalla *et al*., 2019; Edogbo *et al.*, 2020). For determining the electrical conductivity, 10g of soil were weighed in an Erlenmeyer flask to which 25mL of deionized water were added. Then the solution was shaken for 60s in an orbital shaker and filtered through a Whatman No. 1 filter paper (Zhu *et al*., 2018)**.** After that, the electrical conductivity was measured using the multiparameter meter-AK151-AKSO (Weissmannová and Pavlovský, 2017)**.** The organic matter content (% OM) were determined through the Walkley-Black method. For exchangeable bases, an extraction with ammonium acetate and determination of Ca^{2+} and Mg^{2+} by titration with EDTA were performed. Na^{+} and K^{+} were determined using a flame spectrometer (Sellecta UV-3100) (Zakir, 2017)**.** The phosphorus content (%P) was determined through the Olsen's method (Adimalla *et al*., 2019)**.** For texture, soil mechanical analysis was performed using the pipette method and texture classification was obtained based on the limits of the Atterberg scale (Edogbo *et al*., 2020)**.**

2.1.2. *Stoma density*

To quantify the stomatal density, the printing methodology proposed by Bacelar *et al***,** (2004) was adopted to make stomatal impressions, a layer of enamel (colorless) was applied to the abaxial surface of each leaf measuring 1cm^2 and waited until it dried, subsequently the enamel was removed and microscopic preparations were made. The stomatal density was calculated based on an area of 1.76 mm^2, which corresponds to the area observable with a 10x objective (GTAC, 2011)

2.1.3. *Determination of photosynthetic pigments*

The quantification of photosynthetic pigments was determined using Lichtenthaler's, (1987) adjustments methodology. Leaf discs measuring 1.5 cm from the third pair of leaves, starting from the apex, were cut with the aid of pruning shears, macerated in a mortar with the addition of washed sand, with as little light as possible to avoid photooxidation of the pigments. After maceration, the material was filtered through 80g/m^2 qualitative filter paper with the aid of a 60 mm glass funnel, in a 25 ml volumetric flask, previously wrapped in aluminum foil. At the end of filtration, the volume of the bottle was completed with 80% acetone. After extraction, absorbance readings were taken on a spectrophotometer model DLAB SP-UV1100 at wavelengths of 663, 647 and 470nm, for chlorophyll *a*, chlorophyll *b* and carotenoids. The concentration of photosynthetic pigments was calculated using the equations described by Lichtenthaler, (1987).

$$\mathbf{Cl}\boldsymbol{a} = \mathbf{13.36 \times A - 5.19 \times A} \qquad (1)$$

$$\mathbf{Cl}\boldsymbol{b} = \mathbf{27.43 \times A - 8.12 \times A} \qquad (2)$$

$$\boldsymbol{\beta - caroteno} = \left[\mathbf{(1000 \times \boldsymbol{A}470) - (2.05 \times \boldsymbol{Cla}) - \left(\frac{Cla\boldsymbol{b}}{245}\right)}\right] \qquad (3)$$

2.1.4. *Chemical analyses of heavy metal*

Plant samples, as well as soil, were subjected to the acid digestion process to extract the mineral content Xiao *et al*., (2018) total of 1.0g of the soil and plant samples were weighed. For soil samples, 6mL of HCl 37%, 12mL of NHO_3 55%, and 18mL of HF 48% were added. For plant samples, 15mL and 5mL of NHO_3 55% and $HClO_4$ 70%, respectively were added. After adding the acids, the samples were heated to 180°C until the volume reduced to 1mL (Wang *et al*., 2021; Zakir, 2017). Once the digestion of soil and plant samples was completed, they were filtered through a Whatman No. 1 filter paper into 50mL volumetric flasks, and the volume was completed with deionized water. Subsequently, 15mL of the resulting solution were transferred from the 50mL volumetric flasks to test tubes and placed in the Inductively Coupled Plasma - Optical Emission Spectroscopy (ICP-OES) ICPE-9800 Series: Shimadzu SOPS, for the quantification of heavy metals, under the following conditions: the output power of the ICP-OES system and the radio frequency generator were 1200W and 40MHz, respectively. The nebulizer argon gas flow rate was 10.00L min−1, the auxiliary argon gas flow rate was 0.6L min−1 and the plasma argon gas flow rate was 0.7L min^{-1} (Weissmannová and Pavlovský, 2017; Zakir, 2017; Zhu *et al*., 2018).

The quality control of the ICP-OES analyses was performed using chemical quality analytical methods, as well as the incorporation of duplicate samples and blanks into the analytical process. The equipment was sterilized and protected against contamination. Pre-cleaning of the glassware was carried out properly, immersing all containers used in 10% NHO_3 for 24h, after which they were carefully rinsed with double-distilled water and dried in an oven at 105°C. Blanks were analyzed in the same way as samples. A recovery percentage greater than 95% was achieved for each of the elements with linear calibration curves (R^2 of 0.9978).

2.1.5. Bioconcentration and translocation factor

The bioconcentration factor (BF) is a ratio of the metal concentration in specific tissues (root, stem, leaf and fruit) to the concentration in the surrounding environment (soil or rhizosphere) and the translocation factor (TF) is the plant's ability to capture and distribute the metal throughout its tissues (Souza *et al*., 2019; Khalid *et al*., 2017).The BF as well as the TF were calculated using the equations of Arumugam *et al*, (2018).

$$\mathbf{BF} = \frac{\mathbf{C}_{leaf}}{\mathbf{C}_{roots}} \qquad (4)$$

$$\mathbf{TF} = \frac{\mathbf{C}_{pant}}{\mathbf{C}_{soil}} \qquad (5)$$

Where C_{leaf}, C_{roots}, and C_{soil} are the concentrations of Cr in plants tissues and soils samples (Souza *et al*., 2019; Khalid *et al*., 2017). Factors higher than 1 indicate the plant's ability to accumulate heavy metals (Arumugam *et al*., 2018).

2.2. Ecological risk assessment

It is a scientific process that determines the likelihood of damage to plants and animals that may be exposed to hazardous substances from a contaminated site (EPA, 2017).

2.2.1. Pollution Index

The equation for calculating the pollution index (PI) represents the ratio between the metal concentration in the soil divided by the quality value for a given metal. Mozambique does not have standard soil quality values for heavy metals, so the quality values of South African legislation were adopted.

$$\boldsymbol{PI} = \frac{\mathbf{C}_{\boldsymbol{Cr}}}{\mathbf{RfVo}} \qquad (6)$$

Where PI is the pollution index, $\boldsymbol{C_{Cr}}$ corresponds to the Cr value in the soil and the RfVo the standard soil quality value for Cr (6.5mg/kg) (DEA, 2012; Nyika *et al*., 2019)**.** The PI can be defined as follows: $PI \leq 1$ (negligible risk), $1 < PI \leq 2$ (low risk), $2 < PI \leq 3$ (moderate risk), $3 < PI \leq 5$ (high risk) and $PI > 5$ (extreme risk) (Qi *et al.*, 2020; Zhu *et al*., 2018).

2.2.2. Potential ecological risk

The Hakanson method was adopted to assess the potential ecological risk of soil contamination by Cr, using the equation:

$$\boldsymbol{E_i} = \sum \boldsymbol{T_i} \times \frac{\boldsymbol{C_i}}{\boldsymbol{C_{0i}}} \qquad (7)$$

where E_i is the potential ecological risk caused by a metal, T_i is the toxic response factor for a given metal (Cr = 2), C_i is the concentration of a given metal in the soil and C_{oi} is the background concentration (Cr = 0.76 mg kg $^{-1}$). The E_i can be defined as follows: $E_i < 40$ (negligible risk), 40–80 (low risk), 80–160 (moderate risk), 160–320 (high risk) > 320 (extremely high risk) (Qi *et al*., 2020; Rauf *et al*., 2021; Soleimani *et al*., 2023; Zhu *et al*., 2018).

2.3. Human risk assessment

It is a comprehensive assessment of the safety of a product based on its composition, materials, and intended uses (Park and Lek, 2015). To assess the risk to human health from consuming contaminated crops, the Estimated Daily Consumption (EDC) and the Hazard Quotient (HQ) were calculated using the Eqs (8), (9) respectively.

2.3.1. Estimated daily intake

$$\mathbf{EDI} = \frac{\mathbf{C \times dIR}}{\mathbf{BW}} \qquad (8)$$

Where C (mg kg $^{-1}$) is the concentration of the target element in the edible parts of the vegetable; dIR (daily intake rate) (kg day^{-1}) is the average daily consumption of vegetables; BW(kg) the average value of body weight. The dIR was established at 0.8kg.day^{-1} according to Machunguene Jr *et al*, (2024). The value of average body weight was defined at 67.83kg for adults (20-40 years old) in Mozambique (Mohammed *et al*., 2021; Olawale *et al*., 2023; Tchamo *et al*., 2016)**.**

2.3.2. Hazard Quotient (HQ)

$$\mathbf{HQ} = \frac{\mathbf{ECD}}{\mathbf{RfDo}} \qquad (9)$$

ECD is the estimated daily consumption and RfDo is the reference dose according FAO (2001) and WHO (2011) (1.3 for leaves and 0.003 for tubers).

The Chronic Daily Intake and lifetime incremental cancer risk equations were based on the model established by the United States Environmental Protection Agency (USEPA, 2014).

2.3.3. Chronic Daily Intake

$$\mathbf{CDI} = \frac{\mathbf{C \times DI \times ABS \times EF \times EP}}{\mathbf{BW \times AT}} \qquad (10)$$

Where C (mg.kg^{-1}) is the concentration of the heavy metal; DI DoRf in mg.kg^{-1} for leaves Cr=1.3 and DoRf mg.kg^{-1} for roots and tubers Cr (0.003) the daily average intake reference; ABS (0.001, unitless) is the ingestion factor; FE (365 days/year^{-1}) represents the annual exposure frequency; EP (70 years) is the exposure time; BW (67.83Kg/person^{-1}) is the Body Weight and AT (25,550 in days) is the average time (Miletić *et al*., 2023; Olawale *et al*., 2023; Sharma *et al*., 2021).

2.3.4. Lifetime incremental cancer risk

The ILCR is defined as the probability of a person developing any cancer over a lifetime as a result of 24h per day exposure to a given daily amount of a carcinogenic element for seventy years (Gramlich *et al*., 2018; Miletić *et al*., 2023; USEPA, 2004)**.**

$$\mathbf{ILCR} = \mathbf{CDI} \times \mathbf{CSF} \qquad (11)$$

Where CDI is a chronic daily intake and CSF is the cancer slope factor, which is defined as the risk generated by a lifetime average amount of 1mg kg^{-1} day^{-1} of a carcinogen chemical and is contaminant specific. The FIC value for Cr is Cr=0.5 (Miletić *et al*., 2023) (Miletić *et al*., 2023)**.** The allowable limit varies from 10^{-6} - 10^{-4} for a single carcinogenic element (Miletić *et al*., 2023; USEPA, 2004).

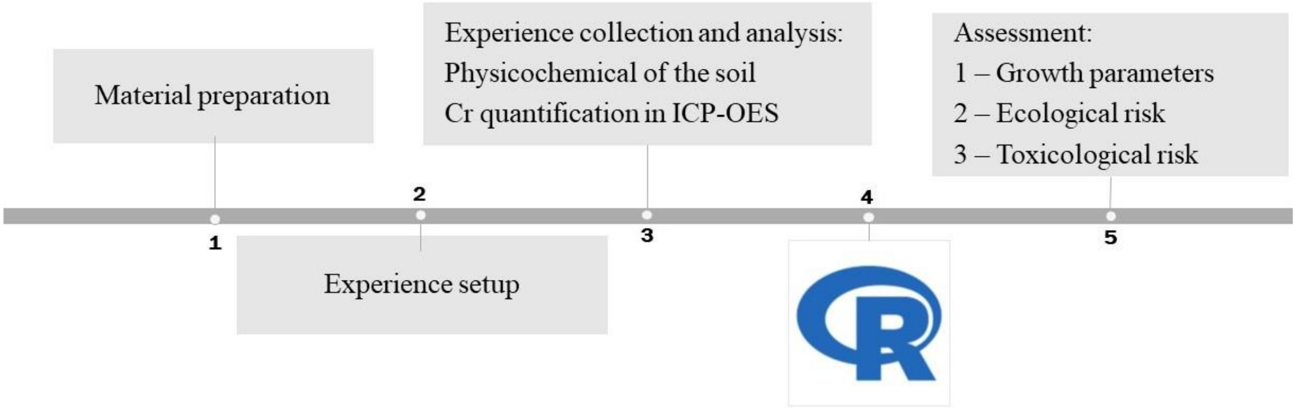

Figure 1: Flowchart of the conducted study.

3. Data analysis

Data normality and homoscedasticity were assessed through the Shapiro-Wilk test and the Levene test, respectively. Correlation analyses between soil and crop heavy metal concentrations were conducted using Pearson's correlation coefficient. All analyses were performed at a 5% significance level with RStudio version 2023.12.1+402.

4. Results and Discussion

4.1. Soil physicochemical characterization

In this study pH (table 1) of the soil was classified as neutral with values between 6.6 and 7.6. For Kim and Kumar Kim *et al*., (2019) under these pH conditions, there are bioavailable free ionic forms of Cr, whose translocation is mediated by coulomb forces. According to (Alderman *et al*, (2018) and Shanker *et al*, (2005) the bioavailability of heavy metals for plants is strictly related to their mobility in the soil, which is dependent on soil pH, a factor also responsible for the translocation of metals up to plant tissues. Meanwhile, Adamczyk-Szabela and Wolf, (2022) describe the pH range from 6 to 12 as being the one with the greatest absorption of metals. However, most micronutrients are better available to plants grown in acidic soils than in neutral or alkaline conditions (Gabriel *et al*., 2019).

Table 1: Physicochemical characteristics of the soil where the three study crops were grown

		Arachis hypogaea soil			***Vigna unguiculata soil***			***Zea mays soil***		
	Control	$10 mg.kg^{-1}$	$30 mg.kg^{-1}$	p-Value	$10 mg.kg^{-1}$	$30 mg.kg^{-1}$	p-Value	$10 mg.kg^{-1}$	$30 mg.kg^{-1}$	p-Value
pH (H_2O)	7.31^{a}	7.41^{a}	7.31^{a}	0.223	6.95^{ab}	7.64^{c}	0.0009	6.65^{bc}	6.87^{bc}	0.0005
Cation Exchange Capacity (Cmol kg^{-1})	3.06^{a}	2.57^{a}	2.58^{a}	0.18	2.93^{a}	2.54^{a}	0.254	2.85^{a}	3.72^{a}	0.848
Ca^{2+} (Cmol kg^{-1})	1.98^{a}	1.83^{a}	1.99^{a}	0.554	2.29^{a}	2.01^{a}	0.237	2.03^{a}	1.87^{a}	0.863
K^{+} (Cmol kg^{-1})	0.06^{a}	0.06^{a}	0.06^{a}	0.975	0.09^{bc}	0.08^{bc}	0.01	0.14^{a}	0.12^{a}	0.27
Na^{+}(Cmol kg^{-1})	0.81^{a}	0.35^{a}	0.31^{a}	0.195	0.32^{a}	0.26^{a}	0.15	1.18^{a}	1.19^{a}	0.562
Mg^{2+}(Cmol kg^{-1})	0.25^{a}	0.32^{a}	0.24^{a}	0.212	0.23^{a}	0.19^{a}	0.347	0.25^{a}	0.34^{a}	0.502
Electrical Conductivity (mS cm^{-1})	0.11^{a}	0.16^{a}	0.16^{a}	0.481	0.22^{ab}	0.14^{ac}	0.003	0.18^{a}	0.23^{ab}	0.01
%P	7^{a}	8^{a}	6.4^{a}	0.733	8.8^{a}	7.2^{a}	0.183	4.87^{a}	6.13^{a}	0.285
%Organic matter (OM)	0.59^{a}	0.72^{a}	0.78^{a}	0.883	0.82^{a}	0.74^{a}	0.857	0.79^{a}	0.73^{a}	0.832
%Loam	2.48^{a}	2.19^{a}	2.15^{a}	0.147	2.7^{a}	2.78^{a}	0.392	2.59^{a}	2.76^{a}	0.466
%Clay	6.03^{a}	$8.66b^{c}$	8.57^{bc}	0.012	8.21^{bc}	9.27^{bcd}	0.0001	8.65^{bc}	8.33^{bc}	0.008
%Coarse sand	33.93^{a}	38.36^{a}	37.05^{a}	0.349	36.81^{a}	37.23^{a}	0.561	32.44^{a}	32.98^{a}	0.869
%Fine sand	57.56^{a}	50.79^{a}	52.23^{a}	0.116	52.27^{a}	50.72^{a}	0.151	56.33^{a}	55.92^{a}	0.838
Texture						Sandy loam				

The values represent the mean of 9 plants ±SD. Values with different letters ab/c horizontally demonstrate significant differences between the means.

A few cation exchange capacity (Table 1) was verified in the soil while the electrical conductivity was low according to the Varennes model (2003). However, significant differences were observed in *V. unguiculata* soil as well as in *Z. mays* soil. For Zhu *et al,* (2018) and Adamczyk-Szabela and Wolf (2022) in these conditions there is an increase in the bioavailability of elements that were observed in this study. In all treatments, Cr was translocated from the soil to the root, and from the root to the shoots. According to Shanker *et al,* (2005) Singh *et al,* (2015) and Adamczyk-Szabela and Wolf (2022) the Cr absorption occurs through transporters used for the absorption of essential elements for plant metabolism. Sharma *et al.* (2020) assumes that the uptake of Cr(III) in plants occurs through a passive mechanism, however, Cr(VI) cross the plasma membrane through the active process that involves transporters of essential phosphate or sulfate anions. These assumptions may explain the low values observed in cation exchange bases, because when Cr binds to active sites for essential elements, it restricts the absorption of nutrients in the soil, forming insoluble compounds (Anitha *et al.*, 2012; Santos *et al.*, 2018). However, Sharma *et al,* (2020) observed that excessive Cr reduces the absorption of essential minerals such as Fe, Mg, P, and Ca, masking the sites of absorption and adsorption, forming insoluble complexes and gradually decreasing the absorption of micronutrients such as Zn, Fe, Mn, and macronutrients such as K, P, and N.

Sharma *et al,* (2020) also observed that plant roots secrete several organic acids, such as citrate and malate, which modify the solubility of metals present in the soil. Similar results were reported by Srivastava *et al,* (2021), who observed an increase in Cr in tomato plants due to the presence of citrate, aspartate, and oxalate, having converted inorganic Cr into organic complexes readily available for absorption by the plant. This study also suggests that the same thing happened, as the higher the concentration of Cr in the soil, the higher the concentration of Cr in plant tissues.

The soil OM was classified as low in all treatments. These values are in line with Gajaje *et al,* (2024) and Ferreira *et al,* (2023) and are probably due to soil texture. According to Li *et al,* (2015) soil texture plays a fundamental role in the mineralization of OM through direct interactions with minerals and indirect effects on soil moisture. It is also known that sandy soils have less OM than clay soils because of its main properties including low water retention and low cations retention and exchange (Yost and Hartemink, 2019). The composition of the soil texture was between 32% and 38% for coarse sand and between 50% and 57% for fine sand. Under these conditions, the availability of heavy metals may be high Edogbo *et al*, (2020) due to low permeability, great water retention capacity, and great ion adsorption (Edogbo *et al*, 2020; Silva *et al*., 2018).

In *A. hypogaea* there was a reduction of 40 and 36% in root biomass (figure 2A), 23 and 38% in leaf biomass (figure 2B) in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. In *V. unguiculata*, was an increase of 24% in root biomass in treatment with 30mg.kg^{-1} of Cr and a reduction in leaf biomass of 1 and 17% in treatments with 10 and 30mg.kg^{-1}of Cr, respectively. In *Z. mays* there was a 33% reduction in root biomass in treatment with 30mg.kg^{-1} of Cr and in leaf there was an increase of 55% in treatment with 10mg.kg^{-1} of Cr and a reduction of 13% in treatment with 30mg.kg^{-1} of Cr. Only leaves of *Z. mays* showed significant differences between treatments. These results corroborate with those obtained by Reis, (2019) and Shanker *et al*, (2005) according to which high concentrations of Cr affect cell metabolism, reducing their division as a consequence of the reduction in palisade parenchyma and an increase in the number of vacuoles along the xylem and phloem walls. Sousa e Santos (2018) studying Cr in mineral nutrition and Chibuzor *et al*, (2018) studying Chromium (III) in soil microbial activities and phytoremediation potentials of *A. hypogaea* and *V. unguiculata* observed that Cr concentrations greater than 20mg/L decrease the dry matter production as well as the absorption of macronutrients (N, P, K, Ca and Mg). The Cr translocation and subsequent accumulation in plants causes changes in plant tissues at physiological and biochemical level (Shanker *et al.*, 2005) manifested through a reduction in biomass (Reis, 2019). According to the same author, this reduction is due to the indirect effects of damage caused to the root system and the absorption and translocation of water and nutrients.

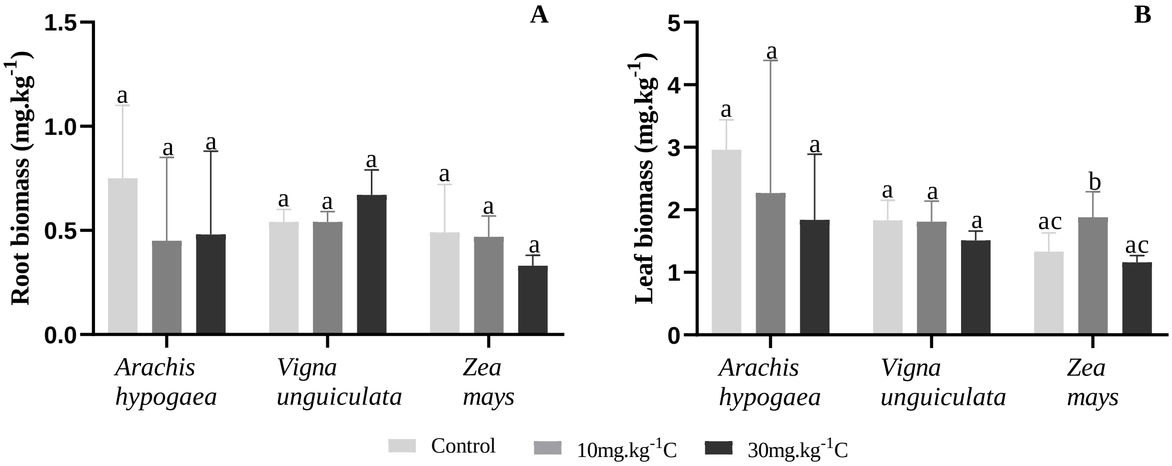

Figure 2: Biomass of roots (A) and leaves (B) of *Arachis hypogaea, Vigna unguiculata* and *Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr. The bars represent the mean of 9 plants ±SD. Bars with different letters a b / c horizontally demonstrate significant differences between the means.

4.2. Photosynthetic pigments

In the chlorophyll *a* (figure 3A) in *A. hypogaea* there was an increase of 37% in the treatment with 30mg.kg^{-1} and in *V. unguiculata* there was an increase of 12% in both treatments. As for chlorophyll *b* (figure 3B) in *A. hypogaea,* an increase of 4 and 10% was observed in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. On the carotenoids (figure 3C) in *A. hypogaea* there was an increase of 7 and 6% in treatments with 10 and 30mg.kg^{-1} of Cr, respectively, while in *V. unguiculata* there was an increase of 5 and 3% in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. According to Srivastava *et al,* (2021) studying the stress caused by Cr in *V. unguiculata* plants, observed a decrease in chlorophyll content in comparison with control treatment, and suggested that gas exchange parameters and chlorophyll *a*, *b* and carotenoids were reduced with increasing concentration of Cr in the soil. Razzaq *et al*, (2024) studying the toxicity of Cr in *Z. mays* reached the same conclusions. Chibuzor *et al.* (2018) studying Chromium (III) in soil microbial activities and phytoremediation potentials of *A. hypogaea* and *V. unguiculata* also observed one decrease in chlorophyll content as Cr increased. This difference to the results obtained in previous studies with *V. unguiculata* under water stress conditions may be related to the varieties used in the study (Martins *et al.,* 2014). According to Taiz *et al*., (2017) the response to abiotic stress, such as heavy metals such as Cr, also depends on the plant genotype.

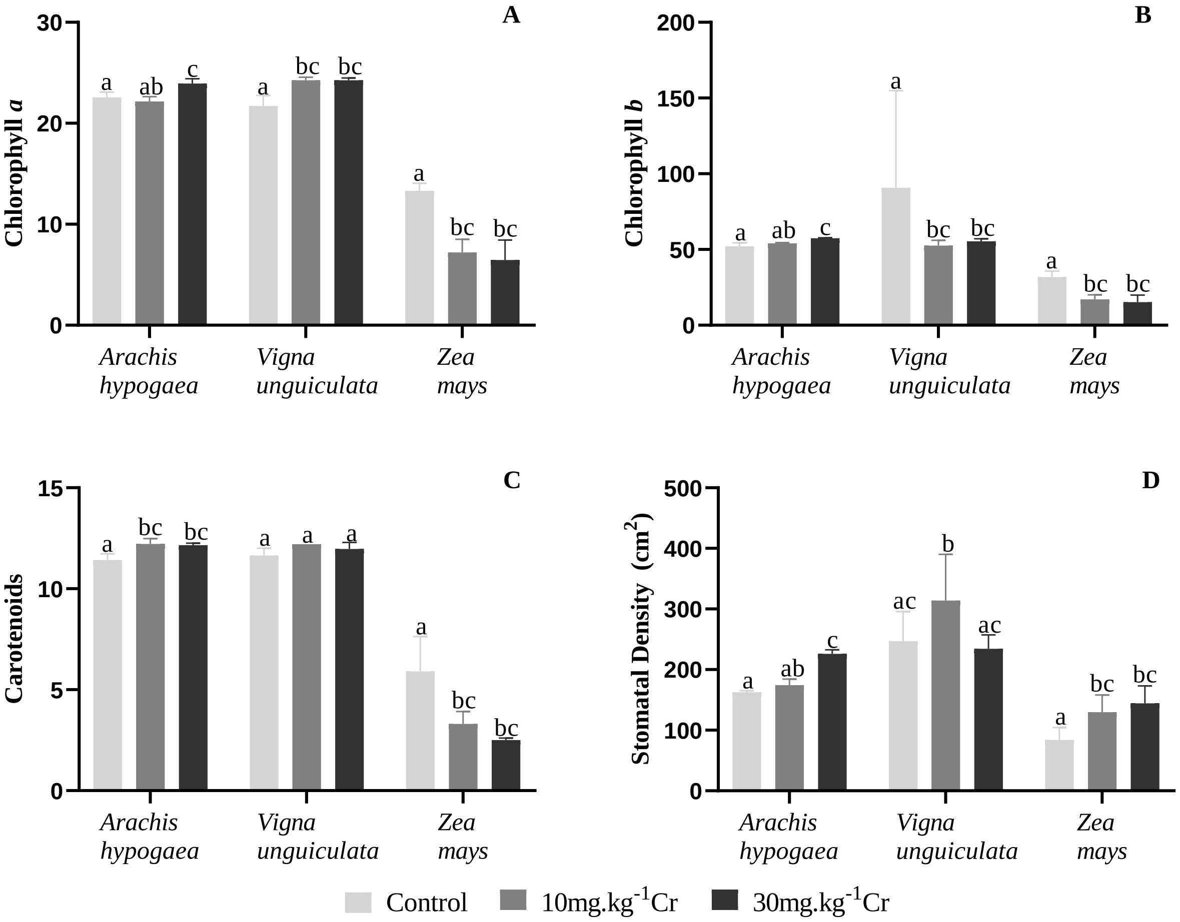

Figure 3: Chlorophyll *a* (A), Chlorophyll b (B), Carotenoids (C) and Stomatal Density (A) in *Arachis hypogaea*, *Vigna unguiculata* and *Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr. The bars represent the mean of 9 plants ±SD. Bars with different letters b /c horizontally demonstrate differences between the means.

In *A. hypogaea* there was an increase of 7 and 39% in stomatal density (figure 3D) in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. While in *V. unguiculata* the increase was 27% in the treatment with 10mg.kg^{-1} of Cr and in *Z. mays* it was 55 and 72% in the treatments with 10 and 30mg.kg^{-1} of Cr, respectively. All species show significant differences in stomatal density between treatments. The Cr translocation level and subsequent accumulation in plants cause changes in plant tissues at a physiological (Srivastava *et al*., 2021). According to Guo *et al*., (2023) to maintain physiological and metabolic functions, plants improve their tolerance to heavy metals by increasing the number of stomata, which increases CO_2 absorption and water availability. Guo *et al*, (2023) concluded that an increase in the number of stomata is one of the first lines of defense against heavy metal stress, which was also observed in this study. However, Mohammed *et al,* (2021) studying the physiological and physicochemical effect of chromium (VI) on the nutritional quality of *Z. mays* observed that the presence of Cr reduced stomatal density.

4.3. Heavy metal concentrations in soils and crops

In *A. hypogaea*, there was an increase in the concentration of heavy metals in the roots by 60 and 280% and by 50 and 850% in the leaves in treatments with 10 and 30mg.kg^{-1} of Cr, respectively (table 2). Significant differences were observed between treatments. In *V. unguiculata* there was an increase of 433 and 1133% in roots and 600 and 1650% in leaves in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. Significant differences were observed between treatments. In *Z. mays* in the roots there was an increase of 33 and 500% and of 0% and 700% in the leaves in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. These results suggest the existence of translocation mechanisms between the species under study. However, a specific mechanism for Cr absorption by plants has not yet been identified as it does not play any vital role in plant metabolism Ali *et al,* (2023). However, it is known that Cr can be transported from the root to the aerial parts involved in transporters of essential nutrients, such as phosphate and sulfate. Cr uses iron (Fe) and sulfur (S) channels for upward translocation, which causes competition between these metals Ali *et al,* (2023). Singh *et al.* (2015) observed that sulfur hyperaccumulating families such as Brassicaceae accumulate high levels of Cr in shoots, meaning that Cr is translocated from the root to the shoot through the sulfur uptake and translocation mechanism.

Table 2: Concentration of heavy metals in the tissues of *Arachis hypogaea, Vigna unguiculata and Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr.

		Arachis hypogaea		
	Control	**10mg.kg^{-1} Cr**	**30mg.kg^{-1} Cr**	**p-Value**
Roots	0.05 ± 0.02^{a}	0.08 ± 0.06^{b}	0.19 ± 0.02^{ab}	0.0132 *
Leaves	0.02 ± 0^{a}	0.03 ± 0^{b}	0.19 ± 0.05^{ab}	0.000343 ***
Soil	NA	0.01 ± 0^{a}	0.05 ± 0^{b}	2.89e-05 ***
		Vigna unguiculata		
Roots	0.03 ± 0.01^{a}	0.16 ± 0.17^{b}	0.37 ± 0.26^{c}	0.0529*

Leaves	0.02 ± 0.01^{a}	0.14 ± 0.11^{c}	0.35 ± 0.13^{c}	0.00703**
Soil	0.02 ± 0^{a}	0.04 ± 0.01^{b}	0.07 ± 0^{c}	2.79e-05***
		Zea mays		
Roots	0.03 ± 0.01^{a}	0.04 ± 0.02^{ab}	0.18 ± 0.04^{c}	0.000459 ***
Leaves	0.02 ± 0.01^{a}	0.02 ± 0.01^{a}	0.16 ± 0.03^{b}	0.000109 ***
Soil	NA	0.03 ± 0^{a}	0.14 ± 0^{b}	7.04e-07 ***

Values with different letters ab/c horizontally demonstrate significant differences between the means. *** represents extremely significant differences, * represents differences in 95% of significant level. The concentration values of different metals in soil and plants represent the average of 9 plants ±SD. NA is not available (below the detection limit).

In *A. hypogeae* there was a positive correlation (table 3), in stomatal density and carotenoids in the control treatment, in chlorophyll *a* and *b* in the treatment with 10mg.kg^{-1} of Cr, and in leaf biomass, stomatal density and chlorophyll *a* and *b* in treatment with 30mg.kg^{-1} of Cr. In *V. unguiculata* there was a positive correlation in chlorophyll *a* in the control treatment, in the biomass of roots and leaves, in stomatal density and in Chlorophyll *a* and *b* in the treatment with 10mg.kg^{-1} of Cr, and root biomass, Chlorophyll *a* and *b* and carotenoids in treatment with 30mg.kg^{-1} of Cr. In *Z. mays* there was a positive correlation in root biomass in the control treatment, in leaf biomass and chlorophyll *a* and *b* in the treatment with 10mg.kg^{-1} of Cr, and in leaf biomass and stomatal density in the treatment with 30mg.kg^{-1} Cr. These results corroborate those of Guo *et al*, (2023), Mohammed *et al,* (2021), Srivastava *et al*, (2021), Razzaq *et al*, (2024), Chibuzor *et al*, (2018) e Martins *et al*, (2014) who suggested that plants develop compensatory mechanisms to cope with heavy metal stress and these mechanisms vary according to the species, variety and tissue of the plant.

Table 3: Correlation between physiological and biochemical parameters and the concentration of heavy metals

R ^{2}	**Biomass of roots**	**Biomass of leaves**	**Stoma density**	**Chlorophyll *a***	**Chlorophyll *b***	**Carotenoids**
			Arachis hypogaea			
Control	-0.939874	-0.787637	0.927714	-0.737035	-0.954454	0.999954
10mg.kg^{-1} Cr	-0.822279	-0.892792	-0.636233	-0.776460	0.991624	0.765519
30mg.kg^{-1} Cr	-0.200793	0.755961	0.801666	0.634635	0.763712	-0.418954
			Vigna unguiculata			
Control	-0.979696	-0.669597	-0.231121	0.933574	-0.988154	-0.948446
10mg.kg^{-1} Cr	0.993007	0.990207	0.452456	0.993495	0.764268	-0.846137

30mg.kg^{-1} Cr	0.841037	-0.663479	-0.999969	0.869611	0.362276	0.946284
			Zea mays			
Control	-0.517074	0.204275	-0.309356	-0.315601	-0.130609	-0.028810
10mg.kg^{-1} Cr	0.972234	-0.963063	-0.808373	0.168268	0.168268	-0.243809
30mg.kg^{-1} Cr	-0.797880	0.975954	0.987348	-0.035680	-0.035680	-0.999923

4.4. Bioconcentration and translocation factor

All species studied showed bioconcentration factors greater than one, except for *A. hypogaea* and *Z. mays* in the control treatment in which it was not possible to determine this factor (figure 4A). For the translocation factor (figure 4B), all species studied showed values below one, except for *A. hypogaea* in the treatment contaminated with 30mg.kg $^{-1}$ of Cr which had a translocation factor equal to one. These results corroborate those obtained by Khalid *et al*, (2017) who suggest that plants with a bioconcentration and/or translocation factor greater than one are hyperaccumulators, as they can tolerate Cr through chelation by biotransformation with reductants, high affinity ligands such as amino acids, organic acids, peptides and polypeptides and compartmentalization in the vacuole or cytoplasm.

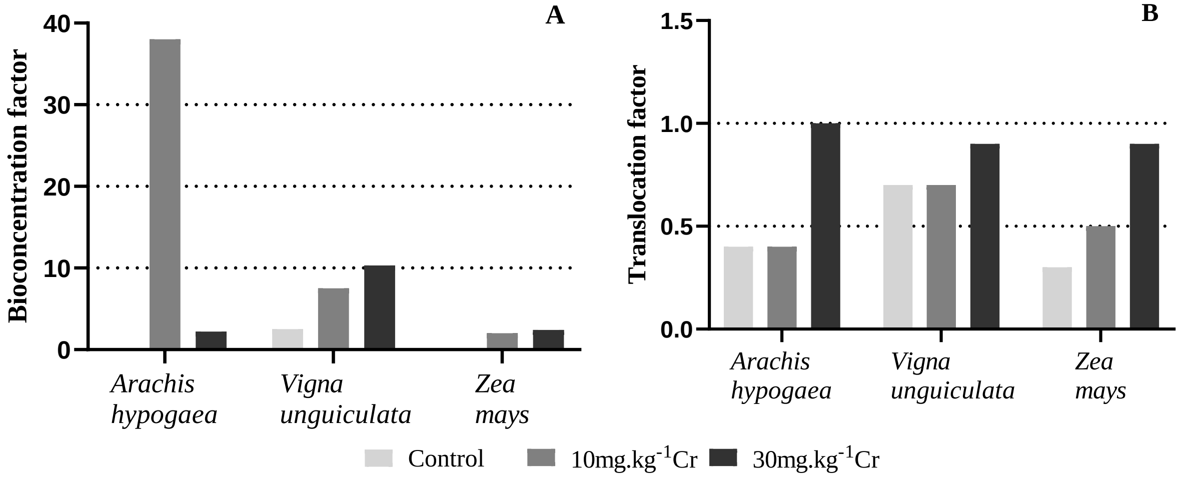

Figure 4: Bioconcentration factor (A) and Translocation factor (B) in *Arachis hypogaea*, *Vigna unguiculata and Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr.

The accumulation of Cr and other heavy metals by plants depends on different factors, such as the mobilization of metals in the soil from the rhizosphere, the increase in absorption and translocation to the shoot tissues through the xylem, chelation and detoxification of metals within plant cells (Guo *et al.*, 2023). Unlike what is reported in other studies about the low accumulation of Cr in plant tissues, the results of this study suggest that the three species studied accumulate Cr in their tissues. Similar results were observed by Chen *et al*, (2022) and suggest that transfer of Cr to vegetables tissues increases toxic exposure in each trophic level and, in last analysis, for human beings. Xu *et al*, (2023) also observed similar results and suggest that Cr accumulation and translocation in plants varies between plant species are influenced by the genetic and morphological characteristics of each plant. They also point out that several factors, such as metal concentration in the soil, bioavailability, physical and chemical properties of the soil, affect the absorption and translocation of metals in plants.

4.5. Ecological risk assessment

The pollution index (figure 5A) in *A. hypogeae* was 0.002 and 0.008 for treatments contaminated by 10 and 30mg.kg^{-1} of Cr, respectively. In *V. unguiculata* they were 0.003, 0.006 and 0.01 in the control treatment, in treatments with 10 and 30mg.kg^{-1} of Cr, respectively. In *Z. mays*, pollution indices were 0.005 and 0.02 in treatments contaminated with 30mg.kg^{-1} of Cr, respectively. Similar results were observed by Chukwu and Oji, (2018) in Nigeria, having found a pollution index of 0.215. In another study by Chen et al. (2022) they also observed a pollution index less than one. These authors concluded that regardless of the source of Cr emissions, whether industrial or agricultural, when the pollution index is less than one, soil quality cannot be lost. Thus, the results of the pollution index (IP<1) demonstrated an unpolluted nature of the soil, even though it was contaminated with 30mg.kg^{-1} of Cr, a concentration four times greater than the reference value for soil quality (6.5 mg.kg $^{-1}$) (Nyika *et al.*, 2019; DEA, 2012). However, there is a potential effects of long-term exposure, mainly because Cr is non-biodegradable and its accumulation in soil can lead to bioaccumulation and loss of local biodiversity (Chukwu and Oji, 2018; Rauf *et al*., 2021)**.**

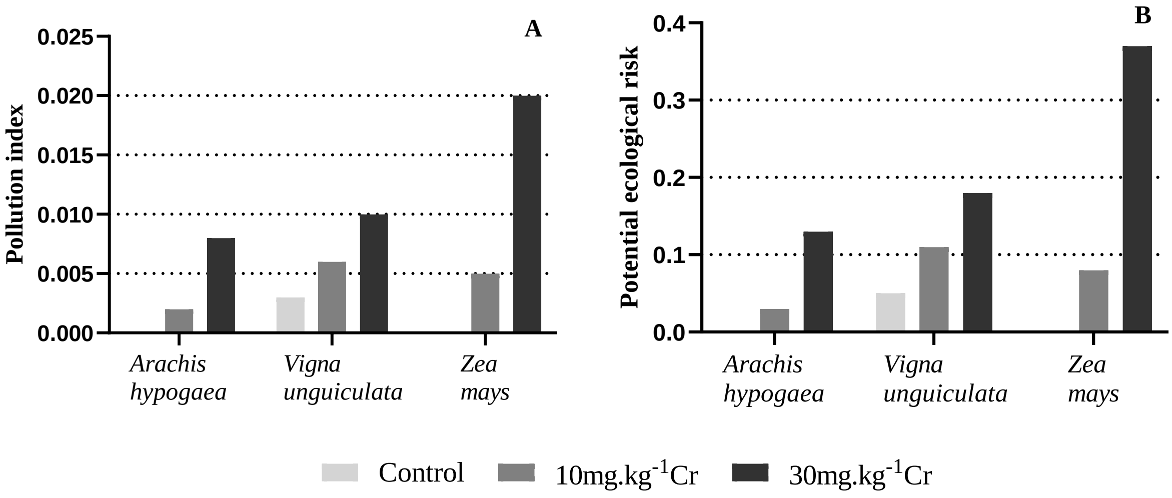

Figure 5: Pollution index **(A)** and Potential Ecological Risk **(B)** of Cr in the soil in treatment with 10 and 30mg.kg^{-1} of Cr.

Regarding the potential ecological risk (figure 5B), values of 0.03 and 0.13 were observed for *A. hypogaea* in treatments contaminated with 10 and 30mg.kg^{-1} of Cr, respectively. In *V. unguiculata*, potential ecological risk values of 0.05, 0.11 and 0.18 were found in control treatments and treatments with 10 and 30mg.kg^{-1} of Cr, respectively. In *Z. mays* the potential ecological risk was 0.08 and 0.37 in treatments contaminated with 10 and 30mg.kg^{-1} of Cr, respectively. These results corroborate what was observed by Sobhanardakani (2018) in the rural area of Hamedan in China, where concluded that with $E_i \leq 40$, the ecological risk can be neglected, not representing a danger to the local biodiversity. Ramos-Miras *et al*, (2020) also obtained $E_i \leq 40$ and suggested that their results indicate that there is negligible ecological risk. Ke *et al*, (2017) assessing the risk ecological of heavy metal at sediment surface of the Liaohe River, also came to the conclusion that with $E_i \leq 40$ the ecological risk is negligible. Qi *et al*, (2020) evaluating the ecological risk due to heavy metal contamination and Rauf *et al*, (2021) assessing the risk ecological of hexavalent chromium observed similar results and suggest that with $E_i \leq 40$ the ecological risk is negligible. These results suggest that thea potential negligible ecological risk is due to the role of the studied species in removing Cr from the soil, reducing its availability. As previously mentioned, these species hyperaccumulated Cr in their tissues.

4.6. Human risk assessment

In the daily intake rate (figure 6A), higher values were observed in the treatment with 30mg.kg^{-1} of Cr in *A. hypogaea* with 2.24, *V. unguiculata* with 4.36, and in *Z. mays* with 1.89 and 1.65 in the treatment with 10mg.kg^{-1} in *V. unguiculata*. In the hazard coefficient (figure 6B), higher values were observed in the treatment with 30mg.kg^{-1} of Cr in *A. hypogaea* with 1.72, *V. unguiculata* with 3.36, and in *Z. mays* with 1.45 and 1.27 in the treatment with 10mg.kg $^{-1}$ in *V. unguiculata* and the remaining values were less than one. Hazard coefficient values greater than one indicate a potential adverse health risk and the higher the coefficient, the greater the health risk (Li *et al*., 2015; Zhu *et al*., 2018). Risk assessment for human health involves determining the nature and magnitude of adverse health effects in humans (Mohammadi *et al.*, 2019). For heavy metal, a lifetime incremental cancer risk (ILCR) of less than 1×10^{-6} is considered insignificant and the risk of contracting cancer can be neglected, while an ILCR above 1×10^{-4} is considered harmful and the risk of cancer is significant.

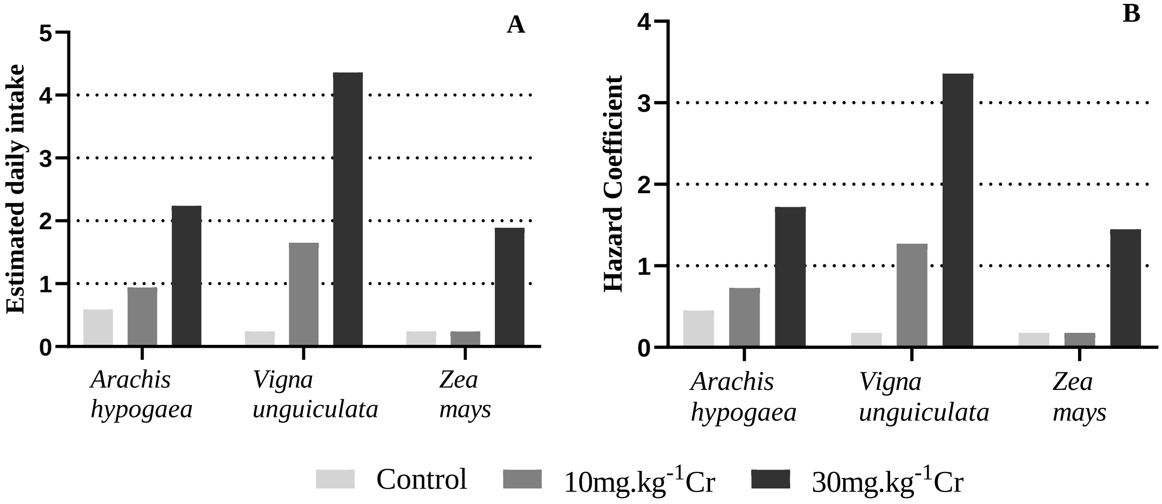

Figure 6: Daily Consumption Rate (A) and Hazard Coefficient (B) from the edible parts of *Arachis hypogaea*, *Vigna unguiculata* and *Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr.

In this study, all treatments for all species showed values below or within the permitted limits for lifetime incremental cancer risk (table 8), according to USEPA guideline values USEPA (Gabriel *et al*., 2019; Miletić *et al.*, 2023). These results are also within the limit values established by

South African legislation of 1×10^{-5} (Kamunda *et al.,* 2016). Similar results were also observed by Bello *et al,* (2019) evaluating the carcinogenic risks of exposure to heavy metals in artisanal gold mining in Nigeria and suggesting that it is unlikely that people can get cancer or any carcinogenic risks arising from foods with ILCR below 10^{-6}. Boluspayeva *et al,* (2023) evaluating the health risk of consuming vegetables contaminated by heavy metals in the industrial area of Kazakhstan obtained similar results. Miletić *et al*, (2023) studying exposure factors in the health risk assessment of heavy metals also observed that with ILCR below 10^{-6} there is no health hazard.

Tabela 4: Chronic daily intake and Lifetime incremental cancer risk (ILCR) from the edible parts of *Arachis hypogaea*, *Vigna unguiculata* and *Zea mays* in treatment with 10 and 30mg.kg^{-1} of Cr.

	Chronic Daily Intake		
	Arachis -Roots	*Vigna* -Leaves	*Zea* -Leaves
Control	9.6×10^{-07}	3.8×10^{-07}	3.8×10^{-07}
10mg.kg^{-1} Cr	1.5×10^{-06}	2.7×10^{-06}	3.8×10^{-07}
30mg.kg^{-1} Cr	3.6×10^{-06}	6.7×10^{-06}	3.1×10^{-06}
	ILCR		
Control	4.79×10^{-07}	1.92×10^{-07}	1.92×10^{-07}
10mg.kg^{-1} Cr	7.67×10^{-07}	1.34×10^{-06}	1.92×10^{-07}
30mg.kg^{-1} Cr	1.82×10^{-06}	3.35×10^{-06}	1.53×10^{-06}

5. Conclusion

This study was carried out to evaluate the ecological and toxicological risk of *Arachis hypogaea, Vigna unguiculata and Zea mays* grown in Cr-contaminated soil. The three species showed through growth parameters that they have physiological and biochemical mechanisms to accumulate Cr in their tissues, which suggests that they are Cr hyperaccumulators. They were able to extract Cr from the soil to the roots and from the roots to the aerial part, however soil contamination by Cr in concentrations of up to 30mg.kg^{-1} does not represent an ecological risk when plants have the potential to accumulate the metal in their tissues. Regarding toxicity, all species showed values within the allowed limits for incremental risk of contracting cancer throughout life. *Arachis hypogaea, Vigna unguiculata* and *Zea mays* can be grown in contaminated soil with up to 30mg.kg^{-1} of Cr and consumed without posing a danger to human health.

Limitações

A grande limitação enfrentada nesse estudo foi o alto custo das análises químicas em Espectrometria de Emissão Atómica por Plasma Acoplado Indutivamente (ICP-OES), tendo limitado o número de tratamentos, metais e espécies a testar.

Outra limitação foi a falta de laboratórios devidamente equipados para proceder com determinadas analises, a título de exemplo não foi possível fazer a especiação do Cr, bem como observar seu acúmulo a nível dos organelos celulares.

A falta de legislação nacional para a concentração de metais pesados, tanto no solo assim como em plantas alimentares, continua a ser um grande desafio, pois por falta desta, somos condicionados a utilizar legislações internacionais, que muitas vezes não refletem a realidade nacional.

Recomendações

Com base nos resultados deste estudo recomenda-se a realização de estudos semelhantes em áreas industriais em todo território nacional de modo se ter uma noção realista do risco ecológico e toxicológico. Recomenda-se também que Moçambique comece a trabalhar no sentido de ter sua própria legislação sobre a emissão de resíduos contendo metais pesados, uma vez que é signatário da UNO para os Objectivos de Desenvolvimento Sustentável, dada crescente explosão industrial no país.

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