Intercurrent Flaviviral Viremia and *Plasmodium vivax* and *Plasmodium ovale* Infections in Ill-Returned Travelers to Ontario

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INTRODUCTION

- Flaviviruses: transmitted to humans through infected bites of *Culex* spp. and *Aedes* mosquitoes[1]
- *Plasmodium vivax* and *plasmodium ovale*: spread by the bite of *Anopheles* mosquito[2]
- Flaviviral infection could precipitate a *P. vivax* or *P. ovale* relapse[3]
- Given overlap of epidemiological and clinical presentations of both flaviviral and malaria infections, diagnostic testing where malaria is confirmed or excluded, without subsequent flaviviral testing may mask true epidemiology of co-infections[4]

Objective: We aim to understand the incidence of intercurrent flaviviral infection in confir med *Plasmodium vivax* and *Plasmodium ovale* infection in whole blood specimens from ill returned travelers

METHODS

- DNA extracted from whole blood specimens and tested for malaria by microscopy and rapid diagnostic test (RDT) between 2006 and 2018 at Public Health Ontario Laboratory-Toronto[5,6]
- RNA extracted from *P. vivax* positive and *P. ovale* positive whole blood specimens and examined by real-time PCR (qPCR) for the following targets: flaviviruses (pan-FLAV) and dengue virus types 1-4 (DEN1, DEN2, DEN3, DEN4)[7-9]

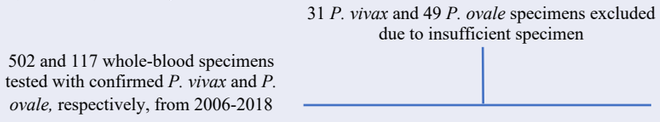

Figure 1: Workflow highlighting *P. vivax* and *P. ovale* confirmed diagnostic testing for intercurrent flaviviral infection using qPCR.

RESULTS

Total	*P. vivax* [n=471, (%)]	*P. ovale* [n=69, (%)]
Median age, years (range)	33.6 (1 month – 87.6 years)	26.7 (1.2 years – 60.9 years)
Median Parasitemia, % (range)	<0.1% (<0.1% - 2.5%)	< 0.1% (< 0.1% - 0.6%)
Sex Male Female Unknown	 297 (63.1%) 149 (31.6%) 25 (5.3%)	 40 (57.9%) 25 (36.2%) 4 (5.8%)
Travel History Yes Unknown	 169 (35.9%) 302 (64.1%)	 35 (50.7%) 34 (49.3%)

Table 1: Clinical and parasitological characteristics of *P. vivax* and *P. ovale* cases.

Location	*P. vivax* [n=471, (%)]	*P. ovale* [n=69, (%)]
Sub-Saharan Africa Nigeria	6 (1.3%) 1 (0.2%)	34 (49.3%) 23 (33.3%)
Indian Subcontinent India Pakistan	143 (30.4%) 97 (20.6%) 44 (9.3%)	1 (1.5%) NA 1 (1.5%)
Latin America Guyana	19 (4.03%) 11 (2.3%)	NA NA
Southeast Asia	1 (0.2%)	NA
Unknown	302 (64.1%)	34 (49.3%)

Table 2: Known travel history

qPCR Type	*P. vivax*	*P. ovale*
DENV qPCR	1/471 (0.2%)	0/69 (0%)
Flavivirus qPCR	1/471 (0.2%) untypeable	1/69 (1.41%) untypeable

Table 3: DENV and Flavivirus qPCR positive results.

DISCUSSION & CONCLUSIONS

- For *P. vivax,* both the Pan-FLAV and the Pan-DENV assays yielded a 0.2% positivity rate (1/471) each (Table 3) and for *P. ovale,* Pan-FLAV yielded a 1.41% positivity rate (1/68), while the DENV assays did not yield a positive result (Table 3)
- For *P. vivax,* type-specific real-time PCR revealed DEN1, detected on both Pan-FLAV and Pan-DENV assays
- *P. vivax* infections rates are highest from India, Pakistan and Guyana[10] and *P. ovale* infections are highest from Sub-Saharan Africa (Table 2)
- Intercurrent flaviviral viremia was noted in 0.2% of *P. vivax* specimens and 1.41% of *P. ovale* specimens, suggesting primary flaviviral infection could have triggered relapse of *P. vivax* and *P. ovale,* respectively
- Alternatively, co-infections may suggest primary infection with both organisms given the overlap of vector populations in these endemic areas
- Consideration of flaviviral co-infection should be given to *P. vivax* and *P. ovale* patients to appropriately manage clinical manifestations including deep thrombocytopenia, lymphopenia, and high yield arboviral symptomology including rash and retro-orbital headache[4]

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Rifampin-Ofloxacin-Minocycline (ROM) for the Treatment of Paucibacillary Leprosy: A Systematic Review

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Introduction

- Standard WHO multi-drug treatment (MDT) for leprosy consists of medications that are potentially harmful and cause a range of adverse systemic effects
- Paucibacillary leprosy, characterized by limited skin lesions and a low bacillary load, may be most amenable to a fluoroquinolone-based treatment protocol
- Monthly- or single dosing of ROM has emerged as a potential treatment option for leprosy, however, a synthesis of the evidence supporting ROM does not exist

Methods

- Abstracts reporting the efficacy & safety of monthly ROM treatment in paucibacillary leprosy in human patients were targeted using combinations of the search terms “ROM” & “Leprosy” from inception to March 2019
- During all phases of screening a tertiary arbitrator will mitigate any inclusion/exclusion discrepancies
- Inclusion Criteria: Systematic reviews, randomized controlled trials, clinical trials, cohort studies, observational studies, case-control studies, case series (N>5), non-English publications
- Exclusion Criteria: Case reports, case series (N<4)

Results

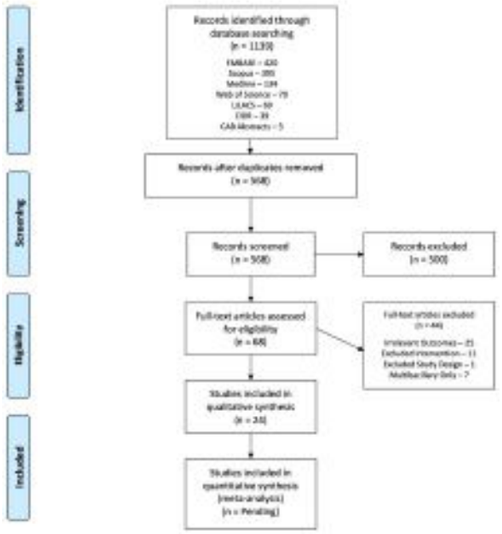

Figure 1. PRISMA Flowchart

Study	Country	Study Design	Sample Size, No.	Mean Age, y	Male, %	Follow-Up, (SD), mo	Diagnosis of Leprosy	# Lesions	Treatment	Comparator
[1]Alam et al., 2007	Bangladesh	Retrospective	270	-	-	96	Not reported	Single	ROM, single dose	No Comparator
[2]Babu et al., 1997	India	Randomized Control Trial	1483	23	42.28	12	Clinical	Single	ROM, single dose	WHO-MDT
[3]Desikan & Gupte, 2001	India	Randomized Control Trial	236	-	46.19	12-18	Clinical + Histological	2-3	ROM, single dose	WHO-MDT
[4]Deshmukj et al., 2003	India	Randomized Control Trial	32	-	75	6	Clinical + Histological	1-3	ROM, single dose	WHO-MDT
[5]Diniz et al., 2010	Brazil	Cohort	54	31	31.48	12	Clinical + Histological	Single	ROM, single dose	No Comparator
[6]Ebenezer et al., 1999	India	Case series	13	26 (11.4)	62	12	Clinical	1-3	ROM, single dose	No Comparator
[7]Emmanuel & Gupte, 2005	India	Randomized Control Trial	51	-	58.82	24	Clinical + Histological	2-3	ROM, single dose	WHO-MDT
[8]Ganapati et al., 1999	India	Case series	634	-	-	-	Clinical	2-5	ROM, single dose	No Comparator
[9]Girdhar et al., 2011	India	Randomized Control Trial	300	30.9 (16.2)	41	36.76 (14.8)	Clinical	Single	ROM, single dose	ROM + clarithromycin
[10]Gomes et al., 2008	Brazil	Cohort	259	32.4 (16)	38.2	36	Clinical + Histological	Single	ROM, single dose	No Comparator
[11]Kumar et al., 2015	India	Randomized Control Trial	268	-	37.7	60	Clinical	1-5	ROM, monthly	WHO-MDT
[11]Kumar et al., 2014	India	Cohort	289	41.6	61.8	12	Clinical	1-5	ROM, monthly	WHO-MDT
[12]Majumder et al., 2000	India	Clinical Trial	90	-	-	12	Clinical + Histological	Single	ROM, single dose	ROM, single dose + Convit vaccine*
[13]Mane et al., 1997	Senegal	Case series	220	-	60	12	Clinical + Histological	2-5	ROM, monthly	No Comparator
[14]Manickam et al., 2012	India	Randomized Control Trial	1526	27	47.5	36	Clinical	2-5	ROM, single dose	WHO-MDT
[15]Martelli et al., 2000	Brazil	No outcomes reported	259	32.4 (16.0)	38.22	-	Clinical + Histological	Single	ROM, single dose	No Comparator
[16]Pai et al., 1999	India	Case series	634	-	-	-	Clinical	1-5	ROM, single dose	No Comparator
[17]Ravenkar et al., 2002	India	Cohort	335	-	-	6-70	Clinical	2-5	ROM, single dose	No Comparator
[18]Shetty et al., 2011	India	Retrospective cohort	62	-	-	-	Clinical + Histological	1-5	ROM, single dose	i) WHO-MDT, ii) dapsone, iii) RO
[19]Shinde et al., 2000	India	Case series	26	-	-	-	Clinical	Single	ROM, single dose	No Comparator
[21]Shukla et al., 2000	India	Clinical Trial	61	-	55.7	12	Clinical + Histological	Single	ROM, single dose	No Comparator
[22]Sousa et al., 2007	Brazil	Case series	135	30.5 (15.4)	44.4	31.4	Clinical	Single	ROM, single dose	No Comparator
[23]Stefani et al., 2003	Brazil	Case series	39	33.4 (15.3)	51.28	32.4 (16.0)	Histological	Single	ROM, single dose	No Comparator
[24]Vivekkumar et al., 2010	India	Randomized Control Trial	72	-	61	6	Clinical	1-5	ROM, single dose	RLM, single dose

Table 1. Preliminary Baseline Characteristics of Included Studies
Abbreviations: Rifampin + Ofloxacin (RO), Standard World Health Organization Multi-drug therapy (WHO-MDT), Rifampin + Levofloxacin + Minocycline (RLM)
*Low-dose Convit vaccine contained $1.6x10^7$ heat-killed M. *leprae* in 0.1ml saline and $1.5x10^7$BCG in 0.1ml saline

Results

Outcome	Study	ROM		Comparator		Difference (%)
		% of patients	Proportion	% of patients	Proportion	
Lesion Clearance	[1]Alam et al., 2007	75.93	205/270	-	-	-
	[2]Babu et al., 1997	44.25	327/739	50.27	374/744	-6.02
	[3]Desikan & Gupte, 2001	96.22	102/106	96.15	100/104	0.07
	[5]Diniz et al., 2010	85.20	45/54	-	-	-
	[6]Ebenezer et al., 1999	84.62	11/13	-	-	-
	[7]Emmanuel & Gupte, 2005	-	-	-	-	-
	6mo	3.85	1/26	16.00	4/25	-
	12mo	38.46	10/26	44.00	11/25	-
	18mo	42.31	11/26	60.00	15/25	-
	24mo	46.15	12/26	64.00	16/25	-
	Mean of first 4 f/u	32.69	-	46.00	-	-13.31
	[10]Gomes et al., 2008	80.69	209/259	-	-	-
	[9]Girdhar et al., 2011	-	-	-	-	-
	6mo	72.85	110/151	78.52	117/149	-
	12mo	89.40	135/151	89.26	133/149	-
	18mo	94.59	140/148	91.72	133/145	-
	Mean of first 3 f/u	86.61	-	86.50	-	0.11
	[11]Kumar et al., 2015	97.22	105/108	93.27	97/104	3.95
	[12]Majumder et al., 2000	46.67	14/30	33.30	20/60	13.37
	[13]Mane et al., 1997	25.00	14/56	-	-	-
	[14]Manickam et al., 2012	72.11	486/674	72.12	494/685	-0.01
	[17]Ravenkar et al., 2002	98.74	626/634	-	-	-
	[23]Stefani et al., 2003	44.00	11/25	-	-	-
	[24]Vivekkumar et al., 2010	36.11	13/36	75.00	27/36	-38.89
	Mean	**52.73**	-	**57.42**	-	**-4.69**
	Median	**75.93**	-	**73.56**	-	**2.37**
	Range	**25.00-98.74**	-	**33.33-96.15**	-	**Negative in favour for ROM**
Treatment Failure	[3]Desikan & Gupte, 2001	3.77	4/106	3.85	4/104	-0.08
	[11]Kumar et al., 2015	0.93	1/108	3.87	4/104	-2.94
	[12]Majumder et al., 2000	23.33	7/30	18.33	11/60	5.00
	[13]Mane et al., 1997	0.98	1/102	-	-	-
	[14]Manickam et al., 2012	0.30	2/674	0.58	4/685	-0.28
	[17]Ravenkar et al., 2002	3.79	24/634	-	-	-
	[22]Sousa et al., 2007	1.48	2/135	-	-	-
	[23]Stefani et al., 2003	2.70	1/37	-	-	-
	Mean	**4.66**	-	**6.66**	-	**-2.00**
	Median	**2.09**	-	**3.86**	-	**-1.77**
	Range	**0.30-23.33**	-	**0.58-18.33**	-	**Positive in favour for ROM**
Relapse	[1]Alam et al., 2007	3.70	10/270	-	-	-
	[2]Babu et al., 1997	0.81	6/739	0.81	6/744	0.00
	[5]Diniz et al., 2010	9.3	5/54	-	-	-
	[17]Ravenkar et al., 2002	1.49	5/335	-	-	-
	[9]Girdhar et al., 2011	2.22	3/135	1.43	2/140	0.79
	[11]Kumar et al., 2015	2.78	3/108	6.73	7/104	-3.95
	[14]Manickam et al., 2012 *	-	29/100py	-	9/100py	20/100py
	Mean	**3.38**	-	**2.99**	-	**0.39**
	Median	**2.50**	-	**1.43**	-	**1.07**
	Range	**0.81-9.3**	-	**0.81-6.73**	-	**Negative in favour for ROM**
Side Effects	[2]Babu et al., 1997	0.68	5/739	0.94	7/744	-0.26
	[3]Desikan & Gupte, 2001	0.00	0/118	1.69	2/118	-1.69
	[12]Majumder et al., 2000	0.00	0/30	0.00	0/60	0
	[13]Mane et al., 1997	0.00	0/220	-	-	-
	[15]Martelli et al., 2000	5.79	15/259	-	-	-
	[24]Vivekkumar et al., 2010	0.00	0/36	0.00	0/36	0
	Mean	**1.08**	-	**0.66**	-	**0.42**
	Median	**3.24**	-	**1.32**	-	**1.93**
	Range	**0.68-5.79**	-	**0.94-1.69**	-	**Negative in favour for ROM**
Reversal Reactions (Type 1&2)	[2]Babu et al., 1997	0.95	7/739	0.40	3/744	0.55
	[5]Diniz et al., 2010	1.85	1/54	-	-	-
	[7]Emmanuel & Gupte, 2005	7.69	2/26	0.00	0/25	7.69
	[10]Gomes et al., 2008	16.20	42/259	-	-	-
	[13]Mane et al., 1997	3.33	1/30	-	-	-
	[21]Shukla et al., 2000	6.50	4/61	-	-	-
	[22]Sousa et al., 2007	14.81	20/135	-	-	-
	[23]Stefani et al., 2003	33.33	13/39	-	-	-
	Mean	**8.35**	-	**0.2**	-	**8.15**
	Median	**7.69**	-	**0.2**	-	**7.49**
	Range	**0.95-33.33**	-	**0.00-0.40**	-	**Negative in favour for ROM**

Table 2. Preliminary Summary of Primary Outcomes; *Not included in mean/median/range

Discussion

- Preliminary outcomes suggest that ROM is less efficacious than its comparator, however a more robust analysis is necessary
- Qualitatively, several determinants of health were identified throughout this analysis including:
  - Social environments – 50% of non-compliant patients denied having leprosy due to potential loss of jobs and/or marriage prospects[25]
  - Patient education – 86% of respondents did not understand the concept of their disease[12]
  - Gender – Women only completed treatment at a rate of 65.6% and men at 79.2% ($p<0.05$)[26]
- Synthesizing the current evidence discussing the efficacy of monthly ROM, will strengthen the current body of knowledge surrounding the treatment of paucibacillary leprosy, and may allow for the development of standardized fluoroquinolone-based treatment protocols.

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Rifampin-Ofloxacin-Minocycline (ROM) for the Treatment of Multibacillary Leprosy: A Systematic Review

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Introduction

- From a diagnostic and management perspective, leprosy is a complex tropical infection.
- Patients who are affected by leprosy are at risk of several complications associated with the disease itself and its treatment
- Standard WHO multi-drug treatment (MDT) for leprosy consists of medications that are potentially harmful and cause a range of adverse systemic effects
- Alternative options for potential treatment have emerged such as monthly dosing of Rifampin-Ofloxacin-Minocycline (ROM) combination therapy, however, there is limited synthesized evidence of efficacy
- Multibacillary leprosy, characterized by many skin lesions and a high bacillary load, may be most amenable to a fluoroquinolone-based treatment protocol
- Monthly- or single dosing of ROM has emerged as a potential treatment option for leprosy, however, a synthesis of the evidence supporting ROM does not exist

Methods

- Abstracts reporting the efficacy & safety of monthly ROM treatment in multibacillary leprosy in human patients were targeted using combinations of the search terms “ROM” & “Leprosy” from inception to March 2019
- Non-English publications were included and translated using Google Translate
- During all phases of screening a tertiary arbitrator will mitigate any inclusion/exclusion discrepancies

Included
Systematic reviews
Randomized controlled trials
Clinical trials
Cohort studies
Observational studies
Case-control studies
Case series (N>5)
Excluded
Case reports
Case series (N<4)

Table 1. Inclusion and exclusion criteria implemented during title and abstract screening

Primary Outcome Measures	Secondary Outcome Measures
Lesion clearance	Social environments
Treatment failure	Patient education
Relapse	Health services
Side effects	Income
Reversal reactions	Gender

Table 2. Preliminary outcome measures to be assessed during full text screening

Results

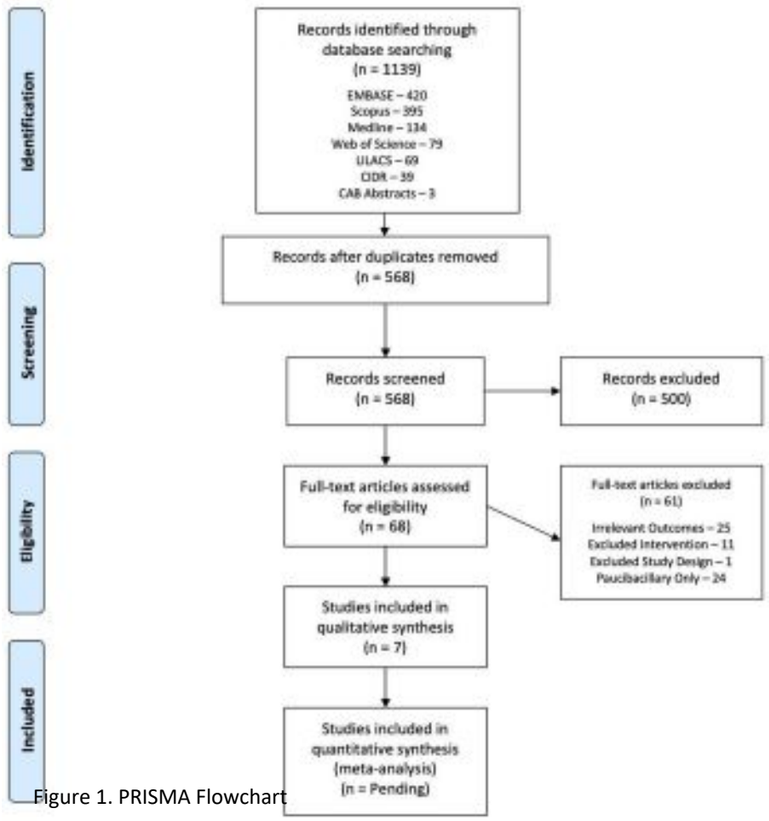

Figure 1. PRISMA Flowchart

Author, Year	Country	Study Design	Sample Size	Mean Age (SD), y	Male, %	Follow-Up, mo	Diagnosis of Leprosy	Treatment	Comparator
[1]Ji et al., 1998	Mali	Randomized Control Trial	20	34 (14)	80	0.25	Clinical + Histological	ROM, single dose	Ofloxacin + minocycline
[3]Kumar & Girdhar, 2014	India	Case Series	19	40.2 (4.0)	68.42	-	Clinical	ROM, monthly	No Comparator
[7]Kumar et al., 2014	India	Cohort	289	41.6	61.8	12	Clinical	ROM, monthly	WHO-MDT
[8]Mane et al., 1997	Senegal	Case series	220	-	60	12	Clinical + Histological	ROM, monthly	No Comparator
[9]Shetty et al., 2011	India	Retrospective cohort	62	-	-	-	Clinical + Histological	ROM, single dose	i) WHO-MDT, ii) dapsone, iii) RO
[4]Ura et al., 2007	Brazil	Randomized Control Trial	26	-	-	24	Clinical + Histological	ROM, monthly	WHO-MDT
[5]Villahermosa et al., 2004	Philippines	Randomized Control Trial	21	29.4	81.5	24	Clinical + Histological	ROM, monthly	WHO-MDT

Table 3. Preliminary Baseline Characteristics of Included Studies; Rifampin + Ofloxacin (RO), Standard World Health Organization Multi-drug therapy (WHO-MDT)

Discussion

- Several determinants of health were identified qualitatively throughout this analysis including:
  - Social environments – 50% of non-compliant patients denied having leprosy due to potential loss of jobs and/or marriage prospects[3]
  - Patient education – 86% of respondents did not understand the concept of their disease[8]
  - Gender – Women only completed treatment at a rate of 65.6% and men at 79.2% ($p<0.05$)[9]
- Synthesizing the current evidence discussing the efficacy of monthly ROM, will strengthen the current body of knowledge surrounding the treatment of paucibacillary leprosy, and may allow for the development of standardized fluoroquinolone-based treatment protocols.

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Solid Organ Transplant in Acute Tropical Infectious Diseases - A Systematic Review and Meta-Analysis of Indications and Outcomes for the Liver

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Introduction

- We aim to understand the frequency, indications for, and outcome of SOT in the liver for treatment of acute tropical infectious diseases (such as yellow fever) presenting with fulminant organ failure.
- Fulminant life-threatening presentations of acute tropical infectious diseases may occur, and the degree of end-organ impairment may qualify patients for emergency liver solid-organ transplantation (SOT).
- However, liver SOT may not be beneficial in all cases as failure of the transplanted organ is only one possible cause of death[1]. The outcomes from such an intervention are largely unknown for many acute tropical infectious diseases.
- Due to a paucity of synthesized data, there is a knowledge gap around indications for and outcomes in liver SOT for severe acute tropical infectious diseases.

Methods

- We will be conducting a systematic review and meta-analysis.
- PubMed, Embase, Scopus, Cochrane, and LILACS were searched using combinations of search terms such as the following: “liver” or “hepatic”, “transplant”, “yellow fever”, “dengue” from database inception to November 30, 2020.
- Full-text articles will be divided according to the type of SOT examined (e.g. liver SOT or kidney SOT).

Results

Figure 1. PRISMA Flow Diagram

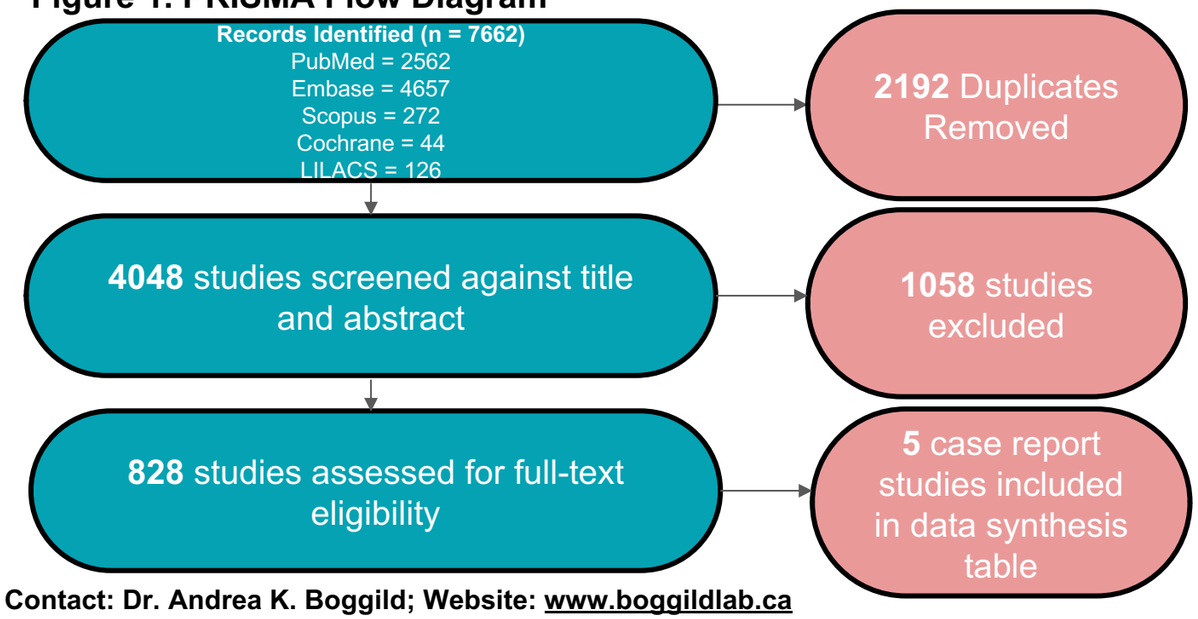

Table 1. Data Synthesis Table

Study	Year of Publication	Organ	Tropical Disease(s)	Pathogen (Full Name)	Method of Diagnosis	Outcome (ex. Mortality/Survival; Temperature; Biochemical Parameter, etc.)	Study Type	Other Comments
Song, Alice Tug Wang	2018	Liver	Yellow Fever Virus (YFV), Fulminant Hepatitis (Acute Liver Failure)	Hepatitis D Virus (HDV)	Antibody Detection	Patient survived	Case Report	N/A
Shimata, Keita	2018	Liver	Fulminant Hepatitis (Acute Liver Failure)	Hepatitis E Virus (HEV)	Antibody Detection	Patient survived; It took 2.5 months for HEV RNA to become undetectable; Patient was discharged from the hospital on postoperative day 43; 8 months post transplant, his graft function is normal & HEV RNA has remained negative	Case Report	N/A
Tenorio González, Elena	2018	Liver	Fulminant Hepatic Failure (Acute Liver Failure)	Hepatitis E Virus (HEV)	Antibody Detection	Patient survived; Negative HEV RNA; One year later, patient is in excellent post-transplant condition on treatment with tacrolimus	Case Report	In this case, diagnosis of HEV was confirmed after liver transplant was performed
Li, Iris Wai Sum	2017	Liver	Hepatitis E Infection (Acute Liver Failure)	Hepatitis E Virus (HEV genotype 3)	Antibody Detection	Patient survived; no HEV reactivation 4 years post liver transplant	Case Report	N/A
Paskaran, P.	2008	Liver	Hepatitis E Infection (Acute Liver Failure)	Hepatitis E Virus (HEV)	Antibody Detection	Survival	Case Report	N/A

Discussion & Conclusion

1. Due to a paucity of synthesized data, there is a knowledge gap around indications for and outcomes in liver SOT for severe acute tropical infectious diseases.
2. Most published literature on SOT in acute tropical infectious diseases is related to liver transplantation for acute Hepatitis E Virus infection. All 5 cases survived.

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Solid Organ Transplant in Acute Tropical Infectious Diseases - A Systematic Review and Meta-Analysis of Indications and Outcomes for the Kidney

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Introduction

- We aim to understand the frequency, indications for, and outcome of SOT in the kidney for treatment of acute tropical infectious diseases (such as Malaria) presenting with fulminant organ failure.
- Fulminant life-threatening presentations of acute tropical infectious diseases may occur, and the degree of end-organ impairment may qualify patients for emergency kidney solid-organ transplantation (SOT).
- However, kidney SOT may not be beneficial in all cases as failure of the transplanted organ is only one possible cause of death[1]. The outcomes from such an intervention are largely unknown for many acute tropical infectious diseases.
- Due to a paucity of synthesized data, there is a knowledge gap around indications for and outcomes in kidney SOT for severe acute tropical infectious diseases.

Methods

- We will be conducting a systematic review and meta-analysis.
- PubMed, Embase, Scopus, Cochrane, and LILACS were searched using combinations of search terms such as the following: “kidney” and “transplant”, and “malaria”, “Plasmodium spp.,”, and “Lepto*” from database inception to November 30, 2020.
- Full-text articles will be divided according to the type of SOT examined (e.g. liver SOT or kidney SOT).

Results

Figure 1. PRISMA Flow Diagram

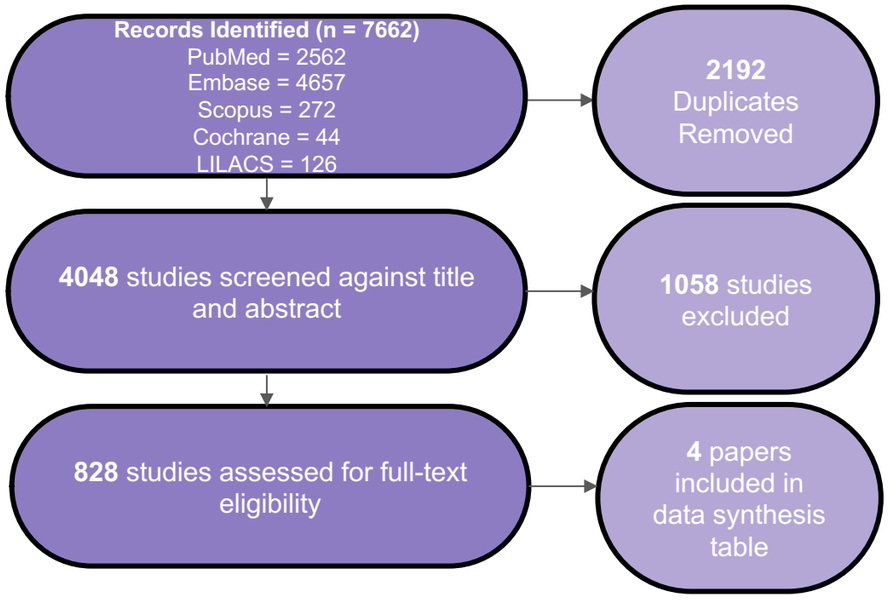

Table 1. Data Synthesis Table

Study	Year of Publication	Organ	Tropical Disease(s)	Pathogen (Full Name)	Method of Diagnosis	Outcome (ex. Mortality/Survival; Temperature; Biochemical Parameter, etc.)	Study Type	Other Comments
Sheerin, Neil	1999	Kidney	Malaria	Plasmodium falciparum	Microscopy	Patient survived	Letter	N/A
Rajesh, Jhorawat	2015	Kidney	Malaria	Plasmodium Vivax	Microscopy	Patient was dialysis dependent and later underwent renal transplantation successfully	Review	N/A
Reynaud, F.	2005	Kidney	Malaria	Plasmodium falciparum	Microscopy	Patient survived but lost her eyesight after complete recovery	Letter	N/A
Naqvi, R.	2003	Kidney	Malaria	Plasmodium falciparum, Plasmodium vivax	Microscopy	62% patients had complete renal recovery, 26% died. 62% had complete recovery of renal function, 12% were progressing towards recovery when lost to follow-up	Research Article	78% of deaths occurred within the first 48 h of admission. Among the patients who survived, 61% were oliguric.

Discussion & Conclusion

- Due to a paucity of synthesized data, there is a knowledge gap around indications for and outcomes in kidney SOT for severe acute tropical infectious diseases.
- Most published literature on kidney SOT in acute tropical infectious diseases is related to kidney transplantation for malaria. One of the common health outcomes is survival.

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Influence of Host Nutriome on Immunological Control of Leishmania Infection

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Introduction

- Immunologic control of parasitic infections arises from a combination of humoral and cellular mechanisms, both of which may be influenced by host nutritional status
- Micronutrient depletion or over-repletion impairs the functioning of the immune system, potentially resulting in increased susceptibility to and poor immunologic control of protozoal infections
- Leishmaniasis is a tissue-dwelling parasitic infection in which disease severity is determined by the host's immune system
- Research suggests that acquired factors such as nutritional inadequacies play a significant role in immunosuppression and enhanced pathogenicity
- We aim to synthesize the knowledge surrounding the interplay between host micronutrient status and tissue-based protozoal infections

Methods

- Five electronic databases were searched with combinations of search terms from database inception to February 10, 2020
- Screening was performed independently by two reviewers with discrepancies arbitrated by a tertiary reviewer
- Following screening, a comprehensive bias assessment will be carried out using the Grading of Recommendations Assessment, Development, & Evaluation (GRADE) approach

Results

Author, Year	Country	Design	Population	Sample Size	Assessment / Intervention	Mean Age ± SD	Sex (F:M)	Outcomes
[1]Goyonlo, 2020	Iran	Case-Control	Diagnosis of CL confirmed by Geimsa-stained direct smear versus age and sex matched controls	220 Cases (149) Control (71)	Nutritional status and Vitamin A intake via FFQ	21.32 ± 17.62	Cases (82:67) Contorls (45:26)	Daily intake of Vitamin A (p<0.001) was significantly lower among the CL group, as well as energy intake, fiber, Vitamin E, and potassium
[2]Guzman-Rivero, 2014	Bolivia	Case-Control	Patients aged 15-50 with confirmed CL on blood, or microbiological/biochemical analysis.	29 Cases (14) Controls (15)	Zinc gluconate (315mg) vs placebo (315mg cornstarch) for 60 days	Not Reported	Not Reported	A statistically significant biological or clinical effect due to zinc was not found
[3]Maciel, 2014	Brazil	Case-Control	Children with clinical and laboratory confirmed VL versus healthy controls	26 Cases (10) Controls (16)	Serum vitamin A (retinol) status and immune response (CD4+CD24Foxp3+ T cells)	Cases (7.99 ± 7.85) Controls (8.82 ± 6.26)	Cases (7:3) Contorls (5:11)	Vitamin A (retinol) status (p=0.013) and immune cells (p=0.011) were significantly lower in cases versus controls
[4]Maciel, 2008	Brazil	Case-Control	Biochemically confirmed cases of paediatric VL versus healthy controls	149 Cases: Active VL (20) History of VL (33) Antigen Response to VL (40) Controls (56)	Nutritional status via anthropometry, and serum Vitamin A (retinol) level	Cases: Active VL (4.7 ± 3.9) History of VL (10.1 ± 3.3) Antigen Response to VL (11.2 ± 2.4) Controls (8.1 ± 3.4)	Cases: Active VL (11:9) History of VL 19(:14) Antigen Response to VL (20:20) Controls (31:25)	Serum retinol was significantly lower in patients with active VL versus controls (p=0.037)
[5]Cerf, 1987	Brazil	Case-Control	Children aged 0-15 years old with at least 2 consecutive years of anthropometric and serologic data confirming presence of VL	1066	Nutritional status via weight-for-age index	Not Reported	Not Reported	Low weight-for-age was significantly higher In VL children versus controls (p < 0.0001)
[6]Kumar, 2014	India	Case-Control	Patients with confirmed, active, and untreated cases of VL versus healthy controls	40 Cases (20) Controls (20)	Nutrition status via weight-to-height ratios and immune response (including ROS activity, cytokine levels, leishmania antigen) via biochemistry	Not Reported	Not Reported	Patients found to be malnourished had a statistically significant weakened immune response to VL on several accounts as compared to healthy controls: antigen responsiveness, monocytes, & ROS activity (p<0.05), CD62-L (p<0.001)
[7]Kocyigit, 2002	Turkey	Case-Control	Patients with laboratory confirmed CL versus healthy controls	50 Cases (28) Controls (22)	Serum nutrient levels: copper, zinc, and iron, and immunoregulatory cytokines: IL-1B, IL-2R, IL-6, IL-8, TNF-a	Cases (27.3 ± 3.8) Controls (28.4 ± 4.1)	Not Reported	Plasma selenium, zinc, iron, and IL-2r levels were significantly lower and plasma copper, IL-1B, IL-8, IL-6, and TNF-a were significantly higher in cases versus controls (p<0.01)
[8]Al-Jurayyan, 1995	Saudi Arabia	Cohort Study	Infants and children undergoing active treatment for Leishmania donovani	94	Haematological findings including nutrition via biochemistry	1.8	39:55	Patients with active infection were found to be immunocompromised and iron deficient
[9]Carbone, 2018	Brazil	Clinical Trial	Patients with parasitologically confirmed presence of VL	67 Intervention: With Zinc (33) Without Zinc (29) Controls (15)	Zinc (2mg/kg/day) plus standard treatment (amphotericin B (0.5-1mg/kg/day) or glucantime (20mg/kg/day)) for 20 days versus standard alone	Intervention: With Zinc (46.20 ± 9.66) Without Zinc (43.76 ± 6.50) Controls (44.60 ± 10.20)	Intervention: With Zinc (12:11) Without Zinc (18:11) Controls (9:6)	Patients who received Zinc supplementation exhibited a more rapid reduction in spleen size compared to controls (p<0.05)
[10]Mengesha, 2014	Ethiopia	Cross-Sectional	Patients age >17 years and non pregnant women with a confirmed diagnosis of VL	403	Nutritional status via BMI	Only Range Provided: 68% 18-27 years old 25.8% 28-37 years old 6.2% >37 years old	6:397	The prevalance of malnutrition and VL infection was 95.5% while presence of intestinal parasitic infection was statistically associated with severe malnutrition in VL patients (p<0.001)

Table 1. Preliminary Data Extraction of Included Studies

Abbreviations: Cutaneous Leishmaniasis (CL), Visceral Leishmaniasis (VL), Food Frequency Questionnaire (FFQ), Reactive Oxygen Species (ROS), Body Mass Index (BMI), Interleukin (IL), Tumor Necrosis Factor (TNF)

Results

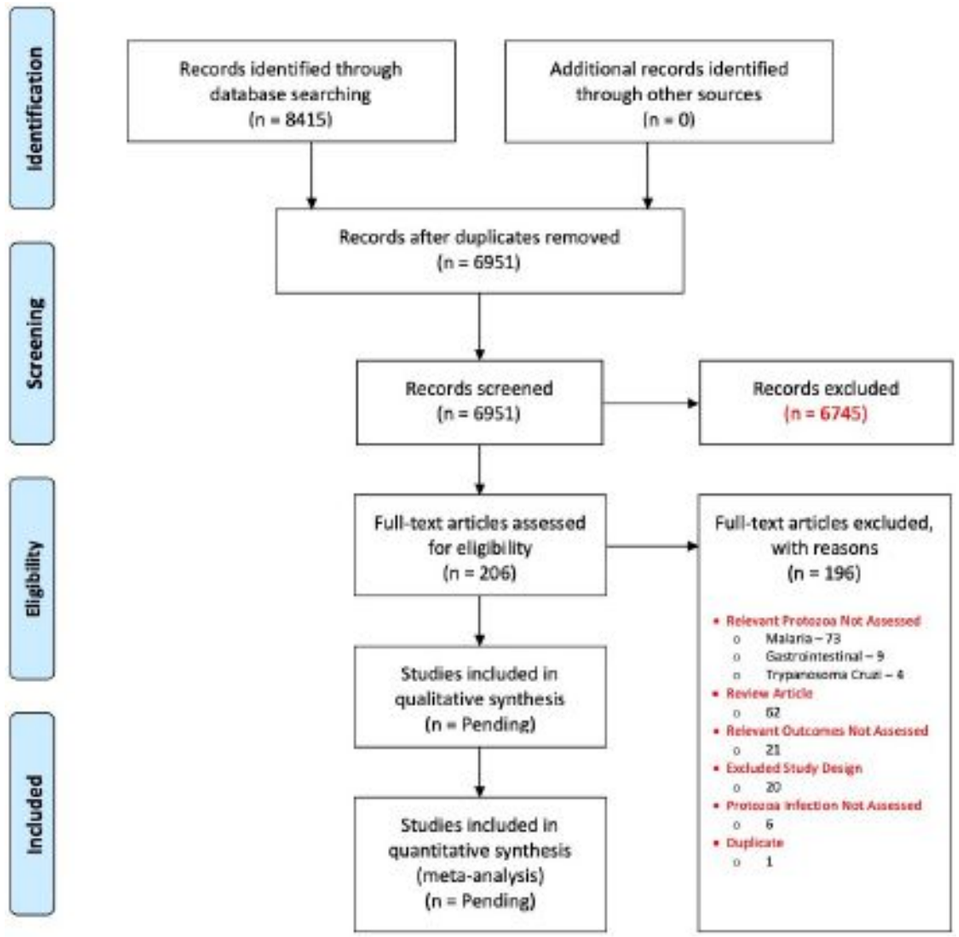

Figure 1. PRISMA Flowchart

Discussion

- Following full-text screening 10 articles remained for absolute inclusion
- Preliminary data extraction suggests that malnourished individuals, including potassium, iron, zinc, and vitamins A & E, are at greater risk of acquiring a significant leishmanial infection
- The data will be summarized to systematically map published literature that will illuminate a number of ways in which nutrient deficiencies or abnormal micronutrient status alter and impair immune function in persons with leishmaniasis
- This synthesized body of information will ultimately inform adjunctive therapeutic decisions in the context of leishmaniasis, which has the potential to improve patient prognosis

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Clinical Medicine Group 1

An Update on the Role of Imaging in the Care of Patients with Intestinal Schistosomiasis

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Background:

- Schistosomiasis leads to significant morbidity and mortality worldwide and infection with *Schistosoma mansoni* and *S. japonicum* can lead to severe hepatic disease including periportal liver fibrosis, portal hypertension and esophageal varices[1]
- World Health Organization (WHO) guidelines recommend the use of abdominal imaging to detect early hepatic changes in order to improve disease outcome[2] but there are limited up-to-date authoritative resources to guide the utilization of imaging in the initial management of those with schistosomiasis
- **We mapped available literature regarding the role of imaging in the evaluation of patients with schistosomiasis to inform clinical recommendations for risk stratification of disease**

Methods:

- Eight electronic databases were searched: Ovid Medline, EMBASE, Cochrane Library of Systematic Reviews, Epistemonikos, Global Health, NICE, TRIP and LILACS from database inception to February 28, 2019 with the following search terms:

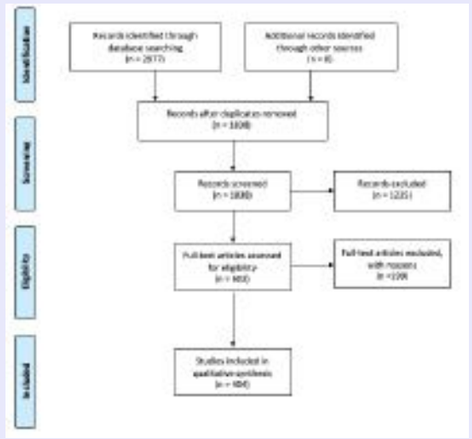
Schistosomiasis		Medical Imaging		Liver
Schistosomiasis		CT		Liver
Schistosoma mansoni		Computed tomography		Periportal fibrosis
Schistosoma japonicum		Ultrasound		Hepatic
		Ultrasonographs		Echogenic
	AND	MRI	AND	Hepatosplenic
		Magnetic resonance imaging		Portal hypertension
		Echo imaging		
		Sonography		
		Sonogram		
Schistosomiasis OR (Schisto* AND (mansoni OR japonicum))		CT OR (computed AND tomography) OR Ultraso* OR Sonogr* OR MRI OR (Magnetic AND resonance AND imaging) OR Echo OR Imaging		Liver OR periportal OR peri-portal OR fibrosis OR hepat* OR echogenic* OR (portal AND hypertension)

- Titles, abstracts and full-text articles were systematically screened by two reviewers with a tertiary arbitrator
- Data extraction was performed by two reviewers and the quality of the articles will be critically evaluated using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach. Meta-analysis was performed in comprehensive meta-analysis software using random effects model

Results:

Figure 1: PRIMSA Flow Diagram

collated from analysis of 9 articles selected for full text review by Dec/20



Based on the data analyzed for this **systematic review** to date:

The pooled **prevalence** for **liver fibrosis** detected by **ultrasound was 68%** in patients with schistosomiasis in Brazil

Abdominal **ultrasound** can detect liver fibrosis **in the absence of clinical disease**

Abdominal ultrasound is an important diagnostic tool in the diagnosis of schistosomiasis-related liver disease

Results Cont'd:

Table 1: Summary of Data Captured

Study	Author (Year)	Study Setting	Baseline Characteristics	Patients with Diagnosis of Schistosomiasis mansoni	Patients with Liver Abnormalities (all degrees) Detected by Ultrasound	Percentage of Patients with Fibrosis Detected by Ultrasound
1	Ndamba (1991)	Zimbabwe	Sugar cane cutters aged 18-54 with schistosomiasis without anti-schistosomal therapy in past 3 years (n=315; n=120 uninfected controls)	315	148	47
2	Domingues (1993)	Brazil	Out-patients with clinical forms of schistosomiasis	176	121	68.8
3	Tanabe (1997)	Brazil	Villagers in an endemic area for schistosomiasis	405	155	51.8*
4	Burchard (1998)	Senegal	Villagers in an endemic area for schistosomiasis	383	268	59**
5	Barata (1999)	Brazil	Patients aged 5-45 with acute schistosomiasis (n=26 acute schistosomiasis, n=26 controls)	26	5	19.2
6	De Jesus (2000)	Brazil	Patients aged 7-38 with schistosomiasis diagnosed x2 tests x2 separate days	164	156	95
7	Hoffman (2001)	Madagascar	Villagers in an endemic area for schistosomiasis	448	99	19***
8	Prata (2010)	Brazil	Group 1: (n=41) clinical evidence of liver disease, Group 2: (n=102) clinical evidence of liver disease in the past, Group 3: (n=268) no clinical evidence of liver disease N=411 with schistosomiasis	411	128	31.1
9	Silva (2013)	Brazil	Patients aged 18-89 with the hepatosplenic form (n=137) and hepatointestinal form (n=41)	178	170	95.5

*Of the 299 with ultrasonography performed assuming all had schistosomiasis infection
** Of the 454 analyzed for the Cairo classification assuming all had schistosomiasis infection
*** Of the 520 analyzed for the Cairo subsegmental classification assuming all had schistosomiasis infection

Results Cont'd:

Figure 2: BREAK DOWN OF SCHISTOSOMIASIS PATIENTS WITH ULTRASOUND EVIDENCE OF LIVER DISEASE

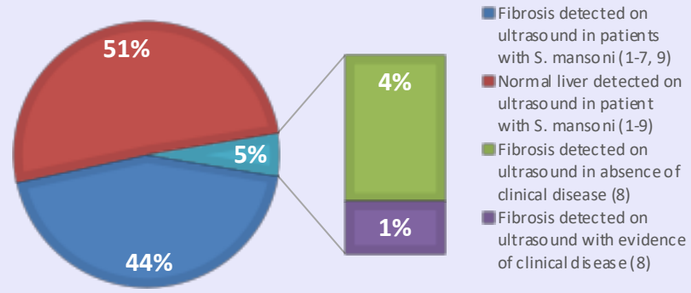
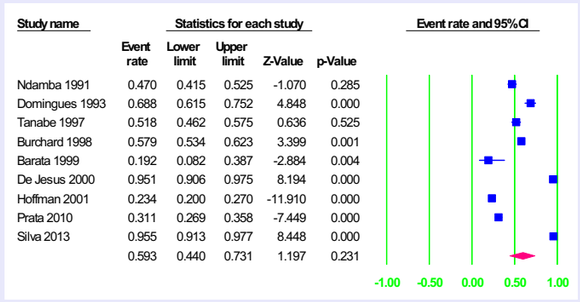


Table 3: **Prevalence of peri-portal fibrosis in *Schistosoma mansoni* patients**



- The pooled prevalence of periportal fibrosis was 59% across the 9 studies evaluating 2600 patients with schistosomiasis
- Sub-analysis in Brazil showed a prevalence of 68%
- The most well represented imaging modality was ultrasound scanning, which documented liver status in 100% of patients
- No included studies reported on use of CT or MRI for liver evaluation

Discussion:

- Abdominal ultrasound is an important diagnostic tool in the detection of schistosomiasis related liver disease
- The prevalence of infection was 59% and was 68% in Brazil
- WHO guidelines support that abdominal imaging can detect early hepatic changes that could indicate downstream periportal fibrosis[2], thereby improving outcomes
- Synthesizing the current literature on abdominal imaging in the evaluation of schistosomiasis can translate into clinical recommendations for improved risk stratification and management of schistosomiasis, and thereby overall improvement of disease outcomes

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2. World Health Organization (WHO). (1996). *Ultrasound in Schistosomiasis. A Practical Guide to the Standardized Use of Ultrasonography for the Assessment of Schistosomiasis-related Morbidity.* Second International Workshop, UNDP/World Bank/WHO/TDR.
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A Systematic Review of Scorpion Envenomation Therapeutics and Antivenom Accessibility

Syed Zain Ahmad[1], Christian Lecce[1], Avinash N. Mukkala[1,2], Michael Klowak[1,2], Aisha Khatib[3], Priyanka Challa[1], Eric Shao[1], Jason Kwan[1,4], Tianna Chong-Kit[1], Jamie Sookhoo[1], Emma Hagopian[1], Dylan Kain[1,4], Mofe Adeosun[1], Andrea K. Boggild[1,2,4,*]

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Introduction:

- Scorpions (*Scorpiones*) are eight-legged arthropods of the class *Arachnida*
- Increased human migration and transcontinental produce shipment may cause the incidence of arachnid envenomations to increase in non-endemic areas[1]
- We aim to compile existing envenomation prevention and treatment data into a clinical resource to be used at the bedside when encountering envenomations

Methods:

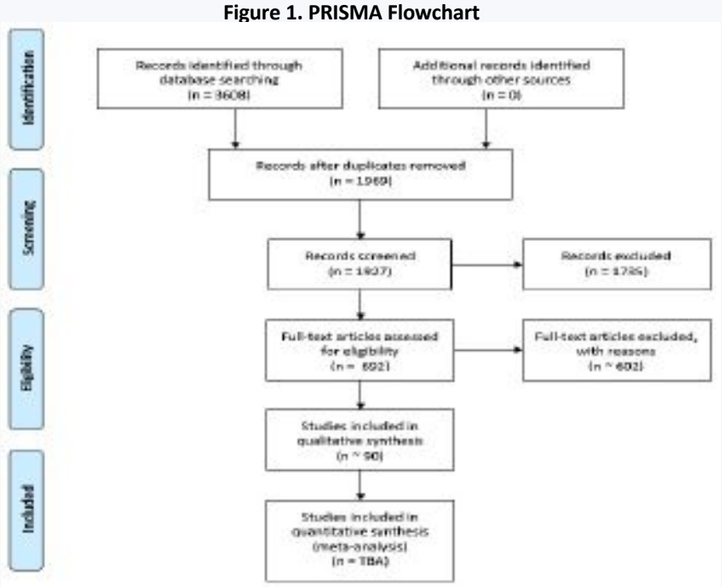
1. PubMed (NCBI), MEDLINE (OVID), EMBASE (OVID), Cochrane Database of Systematic Reviews (CIDR) and TOXLINE (TOXNET) were searched from inception to June 2018 using combinations of the search terms "spider", "scorpion", and "envenomation*"

- We included: observational studies, case reports, case series, and cohort studies, as well as clinical trials, and antivenom safety, tolerability, and efficacy.
- We excluded: Molecular epidemiology and purely mechanistic pathogenesis studies

2. Abstracts underwent double reviewer screening and only titles about spiders that had double inclusion responses were included for the full-text review.
3. A different pair of authors screened the subsequent full-texts and only double inclusion responses were included in the systematic review.

The GRADE approach will be used to assess quality of studies reporting therapeutic interventions. Evidence will be summarized using descriptive measures for each intervention type. Meta-analysis will be planned if sufficient efficacy measures exist.

Figure 1. PRISMA Flowchart



Results:

1. 961 MEDLINE articles, 1053 PubMed, 1486 EMBASE, 0 CIDR and 149 TOXLINE records were retrieved for title and abstract screening; after de-duplication, **1928** remained.
2. Following the abstract screening protocol, **692** titles advanced to full-text review.
3. Full-text screening resulted in the inclusion of **~90** titles to the systematic review.
4. Our analysis captures the reported clinically-relevant data of **529, 469 scorpion envenomation victims.**
5. Of 1,065 envenomated patients, **only 134 (12.6%)** developed some **mild reaction or adverse event** due to antivenom usage.

Discussion:

Increased transcontinental movement of people and tropical produce has facilitated importation of arachnids to non-endemic regions where clinicians lack familiarity with envenomation syndromes and appropriate therapeutics. Synthesizing the current evidence around therapeutic strategies for arachnid envenomations can inform the development of appropriate treatment and prevention protocols.

Figure 2. Preliminary qualitative data

Species	Antivenom Adverse Reactions	Pharmacological Treatments
Indian Red Scorpion *(Mesobuthus tamulus)*	• Allergic reactions are possible • Prazosin + Antivenom will reduce the risk of myocardial dysfunction as compared to Prazosin alone	• Hypertensive → nifepedipine and prazosin • Tachycardic → prazosin, digoxin, aminophylline, and oxygen • Pulmonary edema → digoxin, aminophylline, furosemide and prazosin • Massive pulmonary edema → nitroprusside as well • Children deteriorate quicker without antivenom+prazosin, prazosin alone is not enough
Yellow Scorpion (Tityus serrulatus)	• Children with adrenergic manifestations after *T. serrulatus* scorpion sting had significantly lower anaphylactic reactions to antivenom than those without these manifestations • This finding may also be true for adults victims	• Pain at the site of sting → dipyrone & metoclopramide • Shock → intravenous infusion of dobutamine or dopamine • Premedication with epinephrine, antihistamine plus or minus corticosteroid should be given parenterally to patients before antivenom injection to prevent early anaphylactic reactions • Oral analgesics for pain
Centruroides sculpturatus	• Minor vomiting • Some diarrhea • Rare residual amnesia • No acute serum reactions → safe	NA
Other medically relevant species	***Tityus stigmurus, Tityus obscurus, Hemiscorpius lepturus, Androctonus australis***	

1. Chagla Z, Boggild AK, Chakrabarti S. A venomous visitor from the tropics. The Canadian journal of infectious diseases & medical microbiology = Journal canadien des maladies infectieuses et de la microbiologie medicale. 2015;26(5):243-244.

Spider Envenomation Therapeutics and Antivenom Accessibility: A Systematic Review

Mofe Adeosun[1], Christian Lecce[1], Avinash N. Mukkala[1,2], Syed Zain Ahmad[1], Michael Klowak[1,2], Aisha Khatib[3], Priyanka Challa[1], Eric Shao[1], Jason Kwan[1,4], Tianna Chong-Kit[1], Jamie Sookhoo[1], Emma Hagopian[1], Dylan Kain[1,4], Andrea K. Boggild[1,2,4,*]

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Introduction:

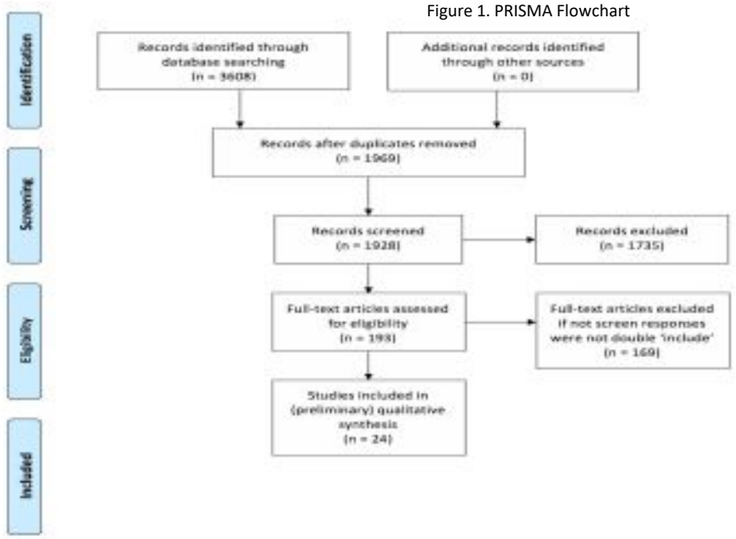
- Spiders (*Araneae*) are eight-legged arthropods of the class *Arachnida*
- Increased human migration and transcontinental produce shipment may cause the incidence of arachnid envenomations to increase in non-endemic areas.
- We aim to compile existing envenomation prevention and treatment data into a clinical resource to be used at the bedside when encountering envenomations.

Methods:

1. PubMed (NCBI), MEDLINE (OVID), EMBASE (OVID), Cochrane Database of Systematic Reviews (CIDR) and TOXLINE (TOXNET) were searched from inception to June 2018 using combinations of the search terms "spider", "scorpion", and "envenomation*"
   - We included: observational studies, case reports, case series, and cohort studies, as well as clinical trials, and antivenom safety, tolerability, and efficacy.
   - We excluded: Molecular epidemiology and purely mechanistic pathogenesis studies
2. Abstracts underwent double reviewer screening and only titles about spiders that had double inclusion responses were included for the full-text review.
2. A different pair of authors screened the subsequent full-texts and only double inclusion responses were included in the systematic review.

Future: A tertiary arbitrator will mitigate any inclusion/exclusion discrepancies experienced during both abstract screening and full-text screening. The GRADE approach will be used to assess quality of studies reporting therapeutic interventions. Evidence will be summarized using descriptive measures for each intervention type. Meta-analysis will be planned if sufficient efficacy measures exist.

Figure 1. PRISMA Flowchart



Results:

- Following the abstract screening protocol, **193** titles advanced to full-text review.
- Full-text screening resulted in the inclusion of **24** titles to the systematic review.
- Reactions to antivenom are rare and adverse events are uncommon
- Of 709 envenomated patients included in the safety analysis, 327 (46.1%) received antivenom, 34 of whom developed adverse events such as: urticarial rash, mild swelling around infusion site, mild Brown reaction, serum sickness, malaise, myalgia, pruritus early anaphylactic reaction, generalized paresthesias, cutaneous rash, nausea/vomiting, itchy rashes, bronchospasm, and generalized flushing
- **Adverse events** as a result of antivenom use occurred in **about ten percent** of patients (34/327, 10.4%)
- Studies report that **4-33%** of patients who receive antivenom for any spider envenomation can develop some mild adverse reaction.

Discussion:

Increased transcontinental movement of people and tropical produce has facilitated importation of arachnids to non-endemic regions where clinicians lack familiarity with envenomation syndromes and appropriate therapeutics. Synthesizing the current evidence around therapeutic strategies for arachnid envenomations can inform the development of appropriate treatment and prevention protocols.

Figure 2. Preliminary qualitative data

Species	Antivenom Adverse Reactions	Pharmacological Treatments
Latrodectus hasseltii **(Redback Spider)**	The adverse reaction rates were similar between IV and IM administration. There were no severe cases of anaphylaxis.	Pre-medication before antivenin with: • Antihistamines • Adrenaline and antihistamine
Latrodectus mactans **(Black Widow Spider)**	Symptoms: • Urticarial rash • Fatal bronchospastic event • Myalgias • Fatigue • Generalized parenthesia • Generalized flushing Antivenom was avoided in patients who tested positive for a skin test or had a history of asthma or allergies	• Morphine • Merperidine • Methocarbamol • Calcium gluconate • Diazepam • Analgesics • Diphenhydramine • Benzodiazepines • Cefaclor • Nebulized albuterol • Opioids • Antihistamines • Antibiotics • Nonsteroidal anti-inflammatories Skeletal muscle relaxants Inefficacious: • Morphine and lorazepam • Hydromorphone, ketorolac, metoclopramide and lorazepam • Morphine and diazepam • calcium gluconate
Loxosceles reclusa **(Brown Recluse Spider)**		• Eculizumab • Steroids • Antihistamines • Dapsone • Topical antibiotics • Nitroglycerine patch • Dapsone • IV Antibiotics • PRBC Transfusion • FFP Transfusion • Oral erythromycin • IM dexamethasone
Latrodectus **spp. (Widow Spider)**	Various adverse drug reactions.	• Benzodiazepines • Calcium • Intravenous fluids
Phoneutria **spp. (Armed Spider)**	No adverse drug reactions	• Local anesthesia alone • Local anesthesia plus analgesics • Oral analgesics alone

A Systematic Review of Virulence Factors in Old World *Leishmania* species

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INTRODUCTION

- Leishmaniasis is a neglected tropical disease divided into three major classifications based on clinical presentation: cutaneous (CL), mucocutaneous (MCL) and visceral (VL)
- Transmitted by the *Lutzomyia* spp. and *Phlebotomus* spp. sandflies, there are up to 2 million cases of Leishmaniasis globally while 350 million people are at risk
- Parasite-determined factors play a complementary role in the pathogenesis of leishmaniasis
- Virulence factors (VFs), or pathogen moieties facilitating disease, can potentiate host cell damage by *Leishmania* spp. by increased expression, host cell invasion, stress tolerance, and modulation of the host immune system
- Due to large eukaryotic genomes in *Leishmania* spp., there is a wide array of VFs which contribute to different aspects of pathogenesis; we aim to synthesize this knowledge by systematically mapping the literature

METHODS

- PubMed (NCBI), MEDLINE (OVID), EMBASE (OVID), Web of Science, and LILACS (VHL) were searched from inception to July 2018 using combinations of the search terms "virulence factor*", "*Leishmania*", and "Leishmaniasis*", while accounting for unique database syntax
- Iterative inclusion and exclusion of search terms was employed to maximize relevant article extraction
- Primarily, molecular and mechanistic pathogenesis studies in various model systems, observational studies, review studies, cohort studies, as well as clinical trials are included
- Synthesis is done by grouping of similar VFs in similar pathogenesis mechanisms, e.g., heat shock
- 760 MEDLINE, 1942 PubMed, 1314 EMBASE, 438 Web of Science, and 8 LILACS records were retrieved for title and abstract screening; after a multi-step de-duplication pipeline, 2620 remained
- All records undergo double-reviewer screening, with tertiary arbitrators to mitigate any discrepancies

Virulence Factor	Mechanisms of Pathogenesis
HSP23 **HSP60** **HSP70** **HSP83** **HSP90** **HSP100** **HSP65**	• Heat shock proteins • Thermotolerance/survival • Chaperones that facilitate the stabilization of proteins in stressful host environments • Significant expression changes in HSPs as parasite is engulfed in host cells • Aid in adapting from poikilothermic insect vector to a homeothermic mammalian host
LPG	• Lipophosphoglycan • Cell surface anchored molecule • Species-specific sugar component • Required to cause infection in the sandfly hindgut
GP63	• Metalloprotease • Cleaves C3b complement • Halts and hinders innate immunity • Protects parasite from cell lysis
CPB	• Lowered virulence in macrophages • Lowered virulence in mice • Required to cause infection
EF-1alpha	• Elongation factor that is part of the parasite exosome • Blocks Nitric Oxide production • Promotes survival
A2	• Exacerbate parasite-derived immunopathogenesis • Significant in visceral leishmaniasis
MPI	• Catalyze the interconversion of F6P and M6P • Required for glycoconjugates • Loss of MPI leads to loss of surface-anchored VF synthesis, such as leishmanolysin

Species	Virulence Factors
L. donovani	eIF2a, HSP70, HSP90, HSP60, HSP83, HSP65, PDI, LBP, LPG, AHA1, AAH, Rab6, HSP100, CPN10, CPB, CATB, HSP23, sAcP, GF1, KMP-11, GP29, ARF1
L. tropica	PDI, A2, ABCA2, GP63, LPG
L. major	DAT, TACI, ACP, PDI, AP3, CPA, CPB, GP63, LACK, LPG, HASP, SHERP, ISP2, HSP100, HSP70, PTR1, GPI12, MIX, ATG5, MGT1, MGT2, MPK10
L. infantum	PNA, KMP-11, CPC, HSP70, LPG, A2, SIR2, GP63, CPB, PTR1, CFAS, CPA

RESULTS

- Some common parasite-derived pathogenesis mechanisms in *Leishmania* include:
  - Heat shock adaptation to the host environment
  - Evading the immune system
  - Increased expression of survival factors
  - Preventing innate immunity opsonisation
  - Modulation of the host immune system
- Heat shock is mainly directed by heat shock proteins (HSPs):
  - Different HSPs are used preferentially in different species
  - HSP23 can protect against thermal, acidic and oxidative stresses
  - CyP40 is thought to be a co-chaperone that helps the parasite infect macrophages
  - Loss of HSP100 renders *L. major* and *L. donovani* non-infective in vitro at physiological temperatures
- Heat shock and resulting thermotolerance is a crucial method by which *Leishmania* species exert their virulence

DISCUSSION

- The ability to comprehensively synthesize all the known literature around parasite-determined virulence factors can open new doors into network-level pathogenesis
- Connecting the dots between virulence factors (if any) to construct a more complete picture of parasite pathogenesis can help better illuminate the underpinnings of different disease manifestations
- Once all parasite-determined VFs are mapped, it can elucidate how they may tie into host-determined immunopathogenesis mechanisms
- Being able to modulate some of these network-level systems can potentially identify novel targets for therapeutics and diagnostics
- This systematic review has implications for painting a fuller picture of parasite-determined *Leishmania* pathogenesis and hence help tie the ends between different VFs, and maybe shed light into host environmental factors

Accuracy of Diagnostics in Old World Tegumentary Leishmaniasis: A Systematic Review

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BACKGROUND

- Old World Tegumentary leishmaniasis (OWTL) is characterized by cutaneous ulcerative skin lesions, caused by *Leishmania* parasites[1], that can potentially disfigure the midface. The geographically OWTL species include *Leishmania (L) tropica*, *L. major*, and *L. aethiopica*,.
- The clinical presentation of OWTL is like that of epidemiologically overlapping fungal and mycobacterial infections, thereby necessitating confirmatory diagnostics to inform appropriate treatment[1].
- Present laboratory diagnostic techniques for OWTL include the leishmanin skin test (LST); microscopic identification of amastigotes from skin aspirates, biopsies and scrapings; culture; and molecular assays[1].
- Current knowledge regarding the best-performing specimen and diagnostic assay for OWTL diagnosis is inadequate, leading to uncertainty as to what specimen to collect and which test to request when encountering a patient suspected to have OWTL.
- **Our objective was to conduct a knowledge synthesis to determine optimal methods to accurately and efficiently diagnose OWTL to improve diagnostic stewardship.**

METHODS

- We searched five databases from inception to October 28, 2019 including Ovid MEDLINE, Ovid Embase, LILACS, Cochrane Library and Scopus.
- The following search terms were used: ("cut* leish*" OR "muc* leish*" OR "teg* leish*") AND (diagnosis OR diagnostic accuracy OR sensitivity OR specificity OR stard OR test*) AND NOT (viscer*).
- All systematic reviews, diagnostic trials and observational studies were included.
- Titles, abstracts and full-texts are systematically doubled screened by two reviewers with a tertiary arbitrator.
- Full texts were excluded if the species were no human, did not involve the old world tegumentary leishmania case and no laboratory diagnostics or diagnostic reference standard included. Full texts were also excluded if they did not include more than 10 human subjects and consisted of reviews or editorials.
- Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA)[2] and Quality Assessment of Diagnostic Accuracy Studies (QUADAS)[3] are employed.

RESULTS

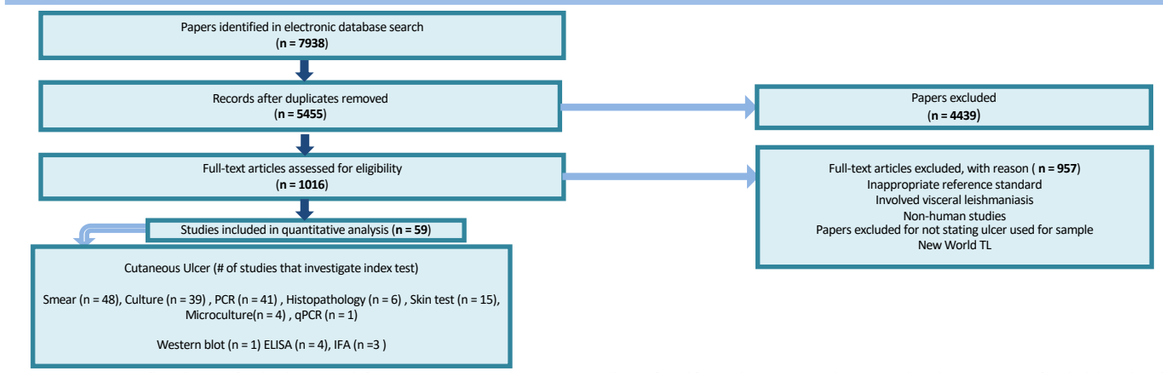

Figure 1. PRISMA flow diagram for database search from inception to October 28, 2019. Full texts found from July 2018 to October 2019 will undergo screening for eligibility. Therefore, the numbers of papers from that stage onwards reflect papers found in a search conducted on October 2019.

Table 1. Descriptive data for eligible full texts including patients with CL, MCL or both

Author, Published Year	Species	Lesion Type	# of Patients	Country	Index Tests	Reference Comparator
Aberra et al., 2019	*L. aethiopica*	CL	124	Ethiopa	TCM with modified Novy-MacNeal-Nicolle (NNN) in tissue culture flask, microcapillary tubes containing RPMI 1640 with 10% fetal bovine serum (FBS) for MCM, Smears (Giemsa)	2/3 test(s) positive (culture, microscpy, smear all from lesion aspirates)
Gunaratna et al., 2018	*L. Donovani*	CL	150	Sri Lanka	Punch biopsies were collected from 150 suspected cutaneous leishmaniasis cases and screened with SpeedXtract/RPA, RNAlater/PCR and ATL buffer/PCR, in addition to Giemsa-stained slit skin smears	3/5 test(s) positive (SSS, SE-RPA, ATL-PCR, RNAlater, PCR)
Khan et al., 2016	*L. Tropica* *L. Major* *L. Infantum*	CL	125	Pakistan	kDNA PCR ITS2 PCR rDNA PCR Microscopy Culture	Only kDNA and rDNA PCR provided significant statistical equivalence with the consensus standard (McNemar's test; P > 0.05)
Kothalawala et al., 2018	L. Donovani	CL	31	Sri Lanka	LAMP assay Nested PCR	Light microscopy, a widely used and universally accepted method was used as the reference standard for confirmation of diagnosis.
Rasti et al., 2016	L. Tropica L. Major	CL	130	Iran	Serosity of ulcer was collected and examined by microscopy, culture, PCR, and nested PCR methods	1/4 test(s) positive (smear, culture, PCR, or nested PCR)
Vink et al., 2018	L. tropica	CL	274	Afghanistan	Loopamp™ Leishmania Detection Kit (Loopamp) and CL Detect™ Rapid Test (CL Detect),	Diagnostic performance of the tests was evaluated against a reference combining microscopy and PCR.

Table 2. Application of QUADAS for full texts investigating CL, MCL or both. There are minimal applicability concerns. However for some studies, the lack of information on patient selection and conduction of index test(s) and reference standard creates uncertainty in bias risk assessment.

Author, Published Year	Risk of Bias				Applicability Concerns		
	Patient Selection	Index Test	Reference Standard	Flow and Timing	Patient Selection	Index Test	Reference Standard
Aberra et al., 2019	O	O	O	?	O	O	O
Gunaratna et al., 2018	?	O	?	?	O	O	O
Khan et al., 2016	X	O	O	?	X	O	O
Kothalawala et al., 2018	X	X	O	O	X	O	O
Rasti et al., 2016	O	O	X	O	O	O	O
Vink et al., 2018	O	X	O	O	O	O	O

O = low risk, **X** = high risk, ? = unclear risk

Table 3. Reported diagnostic performances for papers investigating CL, MCL or both. The objective of all papers were to evaluate sensitivity of diagnostic performance of various index tests, albeit using different materials and protocols.

Author, Published Year	Species	Lesion Type	Reference Comparator	Index Test	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Aberra et al., 2019	*L. aethiopica*	CL	2/3 test(s) positive (culture, microcapillary, smear all from lesion aspirates)	TCM MCM Smears (Giemsa) – Direct Microscopy	69.2 92.3 71.2	98.9 97.8 97.8	97.3 96.0 94.6	84.0 95.7 84.9
Gunaratna et al., 2018	*L. Donovani*	CL	3/5 test(s) positive (SSS, SE-RPA, ATL-PCR, RNAlater, PCR)	SSS SE-RPA ATL-PCR RNAlater PCR	32.2 65.5 92.4 63.4	100 100 100 100	47.5 64.04 89.06 62.64	100 100 100 100
Khan et al., 2016	*L. Tropica* *L. Major* *L. Infantum*	CL	Only kDNA and rDNA PCR provided significant statistical equivalence with the consensus standard (McNemar's test; P > 0.05)	kDNA PCR ITS2 PCR rDNA PCR Microscopy Culture	86.5 70.3 78.4 60.5 67.1	86.5 100 43.2 100 100	92.8 100 73.4 100 100	76.2 62.7 50 51.4 63.2
Kothalawala et al., 2018	*L. Donovani*	CL	Light microscopy, a widely used and universally accepted method was used as the reference standard for confirmation of diagnosis.	LAMP assay Nested PCR	82.6 100	100 100	100 100	66 100
Rasti et al., 2016	*L. Tropica* *L. Major*	CL	1/4 test(s) positive (smear, culture, PCR, or nested PCR)	Microscopic Culture kDNA PCR kDNA-nested PCR	87.9 72.7 99 97	100 100 100 100	100 100 100 100	72.1 53.4 96.9 91.2
Vink et al., 2018	*L. tropica*	CL	Diagnostic performance of the tests was evaluated against a reference combining microscopy and PCR.	CL Detect Loopamp	65.4 87.6	100 70.6	65.3 87.5	100 100

DISCUSSION

- Current laboratory diagnostic techniques for TL include the leishmania skin test (LST) include; microscopic identification of amastigotes from skin aspirates, biopsies, and scrapings; culture; and molecular assays. Microscopy and culture being the "consensus standard" and conventional tests perform poorly especially for cutaneous disease.
- Full-text articles diagnosed CL using **Nested PCR** on the other hand displaying 100% sensitivity, specificity, PPV and NPV values. However, the average time taken for the LAMP assay was 1 hour and 40 minutes when compared to the nested PCR that took approximately 3 hours and 30 minutes (**Kothalawala et al., 2018).**
- Sensitivity and specificity of each diagnostic PCR assay on filter paper samples was assessed against the consensus standard. **kDNA PCR** showed the highest sensitivity (86.5%) and specificity (86.5%). This PCR method was also least prone to producing false negatives (NPV = 76.2%). rDNA PCR was the second most sensitive method (78.4%), although suffered from poor specificity (43.2%). ITS2 PCR was the least sensitive (70.3%) but outperformed other methods in specificity (100%). Parasite **culture and microscopy** provided sensitivity estimates of 67.1% and 60.5%, respectively. Only kDNA and rDNA PCR provided significant statistical equivalence with the consensus standard (McNemar's test; P > 0.05) **(Khan et al., 2016)**
- Based on the results of many articles, PCR had the highest sensitivity and specificity with culture having the lowest sensitivity in the diagnosis of CL.

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A Systematic Review of Ethnopharmaceuticals for the Treatment of New World Cutaneous Leishmaniasis

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Introduction

- **New World Cutaneous Leishmaniasis (NWCL):** neglected parasitic disease caused by members of the genus *Leishmania,* located primarily in Central and South America[1]
- Better drugs needed due to the toxicity, accessibility limits, and expense of first-line treatment options
- **Ethnopharmaceuticals:** plant-based compounds with potential anti-leishmanial effects found in and around local endemic communities[2]
- Potential to overcome the aforementioned therapeutic challenges using ethnopharmaceuticals, are supported by anecdotal evidence of efficacy

Objective: Aim to synthesize existing evidence around available ethnopharmaceuticals, pepper and allium to promote drug discovery for the prevention and treatment of NWCL.

Methods

- PubMed (NCBI), Medline (OVID), Embase (OVID), Web of Science (BioSIS) and LILACS (VHL) were searched using combinations of the search terms **"cutaneous leishmaniasis"** and **"ethnopharmaceuticals"**
- Inclusion and exclusion of search terms was employed to maximize relevant article extraction
- Inclusion criteria: observational studies, case reports, case series, cohort studies, and clinical trials reporting therapeutic outcomes, if possible
- GRADE approach used to assess the quality of studies reporting therapeutic interventions
- LILACS articles screened by native Spanish speaking individuals to ensure proper adherence to inclusion and exclusion criteria
- Data will be grouped and summarized by Leishmania spp. and plant species

Results

Figure 1: Workflow highlighting abstract inclusion and exclusion criteria for full-text review.

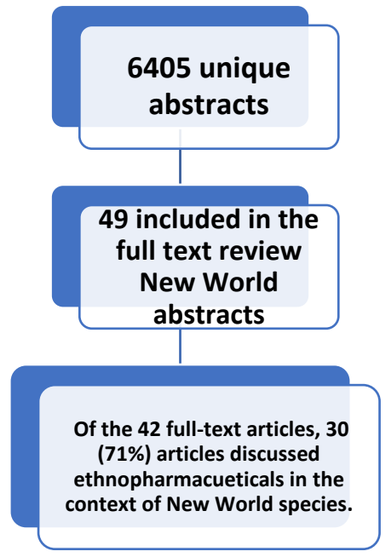

Figure 2: Abstracts discussing ethnopharmacueticals in the context of New World species.

Discussion & Conclusions

- 49 abstracts included for full-text review of NWCL using the GRADE approach from 1957-present (Figure 1 & 2)
- Focus of systematic review will be on the effects of ethnopharmaceuticals in the context of New World species
- Increased human and vector migrations, climate change and travel, and the incidence of CL may increase in non-endemic areas
- Synthesizing current evidence surrounding ethnopharmaceuticals for the treatment of NWCL may contribute to drug discovery pipelines and potentially lead to novel therapeutics

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Transfusion Transmitted Leishmaniasis:
Leishmania Detection in the Blood Supply and Associated Risk Factors

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BACKGROUND

- Blood supply safety has important implications for blood recipients
- Determining *Leishmania* carriage in blood supply of asymptomatic donors in *Leishmania* endemic areas is crucial for recipients' health and limiting transmission
- **Aim: To map the literature on prevalence and detection assays for *Leishmania* detection in blood supplies around the world, which may extrapolate to other non-endemic countries including Canada**

METHODS

- Seven electronic literature databases: Ovid Medline, EMBASE, Global Health, CINAHL Plus, CAB Abstracts, LILACS, and Cochrane Library were searched from database inception to Nov 1, 2019 with restriction to humans only
- A combination of the following search terms: "leishmania" with "blood"; "detection", "diagnosis", "diagnostic accuracy"; "sensitivity", "specificity"; and "smear"; "STARD"; "microscopy", "PCR" were used without language restriction
- Titles, abstracts and full-text articles are systematically screened by 2 independent reviewers, any disagreements were resolved with a tertiary arbitrator
- Inclusion Criteria: 1) *Leishmania* detection 2) Blood 3) Human Systematic reviews, diagnostic trials and smaller observational studies are included
- Data was summarized using qualitative and quantitative measures
- Meta-analysis was performed by comprehensive meta-analysis software using random effects model

RESULTS

Table 1. Prevalence of *Leishmania* by Serology in Asymptomatic Blood Donors

	Sub-analysis Factor	Prevalence
Overall		**6.7% (5.0-8.8%)**
Region	Asia (Bangladesh, Iran, Nepal)	1.2% (0.5-3.0%)
	Europe (France, Greece, Spain, Italy, Turkey)	4.7% (2.7-8.0%)
	South America (Brazil)	10.4% (7.3-14.5%)
Sex	Male	1.4% (1.1-1.7%)
	Female	4.6% (4-5.2%)
Species	Leishmania donovani	7.0% (2.0-12.0%)
	Leishmania infantum	7.0% (5.0-8.0%)

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RESULTS CONT'D – TABLES & FIGURES

Figure 1. PRISMA Flowchart for Identification of Relevant Articles

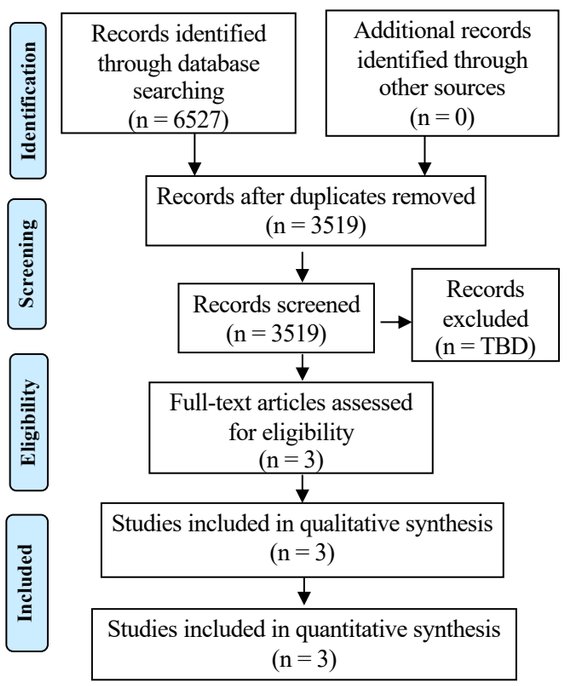

Figure 2. Meta-analysis of Prevalence of *Leishmania* in Blood Donors with Sub-analysis According to Region, Sex, and Species. Overall prevalence was 7%, with Brazil having the highest prevalence. Female blood donors also had a higher prevalence than males

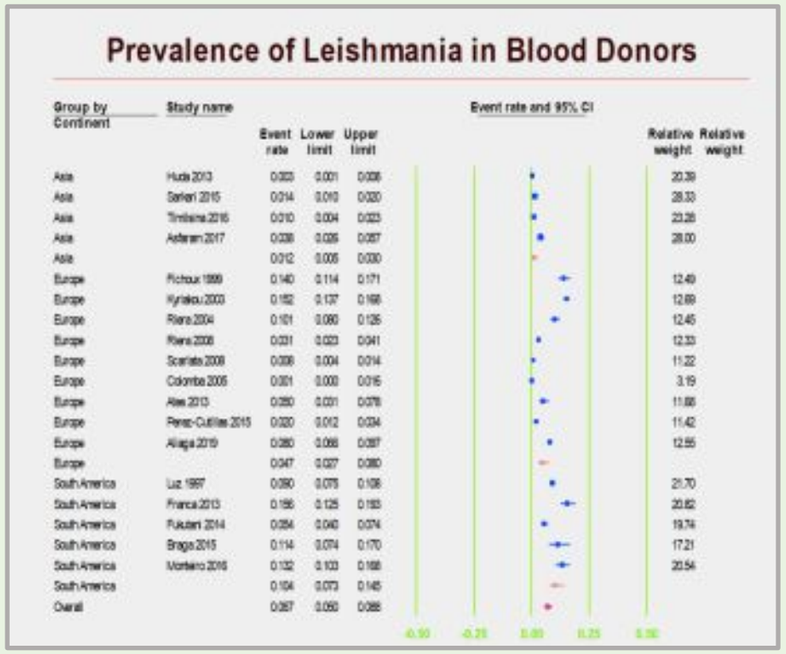

Table 2. Characteristics of Studies in this Systematic Review

Article	Study Design	Study Period	Setting	Sample Size	Sex	Age (years old)	Leishmania species
Asfaram 2017	Systematic Review and Meta-analysis	1997-2016	Brazil, France, Greece, Spain, Italy, Turkey, Bangladesh, Iran, Nepal.	14 243 (16 studies)	Male 69.9% Female 30.1%	16 – 68	Leishmania braziliensis, Leishmania donovani, Leishmania infantum
Asfaram 2017	Cross Sectional	July – Sept 2016	Iran	600 (1 study)	Male 99.3% Female 0.7%	20 – 61	Leishmania infantum
Aliaga 2019	Cross Sectional	June 2015 – May 2016	Spain	1260 (1 study)	Male 48.1% Female 51.9%	18 – 65	-

CONCLUSION

- **Overall prevalence of *Leishmania* in asymptomatic blood donors was about 7%**
- **Highest prevalence was in South America – Brazil (10.4%) and lowest in Asia (1.2%)**
- **Higher detection of *Leishmania* found in female donors**
- ***Leishmania donovani* and *Leishmania infantum* were the primary associated species with *Leishmania braziliensis* (Table 2) also present in the Brazilian population**
- **These data can inform guidelines and policy amendments in blood donor centres**

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- Aliaga L *et al*., Asymptomatic *Leishmania* infection in blood donors from the Southern of Spain. Infection 2019 (47) 739-747

Sequence Analysis of *Plasmodium falciparum* histidine-rich protein 2 and 3 genes from returning travelers to Africa

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Background:

- WHO reported 229 million new cases of malaria with half a million deaths in 2019
- Sub-Saharan Africa and Children under 5 accounted for majority of cases and deaths.
- 5 human species – *Plasmodium falciparum, P. vivax, P. ovale, P. malariae, P. knowlesi*
- *Plasmodium falciparum* accounts for 99% of Malaria deaths
- Malaria is not endemic in Canada but we have competent vectors for transmission
- Travelers from endemic areas account for 500 cases/year
- Current diagnosis of Malaria in Canada include microscopy, rapid diagnostic test by antigen detection of histidine-rich proteins 2 and 3, and PCR
- We aimed to study the sequence heterogeneity of histidine-rich protein 2 and 3 genes in *Plasmodium falciparum* cases which failed detection by HRP2 specific rapid diagnostic test

Materials and Methods:

- *Plasmodium falciparum* whole blood samples from cases which failed BinaxNow rapid diagnostic test were identified
- DNA extraction was performed and species were confirmed by real time PCR
- Plasmodium histidine-rich protein 2 (*pfhrp2*) and 3 (*pfhrp3*) genes were PCR amplified and Sanger Sequenced
- Sequence alignment and analysis were performed by MEGA 6.06 software

Figure 1. Real time PCR to confirm malaria species

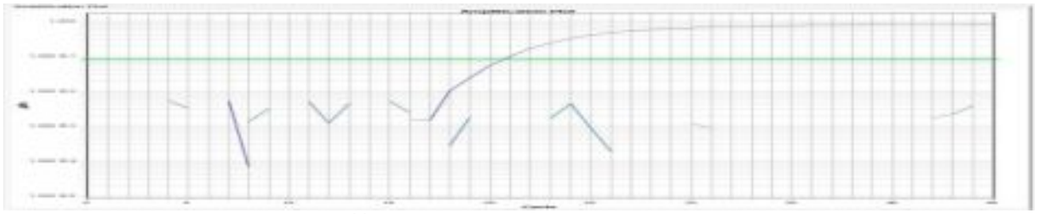

Figure 2. DNA sequence generated by Sanger Sequencing

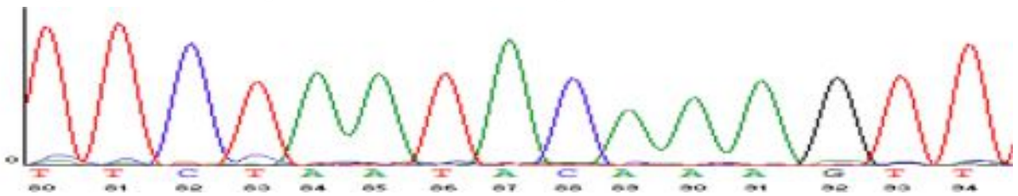

Table 1. Deletion of *pfhrp2* and *pfhrp3* genes described in the literature

Gene	basepair	Gene Structure	Expression	Mutation/Deletion
Pfhrp2	1064	2 exons, 1 intron	HRP2	Deletion (Brazil, Peru) Partial Deletion (Bangladesh) Partial and full deletion (Kenya)
Pfhrp3	983	2 exons, 1 intron	Epitopes homologous to HRP2	Deletion (Brazil, Peru) Mutation (Bangladesh)

Results:

- Four *Plasmodium falciparum* cases that failed BinanxNow rapid diagnostic test were identified
- Real time PCR confirmed three were mono-*Plasmodium falciparum* infections whereas one was a mixed infection

Table 2. *Plasmodium falciparum* cases that failed BinaxNow rapid diagnostic test
TBD – To be determined

Case	Travel (Country of Acquisition)	Year	Age	Sex	Microscopy	Parasitaemia	BinaxNow T1 (PfHRP2)	BinaxNow T2 (Pan-Aldolase)	*Pfhrp2*	*Pfhrp3*
1	Nigeria	2016	39	M	*Pf*, Rings	<0.1%	-	-	No mutation compared to control (400bp of exon2)	No mutation compared to control (400bp of exon 2)
2	Nigeria	2017	42	M	*Pf*, Rings	<0.1%	-	+	No mutation compared to control (700bp of exon 2)	TBD
3	Tanzania	2017	13	M	Neg but *Pf* PCR Pos, low level infection	N/A	-	-	TBD	TBD
4	Ivory Coast	2018	50	M	*Pf* and *Pm* mixed infection. Rings, Trophozoites, Schizonts	0.2%	-	-	TBD	TBD

Conclusions:

- No mutation was found in exon 2 region of pfhrp2 and pfhrp3 DNA sequence analysis for the cases where primary material was available for sequencing
- Detection failure may be due to low expression of PfHRP2 -> detection failure by BinaxNow RDT
- In non-endemic lab setting with waning microscopy expertise
  Ambiguous microscopy smear + PfHRP2 RDT Neg may lead to:
  -> Misdiagnosis as non-*falciparum* Malaria or Neg for Malaria
  -> Inappropriate treatment with Chloroquine for presumed *Plasmodium vivax* or other non *falciparum* malaria
  -> Outcome can be detrimental (Death)!

Future Analysis:

- Further DNA analysis of exon 1 and full exon 2 of both genes
  -> Insight on any sequence heterogeneity that may affect binding to the RDT
- Analysis of remaining PfHRP2 RDT neg Plasmodium falciparum cases
  -> Etiology of detection failure
- Continual Surveillance
  -> Inform lab diagnostics and physicians of potential pitfalls
  -> Possible modification in diagnostic algorithm

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Reactivation of Old World Tegumentary Leishmaniasis following Iatrogenic Immunosuppression: A Systematic Review of Secondary Prophylaxis

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Introduction

- Old World Leishmaniasis is a neglected yet detrimental disease caused by protozoal species of the *Leishmania* genus including *L.major, L. tropica, L. aethiopica* and *L. infantum.*
- Although endemic to the Middle East, the Mediterranean basin, the Arabian Peninsula, Africa and the Indian Subcontinent, recent increases in migration and travel have contributed to the disease's spread into non-endemic areas.
- The possibility of leishmaniasis reactivation in the context of immunosuppressive therapy heightens due to alterations in immunological control, thus posing a potential problem for rapid diagnosis and treatment of patients with a prior history of leishmaniasis.[1]

Objective: We aim to synthesize available data in order to guide healthcare providers on the clinical management of patients who require iatrogenic immunosuppressive treatment and have a history of leishmaniasis.

Methods

- PubMed (NCBI), Medline (OVID), Embase (OVID), Web of Science (BioSIS) and LILACS (VHL) were searched for between inception to November 15, 2020 with combinations of the search terms "Leishmania reactivation", "Leishmaniasis" and "Immunotherapy".
- Case series, case reports, cohort studies, clinical trials and relevant systematic reviews and meta-analyses will be included in this systematic review.
- Quality assessment of studies reporting therapeutic interventions will be conducted using the GRADE approach.[2]
- LILACS articles will be assessed by Spanish speaking individuals to ensure accurate rating of the inclusion and exclusion criteria.

Results

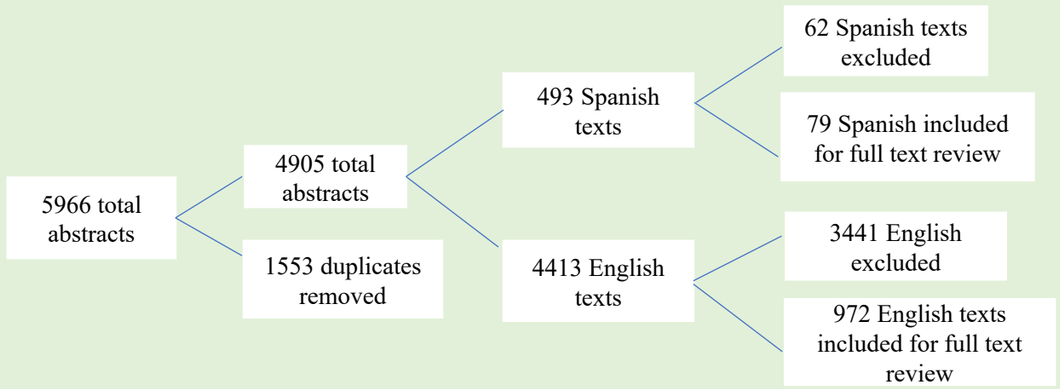

Figure 1: Workflow highlighting title and abstract screening as well as full text screening

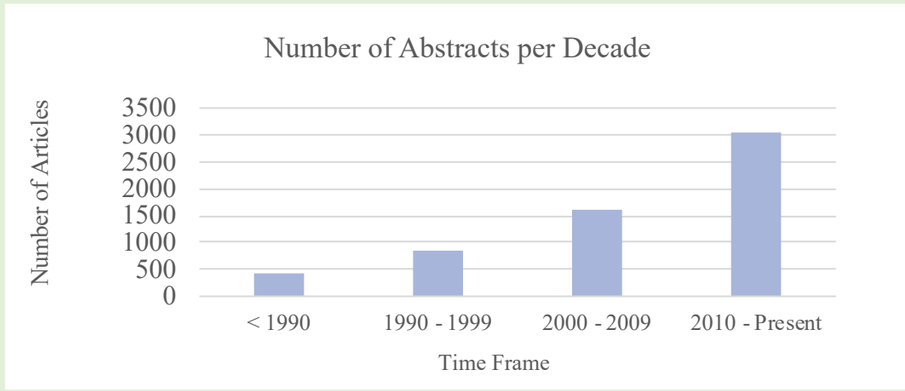

Figure 2: Abstracts by time frame.

Discussion & Conclusions

- Of 4905 abstracts screened, 1051 have progressed to full text review and 3503 have been excluded for not meeting inclusion criteria. English text will begin to undergo full-text screening while the remaining 352 Spanish text will undergo completion of title and abstract screening. (Figure 1 and 2).
- Iatrogenic immunosuppression in patients with a history of leishmaniasis could result in reactivation due to a loss in host control of parasite replication.
- Synthesis of related data can further our understanding of the relationships between iatrogenic immunosuppressive treatment and leishmaniasis reactivation, as well as the role of secondary prophylaxis.
- Necessary information required by healthcare providers will be provided to guide the clinical management of this patient population in advance of immunosuppression in order to reduce the risk of this detrimental disease's reactivation.

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Treatment of Intestinal Protozoa in Pregnancy: A Systematic Review of Maternal, Fetal and Infant Outcomes

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Background:

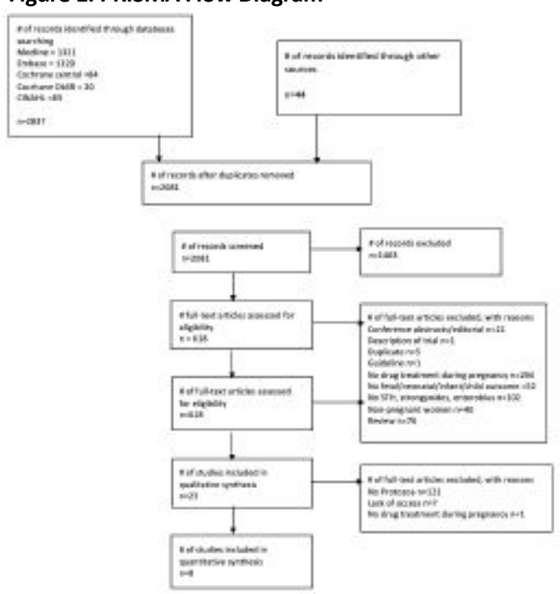
- Treating parasitic infections during pregnancy warrants a consideration of many factors including drug safety, efficacy, and potential impact on maternal and fetal outcomes
- A substantial knowledge gap exists in the treatment of intestinal protozoa infections during pregnancy
- We aimed to map the available literature on the safety, efficacy, maternal and fetal outcomes of the use of metronidazole as a treatment for intestinal protozoal infections during pregnancy.

Methods:

- A literature search was conducted on Medline, EMBASE, CINAHL, Cochrane Library of Systematic Reviews and CENTRAL databases from database inception to March 2020
- Systematic reviews, randomized controlled trials, cohort studies, smaller observational studies, case-control studies, case series, and case reports were screened
- Inclusion criteria were as follows:
  1) Metronidazole treatment during pregnancy;
  2) Diagnosis of intestinal protozoa during pregnancy ;
  3) Maternal, fetal, or child outcome post drug treatment in pregnant women
- Data were extracted from articles and study quality was assessed using the GRADE approach

Results:

Figure 1. PRISMA Flow Diagram



- A total of 2837 articles were retrieved from literature databases and other sources. After title and abstract screening, 618 full text articles were assessed for eligibility and a total of 8 studies were deemed eligible for data extraction.
- Only a limited number of case reports on intestinal protozoa during pregnancy was available for our review.
- For the treatment of amoebiasis with metronidazole during pregnancy, data on maternal and fetal outcomes such as preterm labour, live births, and cesarean delivery will be synthesized

Table 2. Data Synthesis Table

Study	Study Period	Study Population	Study Design	Maternal Trimester of Drug Treatment	Maternal Treatment and Sample Size	Baseline Prevalence of Protozoa in Pregnant Participants (N>1)	Outcomes (Maternal/Fetal/Infant)
Read, 2001 (A14)	N/A	Pregnant woman in third trimester (31 weeks), 37 years old,	Case report: 1 pregnant female	3rd trimester	N=1 Pre labour: Metronidazole and IV Ceftriaxone Post labour: 14 day course of metronidazole + 10 day course of diloxanide furoate	E.Histolytica from serologic tests	Maternal outcomes: Premature labour (32 weeks gestation), complete resolution of liver abscess Fetal Outcomes: Live birth, normal birth weight
Mitchell 1984, (A16)	N/A	Two pregnant women. A 20 year old (patient 1) and a 29 year old (patient 2) whose amoebiasis test were positive	Case Report	Patient 1: 3rd trimester Patient 2: 3rd trimester	Patient 1: intravenous metronidazole and ampicillin Patient 2: intravenous metronidazole and penicillin	N/A	Patient 1: Emergency caesarean section resulted in the birth of a live male infant weighing 3200g Patient 2:The patient remained well and the pregnancy progressed normally until spontaneous rupture of the membranes at term was followed by the normal vaginal delivery of a live female infant weighing 2800 g
Lugo 1981 (A23)	N/A	Pregnant women with gestational age from 9 to 39 weeks. Women diagnosed with amoebic liver abscess and had complications	Case Series	1st-3rd trimester Metronidazole	N=7 Group 1: Metronidazole 400mg/daily + estrogen 100mg/daily for 10 days (n=2) Group 2:Metronidazole 400mg/daily for 10 days (n=5).	All had amoebic liver abscess.	2 mothers died in the Metronidazole only group, 2 days and 3 days after treatment. 5 fetal deaths and 2 live births.

Conclusion:

- There is a significant paucity of information on metronidazole use in pregnancy thus, a broader study on the use of metronidazole in pregnancy for all indications will be considered. With increased international travel and migration, health practitioners will encounter pregnant patients with intestinal protozoal infections. Therefore, synthesizing the current literature on the treatment of such infections during pregnancy can inform treatment, management strategies and referral decisions in pregnancy care.

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Treatment of Schistosomiasis in Pregnancy: A Systematic Review of Fetal and Infant Outcomes

Vivian Chen[1], Alexandra Atayde[1], Hira Raheel[1], Rachel Lau[2], Shveta Bhasker[1], Chelsia Watson[1], Robert B. Chris[1], Swana Kopalakrishnan[1], Leila Makhani[1], Sharmistha Mishra[3], Andrea K. Boggild[1,2]

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BACKGROUND:

- Schistosomiasis remains one of the most prevalent parasitic infections, with an estimated 261 million infected worldwide,[1] and has significant economic and public health consequences.
- Treatment of parasitic infections in pregnancy necessitate considerations of numerous factors, including the potential developmental outcomes for the fetus and newborn.
- A substantial knowledge gap exists in the treatment of schistosomiasis infections during pregnancy, with few published and authoritative resources to guide clinical decision-making.

OBJECTIVE:

- To map the available literature regarding the safety of intestinal schistosomiasis treatments during pregnancy, namely praziquantel, for fetal and infant development.

METHODS:

- A literature search was conducted on Medline, Embase, CINAHL, Cochrane DbSR and Cochrane Central databases with the search terms "intestinal parasites," generic and organism specific; and "pregnant/pregnancy" from database inception to March 2020 without language restrictions.
- Duplicate articles were removed and title, abstract and full-text articles were systematically double-screened and arbitrated by a third reviewer.
- Systematic reviews, randomized controlled trials, cohort studies, smaller observational studies, case series and case reports assessing or reporting the efficacy, safety, or tolerability of praziquantel treatment during pregnancy were screened.
- Inclusion criteria: Pregnant women + Treated with praziquantel during pregnancy + Schistosomiasis + Fetal and/or infant Outcome(s) reported.
- Two independent reviewers extracted the data and assessed quality using the GRADE approach. Risk of bias for each study was determined.
- Data were summarized using qualitative and quantitative measures for safety of praziquantel on the fetus and infant.

RESULTS:

Figure 1. PRISMA Flow Diagram

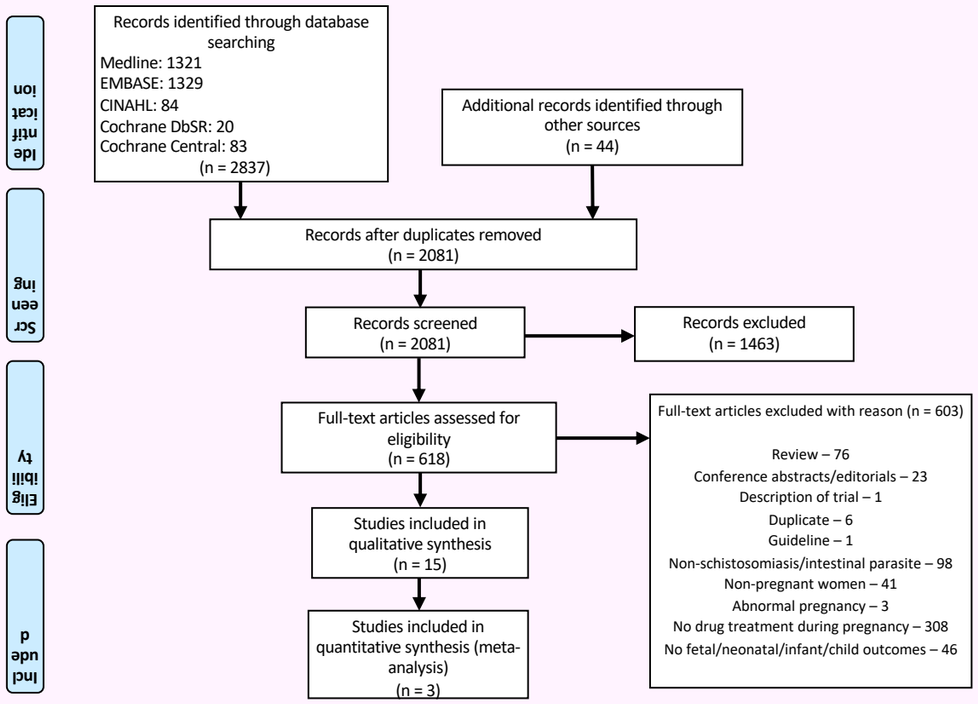

Table 1. Fetal and infant outcomes following praziquantel treatment in pregnant mothers with *S. mansoni* compared to placebo

Fetal and Infant Outcomes	Study Design and Sample Size	Effect of Maternal Praziquantel Treatment Compared to Placebo	Certainty of Evidence (GRADE)
Birth weight; low birth weight (<2.5kg); very low birth weight (<1.5kg)	1 RCT; n = 1953	No difference in birth weight, nor were there differences in incidence of low birth weight and very low birth weight babies.	⊕⊕⊕◯ MODERATE [b]
Height and weight at 15 months	1 RCT; n = 483	No difference in height and weight of infants measured at 15 months.	⊕⊕⊕◯ MODERATE [a]
Fetus small for gestational age	1 RCT; n = 370	No difference in incidence of fetus being small for gestational age.	⊕⊕⊕⊕ HIGH
Apgar score at 10 minutes	1 RCT; n = 483	No difference in Apgar score measured at 10 minutes.	⊕⊕⊕◯ MODERATE [a]
Live birth rate	1 RCT; n = 366	No difference in live birth rates.	⊕⊕⊕⊕ HIGH
Stillbirth at >20 weeks gestation	2 RCTs; n = 2759	No difference in incidence of stillbirths.	⊕⊕⊕◯ MODERATE [c]
Unhealthy newborn	1 RCT; n = 366	No difference in newborn health.	⊕⊕⊕◯ MODERATE
Congenital anomalies	2 RCT; n = 2726	No difference in incidence of congenital anomalies.	⊕⊕⊕◯ MODERATE
Serious infant adverse events	1 RCT; n = 362	No difference in incidence of serious infant adverse events.	⊕⊕⊕◯ MODERATE
Early neonatal death (<7 days)	1 RCT; n = 2345	No difference in incidence of early neonatal death.	⊕⊕⊕⊕ HIGH
Infant cytokine levels (IFN-γ; IL-1, 2, 4, 5, 6, 10, 12, 13; CXCL8, 9; TNF; sTNFRI; sTNFII; IFN-γ:IL-4 ratio)	1 RCT; n = 238	No difference in infant cytokine levels.	⊕⊕⊕⊕ HIGH
Hemoglobin levels (in newborn; in cord blood; in infant at 1 year)	1 RCT; n = 1342 1 RCT; n = 483	No difference in hemoglobin levels measured in newborns, in cord blood nor in infants at 1 year.	⊕⊕⊕⊕ HIGH; ⊕⊕◯◯ LOW [e]; ⊕⊕⊕◯ MODERATE [a]
Newborn serum transferrin receptor level; newborn serum ferritin levels; newborn transferrin receptor:ferritin ratio)	1 RCT; n = 361	No difference in serum transferrin receptor levels of newborns, serum ferritin levels nor transferrin receptor:ferritin ratio.	⊕⊕⊕⊕ HIGH; ⊕⊕⊕◯ MODERATE [d] ⊕⊕⊕⊕ HIGH
Non-anemic at 6 months; non-anemic at 12 months	1 RCT; n = 361 1 RCT; n = 303	No difference in incidence of non-anemic babies, measured at 6 months and 12 months.	⊕⊕⊕⊕ HIGH
Iron-deficiency anemia at 6 months; iron-deficiency anemia at 12 months	1 RCT; n = 320 1 RCT; n = 304	No difference in incidence of iron-deficiency anemia, measured at 6 months and at 12 months.	⊕⊕⊕⊕ HIGH
Non-iron-deficient anemic at 6 months; non-iron-deficient at 12 months	1 RCT; n = 314 1 RCT; n = 310	No difference in incidence of non-iron-deficient anemia, measured at 6 months and at 12 months.	⊕⊕⊕⊕ HIGH

GRADE Working Group: Grades of Evidence

- **High certainty:** We are very confident that the true effect lies close to that of the effect estimate.
- **Moderate certainty:** We are moderately confident in the effect estimate: The true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different.
- **Low certainty:** Our confidence in the effect estimate is limited: The true effect may be substantially different from the estimate of the effect.
- **Very low certainty:** We have very little confidence in the effect estimate: The true effect is likely to be substantially different from the estimate of effect.

Explanations

- Nampijja (2012) had about 50% of loss to follow up, through characteristics of the remaining cohort population in this study were similar.
- Ndibazza (2010) had about 15-20% incomplete report of birth weight (reporting bias).
- Data discrepancy in Olveda (2016) for fetal death in utero.
- Had a wide 95% CI.
- Ndibazza (2010) had about 40% incomplete reporting of cord blood hemoglobin.

CONCLUSION:

- Praziquantel administration during pregnancy for the treatment of *S. mansoni* does not appear to have any adverse birth outcomes for the fetus/infant nor lead to any other adverse outcomes for the child later in life.
- Synthesizing the current literature on the treatment of schistosomiasis may improve the effects of pregnancy care.

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1) WHO. Female genital schistosomiasis. A pocket atlas for clinical health-care professionals. Geneva: World Health Organization, 2015

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Clinical Medicine
Group 6

Treatment of Schistosomiasis in Pregnancy: A Systematic Review of Maternal Outcomes

Chelsia Watson[1], Alexandra L. Atayde[1], Rachel Lau[2], Melissa Phuong[1], Yashvi Bharwada[1], Robert Chris[1], Swana Kopalakrishnan[1], Leila Makhani[1], Sharmistha Mishra[3,4], Andrea K. Boggild[1,2,4]

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INTRODUCTION

- Parasitic infections in pregnancy necessitate consideration of numerous factors including the potential safety, efficacy, and tolerability of antiparasitic drugs for the mother
- A substantial knowledge gap exists in pregnancy-associated schistosomiasis, with few definitive resources to guide clinical decision-making

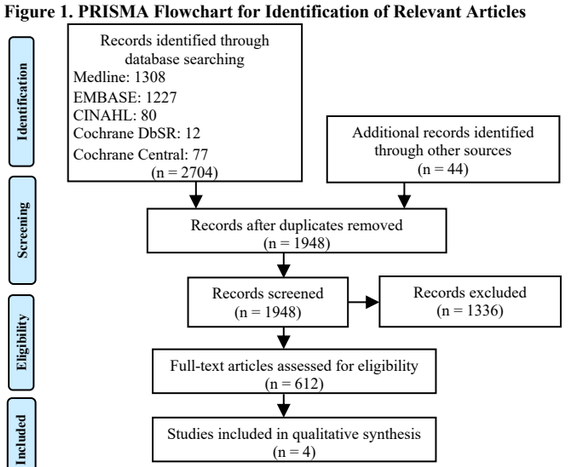
Aim: To systematically map the available literature regarding the efficacy, safety, and tolerability of treatment of intestinal schistosomiasis in pregnancy.

METHODS

- Five electronic databases were searched and titles, abstracts, and full-texts of included studies and reviews were screened from database inception to March 2020, without language restriction
- Systematic reviews, randomized controlled trials, cohort studies, smaller observational studies, case-control studies, case series, and case reports assessing or reporting the efficacy, safety, or tolerability of praziquantel treatment during pregnancy were screened
- Inclusion criteria: Pregnant + Treated with praziquantel during pregnancy + *Schistosoma* Infection + Maternal Outcome(s) reported
- Two independent reviewers screened and extracted the data and assessed quality using the GRADE approach. Risk of bias for each study was determined
- Data were summarized using qualitative and quantitative measures for schistosomiasis as well as efficacy and safety of praziquantel

RESULTS

Figure 1. PRISMA Flowchart for Identification of Relevant Articles



CONCLUSION

- **Praziquantel had a high cure rate of >80% for *Schistosoma mansoni* and *Schistosoma japonicum* infection in pregnant women.**
- **No adverse effects on endotoxin levels, or weight gain were observed.**
- **Treatment with praziquantel during pregnancy did not affect maternal anemia or Hb levels.**

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2. Olveda, Remigio M, et al. Effect of Praziquantel Treatment of Schistosoma Mansoni during Pregnancy On Immune Responses to Schistosome Antigens and Among the Offspring: Results of a of a Randomised, Placebo-Controlled Trial." *The Lancet Infectious Diseases,* 2015:16(**2**)(199-208).
3. Tweyongyere, Robert, et al. "Effect of praziquantel treatment of Schistosoma mansoni during pregnancy on intensity of infection and antibody responses to schistosome antigens: results of a randomised, placebo-controlled trial." *BioMed Central Infectious Diseases*, 2009:9(**32**).
4. McDonald, Emily a., et al. "Endotoxin at the Maternal–Fetal Interface in a Resource-Constrained Setting: Risk Factors and Associated Birth Outcomes." *American Journal of Tropical Medicine and Hygiene,* 2018:99(**2**)(495-501).

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Table 1. Characteristics of Studies included in this study

Study and Design	Study Period	Study Setting	Study Population	Name of Drug and Trimester of Drug Treatment	Sample Size
Ndibazza 2010[1] RCT	April 2003-November 2005	Uganda	Healthy pregnant women	Albendazole; Praziquantel 2nd or 3rd	N=2515 Albendazole (400mg, single dose) + Praziquantel (40mg/kg), N= 628. Albendazole + Placebo, N= 629. Praziquantel + Placebo, N= 628. Placebo + Placebo, N= 630. All single dose. All women received month's supply of daily ferrous sulphate (200mg); 60mg elemental iron); and intermittent sulfadoxine-pyrimethamine for malaria twice after 1st trimester.
Olveda 2015[2] RCT	Not reported	Philippines	Pregnant women infected with *S. japonicum* at 12-16 weeks gestation	Praziquantel 2nd	N=370 Over-encapsulated praziquantel, N=186 (30mg/kgx2 as a split dose over 3h Over-encapsulated placebo (dextrose), N=184 (30mg/kgx2 as a split dose over 3h
Tweyongyere 2009[3] (Nested Cohort of Ndibazza 2010[1]) RCT	November 2003-November 2005	Uganda	Pregnant women with *S. mansoni* infection Exclusion: Pregnancy not normal, history of adverse reactions to anthelminthic, evidence of helminth-induced disease requiring immediate treatment, participation in the study during an earlier pregnancy	Praziquantel 2nd or 3rd	N= 387 Praziquantel, N=186 (40mg/kg, single dose) Placebo, N=201 (dose not stated, single dose)
McDonald 2018[4] (Same trial as Olveda 2015[2]) RCT	Not reported	Philippines	Same as Olveda 2015[2]	Praziquantel 2nd	N=370 Over-encapsulated praziquantel, N=186 (30mg/kgx2 as a split dose over 3h) Over-encapsulated placebo (dextrose), N=184 (30mg/kgx2 as a split dose over 3h)

Table 2. Summary of Findings Table of Praziquantel Compared to Placebo Treatment for Intestinal Schistosomiasis During Pregnancy

Praziquantel Compared to Placebo during Pregnancy

Patient or population: Pregnant women in their 2nd or 3rd trimester
Setting: Developing Countries - Uganda (Ndibazza 2010[1], Tweyongyere 2009[3]) and Philippines (Olveda 2015[2], McDonald 2018[4])
Intervention: Praziquantel
Comparison: Placebo

Outcomes	Anticipated absolute effects* (95% CI)		Relative effect (95% CI)	№ of participants (studies)	Certainty of the evidence (GRADE)	Comments
	Risk with placebo	Risk with Praziquantel				
Anemia at delivery (hemoglobin <11.2g/dL)[1]	349 per 1,000	**349 per 1,000** (307 to 394)	**RR 1.00** (0.88 to 1.13)	1918 (1 RCT)	⊕⊕⊕◯ MODERATE [a]	No difference in maternal anemia
Schistosoma mansoni prevalence at delivery[1]	213 per 1,000	**47 per 1,000** (36 to 64)	**RR 0.22**# (0.17 to 0.30)	2051 (1 RCT)	⊕⊕⊕⊕ HIGH [a]	Praziquantel decreased the prevalence of Schistosoma mansoni at delivery
Mean hemoglobin levels (g/dL) at delivery[1]	The mean mean hemoglobin levels (g/dL) at delivery (Ndibazza 2010) was **0**	MD **0.2 higher** (0.05 lower to 0.45 higher)	-	930 (1 RCT)	⊕⊕⊕◯ MODERATE [a]	No difference in hemoglobin levels
Mean hemoglobin levels (g/dL) at 3rd trimester[2]	The mean mean hemoglobin levels (g/dL) at 3rd trimester (Olveda 2015) was **0**	MD **0.01 higher** (0.24 lower to 0.26 higher)	-	370 (1 RCT)	⊕⊕⊕⊕ HIGH	No difference in hemoglobin levels
Mean weight gain from 2nd to 3rd trimester (kg/week)[2]	The mean mean weight gain from 2nd to 3rd trimester (kg/week) (Olveda 2015) was **0**	MD **0.01 lower** (0.04 lower to 0.02 higher)	-	370 (1 RCT)	⊕⊕⊕⊕ HIGH	No difference in mean weight gain
Cure rate of Schistosoma japonicum at 6-10 weeks post treatment[2]		83.7% (154/184)	not estimable	(1 RCT)	-	
Cure rate of of Schistosoma mansoni at 6 weeks post treatment[3]		81.9% (104/127)	not estimable	(1 RCT)	-	
Endotoxin levels in peripheral blood, cord blood or maternal-fetal interface[4]			not estimable	(1 RCT)	-	Endotoxin levels not associated with praziquantel (no raw data available)

*The **risk in the intervention group** (and its 95% confidence interval) is based on the assumed risk in the comparison group and the **relative effect** of the intervention (and its 95% CI).

CI: Confidence interval; **RR**: Risk ratio; **MD**: Mean difference

GRADE Working Group grades of evidence
High certainty: We are very confident that the true effect lies close to that of the estimate of the effect
Moderate certainty: We are moderately confident in the effect estimate: The true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different
Low certainty: Our confidence in the effect estimate is limited: The true effect may be substantially different from the estimate of the effect
Very low certainty: We have very little confidence in the effect estimate: The true effect is likely to be substantially different from the estimate of effect

Explanations

a. Ndibazza 2010 had about 20% incomplete report of outcomes in both arms (reporting bias)
^ Strong association, RR <0.5 or >2

A Systematic Review of Treatment Strategies for Ingested Marine Toxins

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Background:

- Marine toxins are concentrated in contaminated seafood worldwide.
- Marine intoxications can cause neurological, gastrointestinal, and cardiovascular syndromes, potentially leading to high mortality and lasting morbidity.
- With increasing seafood consumption, globalization, and climate change, there is an increased risk of exposure to these toxins.
- We aim to synthesize existing evidence around diagnosis, treatment, and prevention of marine intoxications into a clinical resource.

Methods:

- Four electronic databases were searched: PubMed (NCBI), Medline (OVID), EMBASE (OVID), and BioSIS (Web of Science) from database inception to February 2021, using combinations of the search terms 'marine' and 'intoxication'
- Iterative inclusion and exclusion of search terms were employed to maximize article extraction
- The search was refined to humans only
- We included observational studies, case reports, case series, and cohort studies, as well as clinical trials and therapeutics tolerability and efficacy
- Abstracts and full-text articles will be systematically double screened by two reviewers and subsequently by a tertiary arbitrator
- The GRADE approach will be employed to assess the quality of studies reporting therapeutic interventions
- Evidence will be summarized using descriptive measures for each intervention type
- Data will be grouped and summarized for ease of clinician use by marine toxin, intoxication syndrome, prevention and therapeutic strategies, and according to geographic location and implicated seafood species.
- Meta-analysis will be planned if sufficient efficacy measures exist

Results:

collated from analysis of 109 abstracts selected for full text review up until November 12, 2019

Type of Intoxication or Syndrome	Type of Toxin Isolated	Seafood Type/Species Ingested	Causative Agent	Countries/Regions Reporting	Treatments
Paralytic	Saxitoxin Endotoxin	Mussels Shellfish Softshell Clams Bivalve Mollusc Finfish Oysters Perna viridis Ostea iridescens Anadara similis Anadara tuberculosa Modiolus sp. Pilecopurpura columellans Gastropds Atlantic Mackeral Scomber scombrus Purple clam (Hiatula diphus)	Gonyaulax tamarensis Dinoflagellates Diatomes Vibrio Norwalk virus Alexandrium catenella/tamarensis Plankton Pyridinium bahamense Algal bloom Prorocentum micans Gymnopdium catenatum	Canada USA Italy Japan Mexico Trinidad El Salvador Taiwan	Symptomatic Exposure Resistance
Neurotoxic	Tetrodotoxin Palytoxin Clupeotoxic	Puffer fish Lagocephalus scleratus (Takifugu oblongus) Crab (Demania reynaudi) Moray fish		Turkey Bangladesh Japan Phillipines	
	Botulism (Type E)	Salted Fish Faseikh Salmo trutta(trout) Fermented seal, arctic fish Whale White fish Kapchunka	Clostridium botulism	Egypt Arab Gulf Canada	Antitoxin polyvalent Supportive
	Brevetoxin		Red Tide Gymnodinium breve		
Diarrhetic	Gempylotoxin Enterotoxin	Oysters Escolar Shrimp Bivalve mollusk Seaweed	Aeromonas hydrophilia Wax esters Vibrio cholerae Dinophysis forti Dinophysis acuminate Enteric viruses Hepatitis A Vibrio alginolyticus Vibrio parahaemolyticus Vibrio Cholerae Vibrio vulnifus	USA Bangladesh Portugal Adriatic Sea Sardinia	
Amnestic		Molluscs Scallops Mussels Oysters Clams	Domoic Acid Diatom Nitzchia pungens	Belgium Canada Angola	
Ciguatera	Ciguatoxin	Tropical Fish Barracuda Grouper Amberjack Snapper Shark Coral Reef fish Turtle Sardine Epipephelus fuscoguttatus	Dinoflagellate Agmbierdiscus toxicus	France Caribbean USA Mexico Puerto Rico South Pacific Islands Madagascar	IV mannitol
Scromboid		Herring Tuna Mahi Mahi	Biogenic Amine Histamine	Russia USA (imported from Vietnam)	Temperature control Anti-histamines
Allergic		Anisakis simplex		Spain	
Hepatotoxic		Sea hare Aplysia kurodai	aplysianin	Japan	IV fluids IV glyzirrhizin
Cytotoxic		Seafood	cyanobacteria	Brazil Australia	
Myotoxic		Buffalo Fish	Haff/rhabdomyolysis	USA	
Heavy Metal	Mercury	Shark Osteichthyes Tuna Bivalve (Marcia optima) Fish (Mullet, Tarli, Surmai, Dahiar) Blackshrimp Sushi/Sashimi Sportfish Crustacea Swordfish		Canary Islands Pelagic Ethiopia Finland Baltic Sea Thailand Colombia Pakistan Brazil USA Turkish Sea Iraq	
	Arsenic	Fish (Cirrhinus reba)		Pakistan Korea Belgium	
Cardiotoxic		Fish	Polychlorinated biphenyls	Sweden	

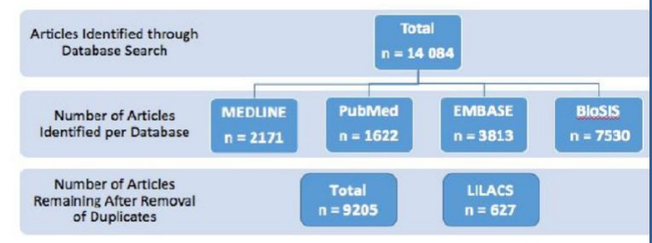

Figure 1: Workflow highlighting breakdown of abstracts by database

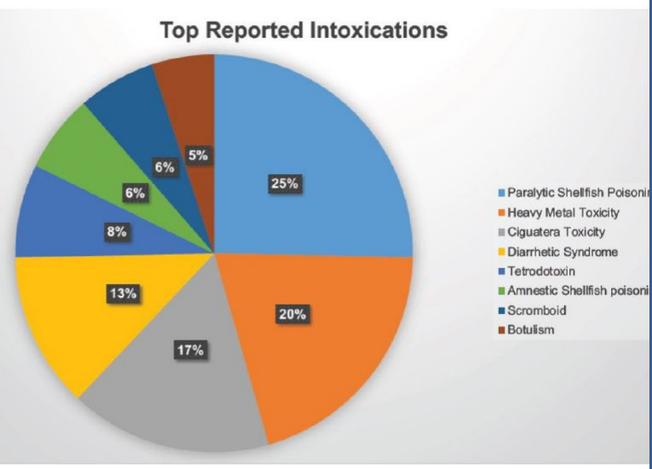

Figure 2: Geographical Distribution of Reported Marine Intoxications

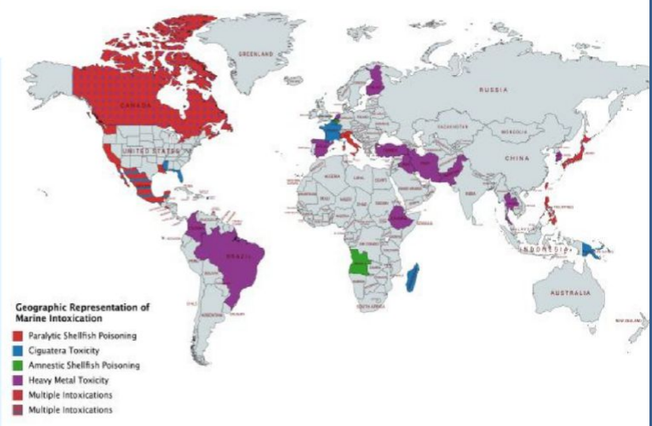

Table 1: Summarized Data of Marine Intoxications from Analyzed Abstracts

Discussion and Conclusion:

- Increased transoceanic movement of people and seafood has facilitated the distribution of contaminated seafood to non-endemic regions where clinicians lack familiarity with intoxication syndromes and appropriate treatment.
- Paralytic shellfish poisoning, ciguatera toxicity, and mercury poisoning are common causes of ingested marine intoxication; usually implicated by contaminated shellfish, large predatory reef fish, and tuna respectively.
- By synthesizing the evidence, we hope to inform the development of appropriate management and risk mitigation protocols.

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A Systematic Review of Treatment Strategies for Percutaneously Introduced Marine Toxins and Venoms

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Background:

- Marine envenomations are common worldwide and can lead to severe morbidity[1-3]
- Effects of marine envenomations can range from mild to severe and can include paralysis, cardiac depression and neurological toxicity, and can be fatal[3]
- There is a rising prevalence of travel and ecotourism, thus leading to increased risk of exposure to marine stings and penetrating marine injuries
- We aim to synthesize existing evidence around diagnosis, treatment, and prevention of marine envenomations into a clinical resource

Methods:

- Four electronic databases were searched: PubMed (NCBI), Medline (OVID), EMBASE (OVID), LILLAC (VHL) and BioSIS (Web of Science) from database inception to August 2019 using combinations of the search terms 'marine' AND 'Intoxications' AND 'envenomations' AND 'syndrome'
- We included observational studies, case reports, case series, and cohort studies, as well as clinical trials and therapeutics tolerability and efficacy; and restricted to humans only
- Abstracts and full-text articles will be systematically double screened by two reviewers and subsequently by a tertiary arbitrator
- The GRADE approach will be employed to assess quality of studies reporting therapeutic interventions
- Evidence will be summarized using descriptive measures for each intervention type
- Data will be grouped and summarized for ease of clinician use by marine organism, syndrome, prevention, and therapeutic strategies, and according to geographic location and species
- Meta-analysis will be performed as appropriate with random effects model

Results: *collated from analysis of 136 abstracts selected for full text review up until October 31, 2019*

Figure 1: PRISMA flow diagram

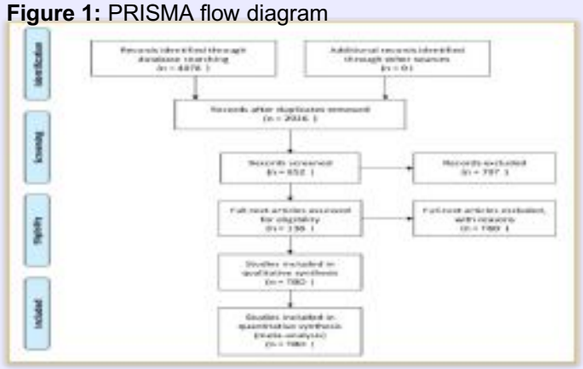


Figure 2: Workflow highlighting breakdown of abstracts by database

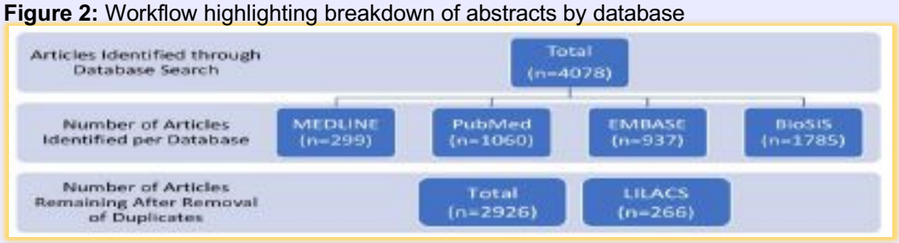


Figure 3:

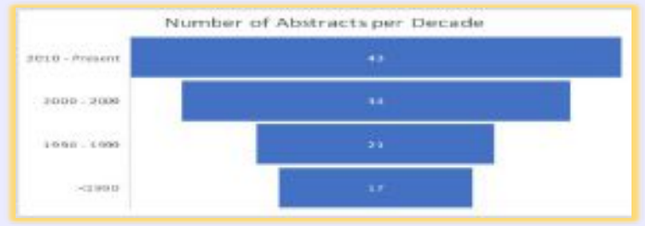
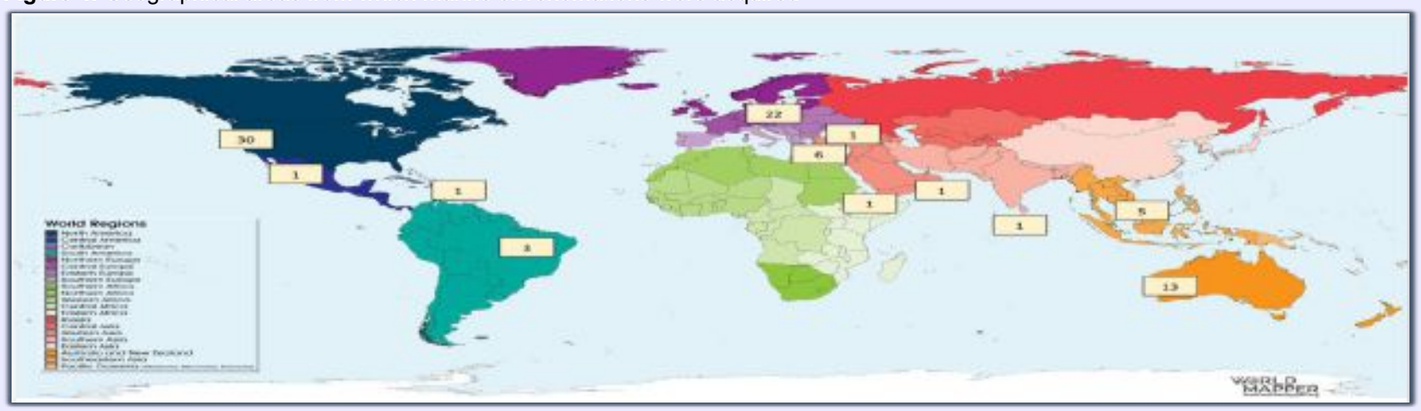


Table 1: Breakdown of type of marine envenomation

Etiology	Total N
Jellyfish	47
Scorpaenidae	26
(Lionfish)	11
(Stonefish)	10
(Scorpionfish)	5
Stingrays	21
Sea Snakes	15
Fish (other)	13
Sea Snails	7
Weeverfish	7
Sea Urchins	6
Octopus	6
Sea Anemones and Corals	5
Fish (other, cartilaginous i.e. ratfish or sharks, eagle ray)	3
Sponge	2

Figure 4: Geographical areas from which marine envenomations were acquired



Discussion and Conclusion:

- With increased globalization and the rising number of clinicians electing to train or work in areas where marine envenomations are common, it is important to synthesize the current evidence around clinical epidemiology, presentation, and management for marine envenomations
- Thus far in our search, jellyfish, scorpaenidae, and stingrays are the leading etiological agent for marine envenomations, and geographical areas of interest for the envenomations include North America, Australia, and Europe
- This synthesis will subsequently help to develop updated public health protocols to ensure timely and effective medical intervention for marine envenomations

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Low Sequence Heterogeneity of *Plasmodium falciparum* Isolates Imported to Ontario, Canada from West Africa over a 10-year Period with Increased Molecular Markers of Resistance to Proguanil

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Background

- Approximately 200 cases of malaria are imported to Ontario by returning travellers annually.
- Majority of cases are *Plasmodium falciparum (Pf)* from West Africa, including Ghana and Cameroon.
- Genetic strain typing is crucial to understand treatment failure outcomes which may indicate a new infection or recrudescence.

Objective

- Perform sequence analyses of *Pf* isolates imported from Ghana and Cameroon over a 10-year period to understand patterns of genetic heterogeneity and molecular drug resistance markers over time.

Results

- We identified 36 *Pf* isolates from Ghana (18 from 2006-2008 and 18 from 2014-2016); and 16 from Cameroon throughout 2006-2016.
- All were confirmed to be mono-*Pf* infections by real-time PCR.
- No molecular resistance to artemisinin derivatives was observed.

Figure 1. Phylogenetic Tree based on bootstrap method with 1000 replications of *Pf* isolates from Ghana 2006-2008 and 2014-2016 (1a) and Cameroon 2006-2016 (1b)

- There was some sequence heterogeneity among the isolates.
- No clustering was observed over the two time periods.

Fig. **1a**

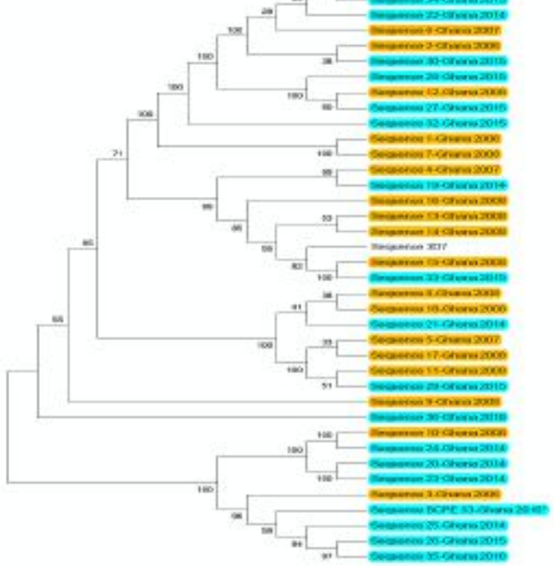
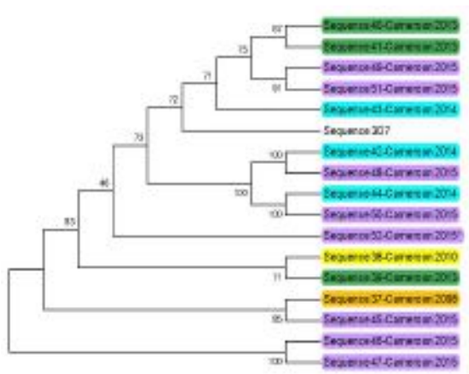


Fig. **1b**



Materials and Methods

- DNA was extracted from frozen bio-banked whole blood samples.
- Species ID was confirmed by multiplex real-time PCR.
- PCR and Sanger sequencing were performed on regions commonly used for strain typing: *merozoite surface protein (msp)* 1 and 2; *erythrocyte binding antigen (eba) 175*; and *glutamate-rich protein (glurp).*
- Sequence assembly, alignment and phylogenetic analyses were performed by MEGA 6.0.
- Molecular resistance markers: *cytochrome B (cytB), dihydrofolate reductase (dhfr), atpase6, kelch13*, and *chloroquine resistance transporter (pfcrt*) were analyzed.

Table 1. Molecular resistance marker analysis of *Pf* isolates from Ghana and Cameroon

- All isolates analyzed were wild type at *cytB* codon 268, *atpase6* codon 769 and *kelch13*
- *dhfr* triple codon mutants increased from 39% in 2006-2008 to 83% in 2014-2016 in Ghanaian isolates
- All isolates from Cameroon had *dhfr* triple codon mutants

Drug	Molecular Resistance Marker	Ghana Mutant (%) 2006-2008	Ghana Mutant (%) 2014-2016	Cameroon Mutant (%) 2006-2016	*p*-value*
Atovaquone	*cytB* Y268SCN	0/18 (0%)	0/18 (0%)	0/16 (0%)	1.0000
Proguanil	*dhfr* N51I, C59R, S108N triple codon mutant	7/18 (39%)	15/18 (83%)	16/16 (100%)	**0.0153**
Artemisinin derivatives	*atpase6* A623E	2/18 (11%)	1/18 (6%)	0/16 (0%)	1.0000
Artemisinin derivatives	*atpase6* S769N	0/18 (0%)	0/18 (0%)	0/15 (0%)#	1.0000
Artemisinin derivatives	*kelch13* codons >440	0/17 (0%)#	0/18 (0%)	0/16 (0%)	1.0000
Chloroquine	*pfcrt* K76T	8/18 (44%)	1/18 (6%)	9/15 (60%)#	**0.0178**

* Comparison of isolates imported from Ghana 2006-2008 vs. 2014-2016
\# Analysis of partial set of samples, some samples not amplifiable

Conclusion

- Low sequence heterogeneity suggests there was no major evolutionary genetic changes in isolates of *Pf* from Ghana and Cameroon.
- Molecular resistance to chloroquine was still prevalent in Cameroon and may due to counterfeit drugs or inappropriate use of chloroquine.
- *Pf* strains from Ghana had decreased molecular markers of resistance to chloroquine over time.
- Not enough data to suggest whether recycling chloroquine back into the treatment regime in West Africa is appropriate.
- *Pf* strains from Ghana had increased molecular markers of resistance to proguanil over time.
- All strains remain wild type to the partner drug atovaquone in Malarone®.
- With the increased molecular resistance to proguanil coupled with the reduced efficacy of atovaquone that may occur with malabsorption, patients should be informed to take Malarone® appropriately with a fatty meal.

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Clinical Medicine

Group 6

Validation of a Multiplex real-time PCR Gastrointestinal Parasite Panel

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Background:

- Microscopy, a conventional method for identification of gastrointestinal parasitic pathogens in fecal samples presents numerous challenges including high technical expertise and prolonged turnaround time
- Molecular methods provide higher throughput and potentially higher sensitivity and specificity
- We sought to validate a commercial multiplex parasitic real-time PCR panel detecting 6 protozoal pathogens in unpreserved fecal specimens for diagnostic parasitology: *Blastocystis hominis (Bh)*, *Cryptosporidium, Cyclospora, Dientamoeba fragilis (Df)*, *Entamoeba histolytica (Eh)* and *Giardia lamblia (Gl)*

Methods and Materials:

- We identified 478 specimens: 461 unpreserved, fresh stool specimens and 17 frozen samples.
- The fresh samples consisting of 80 microscopy positives with mono- or mixed infection, and 381 negatives; 17 banked frozen positive specimens; and 23 specimens for cross-reactivity check (Table-1)
- Cross reactivity panel consisted of 23 samples comprising *Entamoeba dispar,* various gastrointestinal helminths and human DNA (Table-1)
- DNA extraction and PCR were setup with the Hamilton Starlet automated platform, and Seegene's extraction and gastrointestinal parasite PCR kits
- Microscopy for O&P was the reference standard for all organisms with stool ELISA as an additional reference assay for *Entamoeba histolytica* to differentiate it from *Entamoeba dispar*.
- Limit of detection was calculated using unpreserved stool samples for qPCR and SAF stool samples for microscopy quantification. LOD was determined as parasites per gram stool detected at C_t of 43.
- Sensitivity, specificity, PPV and NPV were calculated using SPSS version 21.0 (IBM Corp., USA)
- Comparison of technologist hands-on time and total turnaround time was completed for microscopy and molecular assays for diagnostic analysis. 20 stool samples were chosen for timing comparison as it was the capacity of the microscopy staining machine.

Table 1: Description of Specimens in this Study

Sample Type	Organism	n	Fresh or Frozen Stool
Mono	*Blastocystis hominis (Bh)*	29	Fresh
Mono	*Cryptosporidium*	7	Fresh
Mono	*Cyclospora*	6	Fresh
Mono	*Dientamoeba fragilis (Df)*	16	Fresh
Mono	*Entamoeba histolytica (Eh)*	3	Fresh
Mono	*Giardia lamblia (Gl)*	4	Fresh
Mixed	*Bh+Df*	6	Fresh
Mixed	*Bh+Df+Gl*	1	Fresh
Mixed	*Bh+Gl*	5	Fresh
Mixed	*Cryptosporidium+Gl*	1	Fresh
Mixed	*Bh+Cryptosporidium*	2	Fresh
-	Negative	381	Fresh
-	**Total**	**461**	**Fresh**
Mono	*Entamoeba histolytica (Eh)*	17	Frozen
Cross Reactivity Panel	*Ascaris lumbricoides*	3	Fresh and Frozen
Cross Reactivity Panel	*Dicrocoelium dendriticum*	1	
Cross Reactivity Panel	*Diphyllobothrium latum*	2	
Cross Reactivity Panel	*Entamoeba dispar*	3	
Cross Reactivity Panel	*Enterobius vermicularis*	2	
Cross Reactivity Panel	Hookworm	3	
Cross Reactivity Panel	*Schistosoma mansoni*	2	
Cross Reactivity Panel	*Strongyloides stercoralis*	3	
Cross Reactivity Panel	*Taenia*	2	
Cross Reactivity Panel	*Trichuris trichiura*	2	
Cross Reactivity Panel	Human DNA	1	-

Results:

Table 2: Sensitivity, Specificity, PPV, NPV of Multiplex parasitic real-time PCR panel for fresh and frozen specimens combined

Protozoan Species	Microscopy Positives (N)	Microscopy Negatives (N)	Sensitivity % (95% CI)	Specificity % (95% CI)	PPV % (95% CI)	NPV % (95% CI)
Blastocystis hominis	43	418	93.0 (80.9 - 98.5)	98.3 (96.6 - 99.3)	85.1 (73.2 – 92.3)	99.3 (97.9 - 99.8)
Cryptosporidium	10	451	100 (69.2 - 100)	100 (99.2 - 100)	100 (72.2 - 100)	100 (99.2 - 100)
Cyclospora	6	455	100 (54.1 - 100)	100 (99.2 - 100)	100 (61.0 - 100)	100 (99.2- 100)
Dientamoeba fragilis	23	438	100 (85.2 – 100)	99.3 (98.0 - 99.9)	88.5 (71.3 – 96.0)	100 (99.1 - 100)
Entamoeba histolytica (fresh)	3	458	33.3 (0.84 – 90.6)	100 (99.2 - 100)	100 (99.0 - 100)	99.6 (99.0 – 99.8)
Entamoeba histolytica (fresh+frozen)	20	458	75.0 (50.6 – 90.4)	100 (99.0 – 100)	100 (74.7 - 100)	98.9 (97.3 – 99.6)
Giardia lamblia	11	450	100 (67.9 - 100)	98.9 (97.3 - 99.6)	68.8 (41.5 – 87.9)	100 (98.9 - 100)

PPV and NPV denote Positive Predictive Value and Negative Predictive Value, respectively

Table 3: Limit of Detection

Protozoan Species	Parasites per Gram Stool
Blastocystis hominis	8
Cryptosporidium	9
Cyclospora	38
Dientamoeba fragilis	697
Entamoeba histolytica	47
Giardia lamblia	2

Table 4: Timing of microscopy and Molecular Assays

Procedure	Microscopy Timing in Minutes	Molecular Timing in Minutes	Technologist Hands-On Time (Yes/No)
Iron-Haemoatoxylin Slide - Prep	8	-	Yes
Iron – Haemoatoxylin Slide – Staining	75	-	No
Iron-Haemoatoxylin Slide – Analytical	300	-	Yes
Auramine-Rhodamine Slide – Prep	8	-	Yes
Auramine-Rhodamine slide – Staining	20	-	No
Auramine-Rhodamine Slide – Analytical	32	-	Yes
Wet Prep Concentrate – Prep	74	-	Yes
Wet Prep Concentrate – Analytical	200	-	Yes
Sample Prep	-	40	Yes
Setup of Automated Liquid Handler	-	20	Yes
Automated DNA Extraction and PCR Setup	-	80	No
Real Time PCR Run	-	165	No
PCR Analysis	-	10	Yes
Total Technologist Hands-On Time	622 min = 10.5 h	70 min = 1.2h	
Total Turnaround Time	717 min = 12h	315 min = 5.3h	

Conclusions:

- The platform had high sensitivity for *Blastocystis hominis*, *Cryptosporidium, Cyclospora, Dientamoeba fragilis,* and *Giardia lamblia* , but suboptimal sensitivity for *Entamoeba histolytica* which can be attributed to low number of fresh samples available for *Entamoeba histolytica.*
- Low positive predictive value for *Blastocystis hominis, Dientamoeba fragilis,* and *Giardia lamblia* may reflect challenges to accurately identify these organisms microscopically.
- No cross-reactivity was observed with any of the DNA samples of helminthic parasite species.
- Negative predictive value was excellent for all targets.
- This enteric multiplex platform provides a useful diagnostic tool in complement to microscopy for *Blastocystis hominis, Cyclospora, Cryptosporidium*, *Dientamoeba fragilis*, and *Giardia lamblia*.
- Further recruitment of fresh samples is required to determine more accurate performance characteristics of this platform.

Acknowledgements:

- Molecular Research and Parasitology departments at Public Health Ontario (PHO)
- Funding was provided by PHO, Ontario Agency for Health Protection and Promotion

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Validation of a multiplex real-time PCR gastrointestinal helminth panel

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Introduction

•Microscopy, a conventional method for identification of gastrointestinal parasitic pathogens in fecal samples presents numerous challenges including high technical expertise and prolonged turnaround time requiring multiple staining procedures for definitive diagnosis.
•Molecular methods provide higher throughput and potentially higher sensitivity and specificity
•We sought to validate a commercial multiplex parasitic real-time PCR panel detecting 8 helminthic pathogens in unpreserved fecal specimens for diagnostic parasitology: *Ancylostoma* spp. *(An), Ascaris* spp. *(As), Enterobius vermicularis (Ev), Hymenolepis* spp. *(Hy), Necator americanus (Na), Strongyloides* spp. *(St), Taenia spp. (Ta)* and *Trichuris trichiura (Tt).*

Materials and Methods

• We identified 163 frozen specimens in our biobank: *An* (n=5), *As* (n=26), *Ev* (n=17), *Hy* (n=1), *Na* (n=4), *St* (n=37), *Ta* (n=17), *(Tt)* (n=11), mixed (1 *St* + *Tt*, 2 *St* + *Ta*), and 39 negatives.
• The cross reactivity panel included *Blastocystis hominis, Clonorchis sinensis, Cryptosporidium, Cyclospora, Dientamoeba fragilis, Entamoeba histolytica, Giardia lamblia, Schistosoma mansoni,* and human DNA.
• DNA extraction (with and without Buffer ASL pre-treatment for 10min) and PCR were setup with the Hamilton Starlet automated platform with Seegene extraction kit and gastrointestinal helminth PCR kit.

Results

Table 2a: Performance Characteristics of Helminth molecular assay without ASL lysis buffer pre-treatment

Helminth species	Microscopy Positives	Microscopy Negatives	Sensitivity % (95% CI)	Specificity % (95% CI)	PPV % (95% CI)	NPV % (95% CI)
Ancylostoma spp. (Hookworm)	5	154	80% (28-99%)	100% (98-100%)	100% (40-100%)	99% (96-100%)
Ascaris spp.	26	133	54% (33-73%)	100% (97-100%)	100% (77-100%)	92%(86-96%)
Enterobius vermicularis	17	142	76% (50-93%)	100% (97-100%)	100% (75-100%)	97% (93-99%)
Hymenolepis spp.	1	158	100% (3-100%)	100% (98-100%)	100% (3-100%)	100% (98-100%)
Necator americanus (Hookworm)	4	155	100% (40-100%)	99% (96-100%)	80% (28-99%)	100% (98-% (100%)
Strongyloides spp.	40	119	58% (41-73%)	99% (95-100%)	96% (79-100%)	87% (81-92%)
Taenia spp.	19	140	89% (67-99%)	100% (97-100%)	100% (80-100%)	99% (95-100%)
Trichuris trichiura	12	147	0% (--)	100% (--)	--	92% (--)

PPV – Positive Predictive Value
NPV – Negative Predictive Value

Results

Table 2b: Performance Characteristics of Helminth molecular assay with ASL lysis buffer pre-treatment

Helminth Species	Microscopy Positives	Microscopy Negatives	Sensitivity % (95% CI)	Specificity % (95% CI)	PPV % (95% CI)	NPV % (95% CI)
Ancylostoma spp. (Hookworm)	5	154	80% (28-99%)	100% (98-100%)	100% (40-100%)	99% (96-100%)
Ascaris spp.	26	133	65% (44-83%)	100% (97-100%)	100% (80-100%)	94% (88-97%)
Enterobius vermicularis	17	142	76% (50-93%)	100% (97-100%)	100% (75-100%)	97% (93-99%)
Hymenolepis spp.	1	158	100% (3-100%)	100% (98-100%)	100% (3-100%)	100% (98-100%)
Necator americanus (Hookworm)	4	155	100% (40-100%)	99% (96-100%)	80% (28-99%)	100% (98-100%)
Strongyloides spp.	40	119	58% (41-73%)	100% (97-100%)	100% (85-100%)	88% (81-93%)
Taenia spp.	19	140	89% (67-99%)	100% (97-100%)	100% (80-100%)	99% (95-100%)
Trichuris trichiura	12	147	0% (--)	100% (--)	--	92% (--)

PPV – Positive Predictive Value
NPV – Negative Predictive Value
95%CI by GraphPad Prism 5

Discussion

•High sensitivity for the detection of *Ancylostoma* spp., Hymenolepis spp., Necator americanus and *Taenia* spp. but suboptimal for other Helminth species by multiplex PCR.
•Both extraction methods had similar performance characteristics with the exception of an ASL pre-treatment enhancing the sensitivity of *Ascaris* spp.
•No cross-reactivity was observed to protozoa or helminths not included in the platform, such as trematodes.
•Further prospective recruitment of fresh positive samples is required to determine more accurate performance characteristics of this platform.

Acknowledgement: Grace Jeong from Seegene for technical advice.

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Lifestyle Interventions for Neuropathic Pain

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Introduction

- Neuropathic pain affects 1 in every 10 individuals over the age of 30
- Standard pharmacological interventions for neuropathic pain such as antidepressants, anticonvulsants, and opioids are associated with significant side effects and limited effectiveness
- Lifestyle interventions have recently emerged as accessible and cost-effective strategies to reduce the burden and severity of neuropathic pain, particularly in type 2 diabetes
- Strategies seeking to improve physiological wellness, including those that reduce inflammation and enhance immune responsiveness to neurotoxic factors are powerful tools that can influence underlying neuropathic etiologies
- This systematic review seeks to understand and synthesize the literature surrounding lifestyle interventions - such as diet, physical activity, and smoking cessation - for neuropathic pain.

Methods

- A comprehensive search strategy encompassing underlying neuropathic etiologies, lifestyle interventions, and stratifiers was conducted using 5 databases (Embase, Medline, Pubmed, Scopus, LILACS) from inception to April 2020
- Articles were screened independently by two reviewers and discrepancies were resolved by a tertiary arbitrator during title/abstract, and full-text screening
- The quality assessment tool GRADE (Grading of Recommendations, Assessment, Development and Evaluations) was implemented to assess the quality and bias of evidence
- Inclusion Criteria: Randomized controlled trials, clinical trials, cohort studies, observational studies, case-control studies, case series & reports, non-English publications
- Exclusion Criteria: Systematic reviews, reviews, conference abstracts, editorials, animal studies, in vitro studies, trial descriptions

Results

Study	Author, Year	N	Mean Age (SD)	Range	Sex N (F:M)	Neuropathy	Etiology	Lifestyle	Outcome
Clinical Trial	[1]Henke, 2014	29	-	29-82	-	Peripheral Neuropathy	Cancer	Exercise Regimen	A statistically significant association was not detected on DN4.
	[2]Kneis, 2019	37	65		26:11	Peripheral Neuropathy	Cancer	Exercise Regimen	Reduction of neurotoxicity subscale (p=0.015), and vibration test (p<0.01)
	[3]Kleckner, 2018	355	56 (11)		330:25	Peripheral Neuropathy	Cancer	Exercise Regimen	Reduction of pain severity by numbness & tingling (p=0.061) and QST (p=0.045)
	[4]Malhotra, 2002	40	-		-	Neuropathy (in general)	Diabetes	Exercise Regimen	Increase in conduction velocity
	[5]Fisher, 2007	5	68.4 (6.4)		0:5	Neuropathy (in general)	Diabetes	Exercise Regimen	Improvement in motor CV & amplitdue (p<0.001), & sensory CV (p<0.03)
	[6]Kluding, 2010	6	60.2 (4.7)		4:2	Peripheral Neuropathy	Diabetes	Diet + Exercise Councelling	No statistically significant change on MNSI found
	[7]Ng Wing Tin, 2019	8	65		4:4	Polyneuropathy	Diabetes	Diet + Exercise Councelling	Improvement of Neuropathy total symptom score via QST (p=0.0008)
	[8]Kluding, 2012	17	58.4 (5.98)		9:8	Peripheral Neuropathy	Diabetes	Exercise Regimen	Improvements in pain (p<0.05), MNSI symptom score (p<0.01), and IENF (p<0.008)
	[9]Gholami, 2018	24	42.5		0:24	Peripheral Neuropathy	Diabetes	Exercise Regimen	Improvement of pain on MDNS (p<0.05) and sural sensory nerve CV (p=0.007)
	[10]Bunner, 2015	34	57.5		19:15	Neuropathy (in general)	Diabetes	Low-Fat Plant-Based Diet	Improvement of pain on MPQ (p=0.04), NTSS (P=0.70), VAS (0.39), MNSI-Q (P=0.03)
	[11]Ahn, 2012	39	64.4		19:20	Neuropathy (in general)	Diabetes	Exercise Regimen	Improvement in TSS (p=0.042)
	[12]Smith, 2006	40	60 (8.4)		20:20	Neuropathy (in general)	Diabetes	Diet + Exercise Councelling	Improvement in VAS (p<0.4), GPS (p<0.1), peroneal motor CV (p<0.004), and proximal IENFD & sural sensory amplitude (p< 0.03)
	[13]Ghavami, 2018	74	48.3		56:18	Peripheral Neuropathy	Diabetes	Diet + Exercise Councelling	Improvement of pain on mTCNS (p<0.001)
	[14]Balducci, 2006	78	52.1		30:39	Neuropathy (in general)	Diabetes	Exercise Regimen	Improvement of motor & sural nerve CV (p<0.001)
	[15]Dixit, 2014	87	56.9		34:53	Peripheral Neuropathy	Diabetes	Exercise Regimen	Improvement in pain (p=0.01), sensory symptoms (p=0.01), & overall score (p=0.001) on MDNS
	[16]Look AHEAD Research Group, 2017	5145	-		-	Peripheral Neuropathy	Diabetes	Exercise Regimen	Improvement oF pain on MNSI (p<0.001)
	[17]Toth, 2014	54	55.1		32:22	Peripheral Neuropathy	Diabetes	Exercise Regimen	No statistically significant change on MNSI found
	[18]Maharaj, 2018	136	36.8 (8.23)		79:57	Neuropathic Pain	HIV/Aids	Exercise Regimen	Improvement in pain (p<0.05) on NPRS
	[19]Arnold, 2017	47	60		17:30	Neuropathy (in general)	Kidney Disease	Potassium Reduced Diet	Improvement in neuropathy severity (p<0.01) on NPS
	[20]Albayrak, 2017	39	64		35:4	Neuropathy (in general)	Trauma / Surgery	Exercise Regimen	Improvement in pain on DN4 (p<0.05)
	[21]Hadjivassiliou, 2006	35	69.1		-	Neuropathy (in general)	Not Specified	Gluten Free Diet	Improvement in sural SNAP amplitude (p<0.03), subjectively neuropathy severity improved (p<0.0006)
Cross Sectional	[22]Ammendola, 2001	76	42.2	21-84	21:55	Neuropathy (in general)	Allcohol	Dietary Patterns	TLDE associated with greater neuropathy (p<0.05) & lower NCV (p<0.01)
	[23]Monforte, 1995	107	42		18:89	Autonomic & Peripheral Neuropathy	Alcohol	ARS	TLDE associated with greater neuropathy (p<0.01)
	[24]Tao, 2008	1062	-		-	Peripheral Neuropathy	Diabetes	Dietary Polyunsaturated FA	Intake of PUFA was lower in partiicpants with neuropathy
	[25]Gong, 2011	400	-		-	Neuropathy (in general)	Diabetes	Exercise & Dietary Patterns	No statistically significant association reported
	[26]Loprinzi, 2014	339	61.8		161:178	Peripheral Neuropathy	Diabetes	Physical Activity Level	Normal HbA1c level & greater than average physical activity correlated to lower odds of neuropathy
	[27]Yoo ,2015	14	57 (5.11)		9:5	Neuropathic Pain	Diabetes	Exercise Level	No statistically significant association reported
	[28]Nicholas, 2010	208	44.6 (9.8)		87:121	Neuropathy (in general)	HIV/Aids	Exercise Level	Exercise is subjectively effective for treating neuropathy
	[29]Nicholas, 2007	445	43.58 (9)		129:316	Neuropathy (in general)	HIV/Aids	Physical Activity Level	Exercise is subjectively effective for treating neuropathy
	[30]Mols, 2015	1643	68.7		708:935	Peripheral Neuropathy	Cancer	Physical Activity Level	Worse neuropathy scores on EORTC OLO-C30 correlated to low levels of physical activity.
	[31]Zis, 2018	60	69.9 (10.1)		14:46	Peripheral Neuropathy	Gluten	Gluten Free Diet	Diet was associated with lower odds of neuropathic pain (p=0.006)
	[32]Norrbrink, 2012	13	47		5:8	Neuropathic Pain	Trauma / Surgery	Exercise Level	Subjective improvement of pain severity with exercise
	[33]Ojeda, 2018	326	47.4		208:118	Neuropathy (in general)	Not Specified	Sleep Quality	No statistically significant association reported
Cohort	[34]Fernandes, 2016	25	47 (12)	29-81	13:12	Peripheral Neuropathy	Cancer	Exercise Level	Improvement on mTNS (p=0.00001)
	[35]McCrary, 2019	29	61.6		21:8	Peripheral Neuropathy	Cancer	Exercise Level	Lower TNSc (p=0.001) & CIPN-20 (p<0.001) scores with more exercise
	[36]Galantino, 2019	10	64.4		9:1	Peripheral Neuropathy	Cancer	Exercise Level	Reduced pain severity on BPI (p=0.041) and CIPN (p=0.003)
	[37]Teixeira, 2010	20	74.6		15:5	Peripheral Neuropathy	Diabetes	Dietary Patterns	No statistically significant association reported
	[38]Nolan, 2016	465	63		182:283	Peripheral Neuropathy	Diabetes	Physical Activity Level	Neuropathy was correlated with lower physical activity (p=0.04) on IPAQ
	[39]Hawley, 1982	63	53.3		0:63	Neuropathy (in general)	Alcohol	Dietary Patterns	Reduced sensory symptoms on QST with "normal" diet
	[40]Lange-Maia, 2016	328	73.7 (5.9)		0:328	Neuropathy (in general)	Not Specified	Physical Activity Level	Higher levels of physical activity & fewer minutes of sedentary behaviors correlated with better CMAP (p<0.05)
Case Report / Case Series	[41]Polat, 2018	1	53	33-88	1:0	Neuropathy (in general)	Genetic Syndrome	CPAP for Sleep	Patient's neuropathy symptoms improved with CPAP initiation
	[42]Hungerbuhler, 1985	1	33		1:0	Neuropathy (in general)	Genetic Syndrome	Low Phytanate Diet	Improvement in strength, reflexes, and NCV
	[43]Sahgal, 1975	1	33		0:1	Neuropathy (in general)	Genetic Syndrome	Low Phytol Diet	Improvement in strength, and NCV
	[44]Zagarella, 2016	12	57		9:3	Sensory neuropathy	Trauma / Surgery	Exercise Level	All patients had an improvement in VAS & NPS
Case Control	[45]Ishibashi, 2019	158	50.4	10-69	65:93	Neuropathy (in general)	Diabetes	HbA1c Controlled Diet	Improvements in NDS, median nerve CV, and VPT.
	[46]Clements, 1979	20	33.6		6:14	Neuropathy (in general)	Diabetes	Myo-inositol Diet	Improvement in median & sural sensory CV (p<0.001)
	[47]Gay, 1995	99	43		13:21	Neuropathy (in general)	Diabetes	Dietary Patterns	Neuropathy correlated to less varied diet, and low intake of nutrients.

Table 1. Preliminary Data Extraction of Included Studies

Abbreviations: Avoiding Risky Substances (ARS), Fatty Acids (FA), Continuous Positive Airway Pressure (CPAP), Douleur Neuropathique en 4 (DN4), Quantitative Sensory Testing (QST), Conduction Velocity (CV), Michigan Neuropathy Screening Instrument Questionnaire (MNSI-Q), Intra Epidermal Nerve Fibers (IENF), Michigan Diabetic Neuropathy Score (MDNS), McGill Pain Questionnaire (MPQ), Neuropathy Total Symptom Score (NTSS), Visual Analog Scale (VAS), modified Toronto Clinical Neuropathy Score (mTCNS), Neuropathic Pain Rating Scale (NPRS), Neuropathy Pain Scale (NPS), Sensory Nerve Action Potential (SNAP), Nerve Conduction Velocity (NCV), Total Lifetime Dose Ethanol (TLDE), Polyunsaturated Fatty Acids (PUFA), European Organization for the Research and Treatment of Cancer Quality of Life Questionnaire (EORTC OLO-C30), modified Total Neuropathy Score (mTNS), Total Neuropathy Score clinical version (TNSc), Chemotherapy Induced Peripheral Neuropathy (CIPN-20),Brief Pain Inventory (BPI), International Physical Activity Questionnaire (IPAQ), Compound Muscle Action Potential (CMAP), Vibration Perception Threshold (VPT)

Results

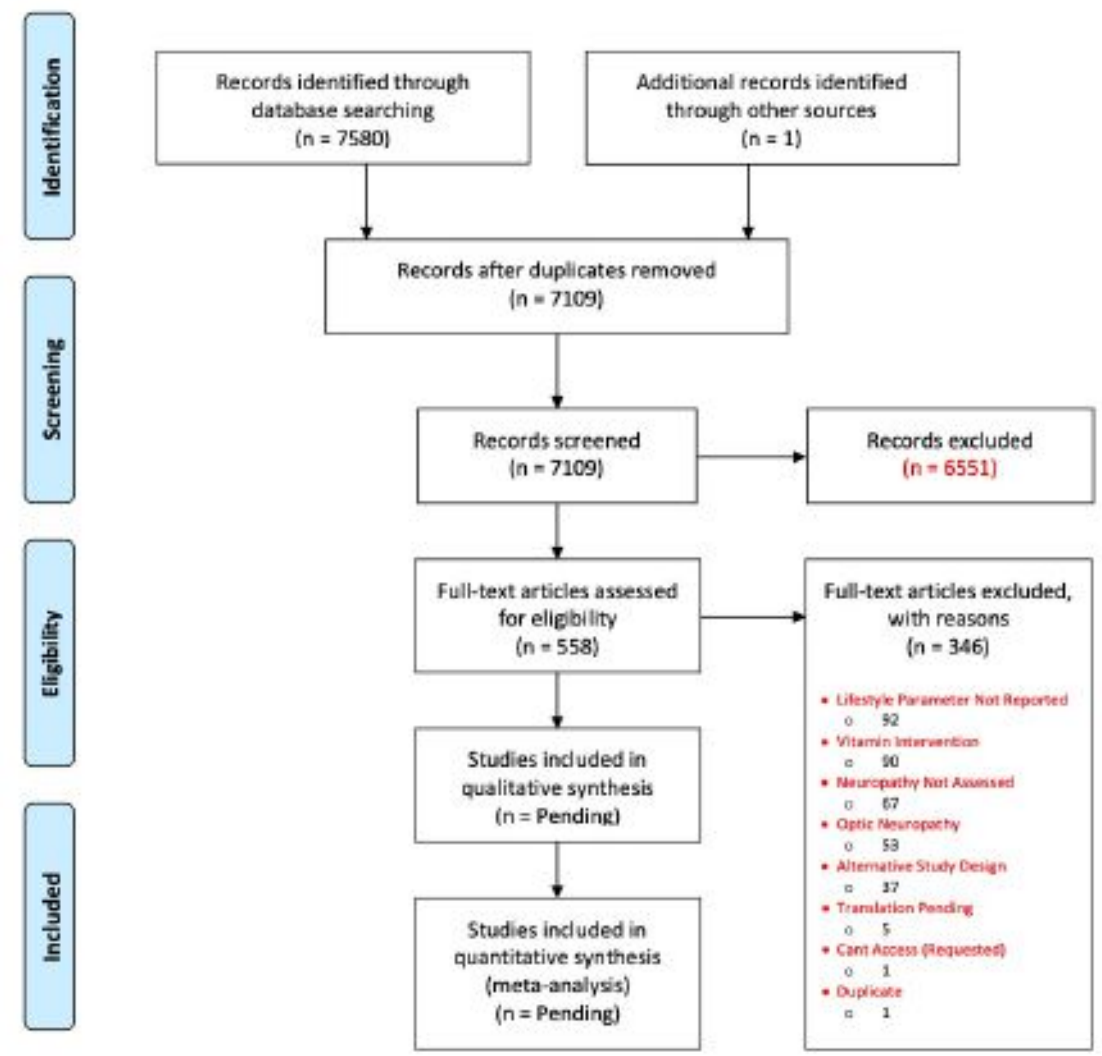

Figure 1. PRISMA Flowchart

Discussion

- Following full-text screening 211 articles remained for inclusion
- Efficacy, safety, tolerability, and harms stratified by age, sex, BMI, and socioeconomic status will be synthesized
- Preliminary findings suggest that the most common lifestyle parameters assessed are diet & exercise in diabetes & cancer cohorts
  - ~90% of studies are reporting a neuropathic improvement or positive associations with lifestyle
- This knowledge synthesis will inform the development of a randomized control trial exploring a low-risk, low-cost, low-tech lifestyle intervention for chronic neuropathic pain in leprosy, thereby improving function and reducing overall morbidity, and mortality.

References

Spectrum of Bacterial Pathogens in Inflammatory and Non-Inflammatory Cutaneous Ulcers of American Tegumentary Leishmaniasis (ATL)

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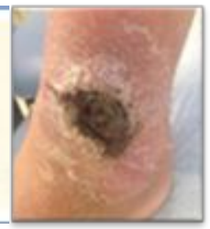
Background

- Leishmaniasis is a Neglected Tropical Disease (NTD) with 2 million cases globally and estimated 350 million people at risk.[1]
- Cutaneous Leishmaniasis (CL) is the most common form characterized by significant skin ulcerations proximate to the site of *Leishmania* spp. injection by the female sandfly vector.[1]
- Patients with CL can present with ulcers that exhibit an inflammatory phenotype characterized by pain, erythema, and purulent exudate and as a result, are empirically treated with antibiotics.
- Although the inflammatory CL phenotype suggests a secondary bacterial infection, bacterial contribution to the pathogenesis of the inflammatory CL ulcer remains to be elucidated.
- Antimicrobial stewardship and evidence-based management guidelines for inflammatory CL ulcers would benefit from knowledge of the microbial differences between inflammatory and non-inflammatory CL ulcers.

Panel 1 & 2: Transmission of *Leishmania* spp. occurs through female sandfly bites.



Panel 3: Inflammatory CL ulcer phenotype

Objective

The objective of this study was to document the spectrum of bacteria present in inflammatory and non-inflammatory ulcers in order to understand the bacterial contribution to CL phenotypes.

Methods

Filter Paper Lesion Impressions (FPLIs) were used to extract tissue samples from CL ulcers

→ Bacterial DNA Extracted from n=39 patients

→ **Real-time PCR:**
- 18S *Leishmania*
- β2MG gene (human)

→ **End-point PCR & Real-Time PCR:**
1. *C. freundii*
2. *S. pyogenes*
3. *P. aeruginosa*
4. *S. aureus*
5. *E. cloacae*
6. *E. coli*
7. *K. pneumoniae*
8. *Enterococcus* spp.
9. 16S rDNA

→ A subset of n=6 FPLI were analyzed using Whole Genome Sequencing (WGS)

→ Sanger Sequencing for confirmation to genus and species level

RECRUITMENT :
- FPLIs submitted to the Ontario Laboratory (PHOL) and the Instituto de Medicina Tropical "Alexander von Humboldt" in Lima, Peru were collected between 2012 and 2018 and stored at -20°C.

STATISTICAL ANALYSIS:
- Comparative statistics on categorical variables were analyzed using Fisher's exact test, chi-square and chi-square test for trend; while continuous variables by Mann-Whitney U-test.
- Statistical analyses were calculated via GraphPad Prism Version 6.01 (La Jolla, California, USA).

Results

- In total, **n=39** CL samples were analyzed, of which **n=19 (49%)** met the criteria for exhibiting an inflammatory phenotype, while **n=20 (51%)** were classified as non inflammatory.
- Patients with the inflammatory phenotype were, on average, **over a decade older (42 vs. 27 years, *p=0.01*)** than patients with the non-inflammatory phenotype.
- CL ulcer phenotype **did not differ** across sex (*p=0.31*), causative *Leishmania* species (all *p>0.05*), or bacterial pathogens detected all *p>0.05*).

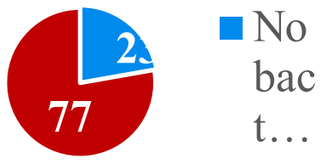


FIGURE 1: Overall prevalence of bacteria detected in all CL ulcer samples (n=39).

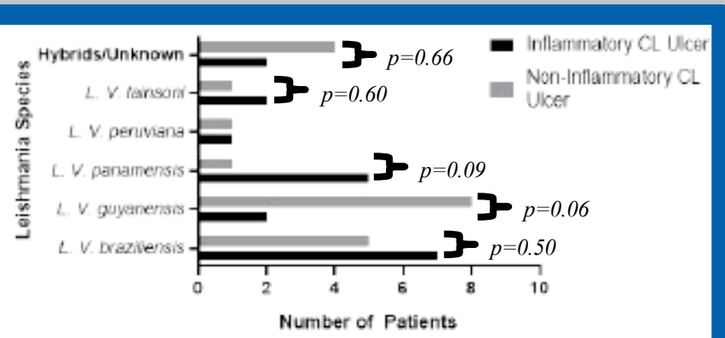


FIGURE 2: Distribution of causative *Leishmania spp.* across the inflammatory and non-inflammatory phenotypes of CL.

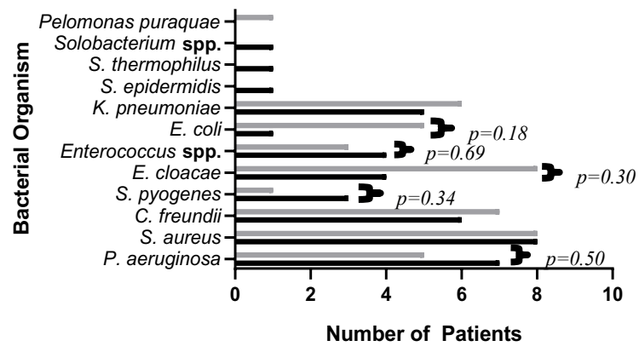


FIGURE 3: Distribution of bacterial organisms amongst patients with the inflammatory and non-inflammatory phenotypes of CL.

Results (Continued)

Sample	*S. aureus*	*C. freundii*	*S. pyogenes*	*Enterococcus spp.*	*Enterobacter spp./ Klebsiella spp.*	*P. aeruginosa*	*E. coli*
1	WGS/PCR	-	PCR	PCR	PCR	WGS	PCR
2	WGS/PCR	-	PCR	WGS	-	-	WGS
3	WGS/PCR	-	WGS	WGS	WGS/PCR	WGS/PCR	WGS/PCR
4	WGS/PCR	-	WGS	PCR	-	WGS	-
5*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*
6*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*

Table 1: WGS Analysis (n=6) – Comparison of WGS vs. Conventional PCR Methods. **Samples did not have sufficient DNA sequence data for analyses.*
- Of samples with sufficient DNA (n=4), all had *Brevundimonas nasdae* (gram-negative, opportunistic pathogen)

Discussion & Conclusions

- Prevalence of bacterial species **did not differ by CL phenotype**, as detectable pathogens and non-pathogens were equally distributed between inflammatory and non-inflammatory CL ulcers.
- It is unclear whether current empiric antibiotic therapy is necessary given the small sample size and a lack of difference observed between bacterial pathogens in inflammatory and non-inflammatory ulcers from our patient population
- Sandfly microbiota overlap with common opportunistic skin flora including *P. aeruginosa, S. aureus* and *C. freundii* could result in co-infections. [2-3]
- Other potential contributions: LRV-1, more infectious *Leishmania* species (*Viannia* subgenus) should be evaluated.
- **Our findings do not support the current practice of empiric antibiotic therapy for those with the inflammatory phenotype of CL ulcers.**
- Further examination using WGS are warranted to better understand other non-bacterial contributions to CL phenotype.

Reference

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3. Sant'Anna, M.R.V., Darby, A.C., Brazil, R.P., Montoya-Lerma, J., Dillon, V.M., Bates, P.A., Dillon, R.D. Investigation of the Bacterial Communities Associated with Females of Lutzomyia Sand Fly Species from South America. PLoS ONE 2012: 7(8):2531.

Molecular Surveillance for Drug Resistant *Plasmodium falciparum* Imported to Ontario

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Background

- *Plasmodium falciparum* can lead to rapid and fatal malaria in humans
- Single Nucleotide Polymorphisms (SNPs) at several loci have been correlated to *P. falciparum* treatment failure, delayed parasite clearance, and/or *in-vitro* drug resistance
- Recently artemisinin resistance has emerged in Southeast Asia and threatens to spread towards the African subcontinent

Objective: We wanted to study the prevalence of molecular resistance SNPs in *P. falciparum* infections imported by returning travelers and migrants to Ontario with cases from three time periods: July 2008-June 2009 (75 cases), July 2013-June 2014 (79 cases) and July 2017-June 2018 (89 cases)

Methods

- *P. falciparum* monoinfection in microscopy-confirmed specimens stored in our malaria biobank was verified by real time PCR [1]
- DNA was extracted from 200uL of whole blood (biobanked at -80) using the Kingfisher DNA blood kit and eluted with 150uL buffer
- Pyrosequencing to detect SNPs associated with drug resistance and/or treatment failure was performed on the following gene targets: *atpase6*, chloroquine resistance transporter (*Pfcrt*), cytochrome b (*cytb*), dihydrofolate reductase-thymidylate synthase (*dhfr*), dihydropteroate synthetase (*dhps*), and multidrug resistance protein (*mdr1*). The use of pyrosequencing allowed the analysis of the frequency of Wild Type and Mutant genotypes in each sample [2]
- Sanger sequencing was used to detect 20 SNPs associated with artemisinin resistance in the Kelch 13 *(k13)* gene [3]

Table 1: Location of single nucleotide polymorphisms (SNPs) associated with drug resistance and/or treatment failure.

Gene	Codon	Wild Type	Mutant	Malaria Drug Associated	Mutant Sensitive/Resistant
Pf ATPase 6	623 + 769	Alanine [A]; Serine [S]	Glutamic acid [E]; Asparagine [N]	Artemether	Resistant
Pf ATPase 6	769	Serine [S]	Asparagine [N]	Artemether	Resistant
Plasmodium falciparum chloroquine resistance transporter (Pfcrt)	72	Cysteine [C]	Serine [S]	Chloroquine	Resistant
	74	Methionine [M]	Isoleucine [I]	Chloroquine	Resistant
	75	Asparagine [N]	Glutamic acid [E]	Chloroquine	Resistant
	76	Lysine [K]	Threonine [T]	Chloroquine	Resistant
Plasmodium falciparum Cytochrome b (Cyt b)	268	Tyrosine [Y]	Cysteine [C]	Atovaquone	Resistant
			Serine [S]	Atovaquone	Resistant
			Asparagine [N]	Atovaquone	Resistant
Plasmodium falciparum bifunctional dihydrofolate reductase-thymidylate synthase (DHFR)	16 + 108	Alanine [A]; Serine [S]	Valine [V]; Threonine [T]	Cycloguanil	Resistant
	50 + 51 + 108 + 164	Cysteine [C]; Asparagine [N]; Serine [S]; Isoleucine [I]	Arginine [R]; Isoleucine [I]; Asparagine [N]; Leucine [L]	Pyrimethamine and cycloguanil	Resistant
	51 + 59 + 108 + 164	Asparagine [N]; Cysteine [C]; Serine [S]; Isoleucine [I]	Isoleucine [I]; Arginine [R]; Asparagine [N]; Leucine [L]	Pyrimethamine and cycloguanil	Resistant
	51 + 59 + 108	Asparagine [N]; Cysteine [C]; Serine [S]	Isoleucine [I]; Arginine [R]; Asparagine [N]	Proguanil	Resistant
Plasmodium falciparum dihydropteroate synthetase (Dhps)	436	Serine [S]	Alanine or Phenylalanine [A, F]	Sulfadoxine	Resistant
	437	Alanine [A]	Glycine [G]	Sulfadoxine	Resistant
	540	Lysine [K]	Glutamic acid [E]	Sulfadoxine	Resistant
	581	Alanine [A]	Glycine [G]	Sulfadoxine	Resistant
	613	Alanine [A]	Threonine or Serine [T, S]	Sulfadoxine	Resistant
Plasmodium falciparum multidrug resistance protein (Pf mdr1)	86	Asparagine [N]	Tyrosine [Y]	Mefloquine, Lumenfantrine, Artemether	Sensitive
	86	Asparagine [N]	Tyrosine [Y]	Chloroquine	Resistant
	184	Tyrosine [Y]	Phenylalanine [F]		
	1034	Serine [S]	Cysteine [C]	Artesunate	Resistant
	1042	Asparagine [N]	Aspartic acid [D]	Artesunate	Resistant
	1042	Asparagine [N]	Aspartic acid [D]	Mefloquine	Sensitive
	1246	Aspartic acid [D]	Tyrosine [Y]		
Pf mdr1 copy number	Increase			Mefloquine; Artesunate; Halofantrine	Resistant
				Mefloquine, Mefloquine+Artesunate	Resistant
Kelch 13 (k13)	449	Glycine [G]	Alanine [A]	Artemisinin	Resistant
	458	Asparagine [N]	Tyrosine [Y]	Artemisinin	Resistant
	474	Tyrosine [Y]	Isoleucine [I]	Artemisinin	Resistant
	476	Methionine [M]	Isoleucine [I]	Artemisinin	Resistant
	481	Alanine [A]	Valine [V]	Artemisinin	Resistant
	493	Tyrosine [Y]	Histidine [H]	Artemisinin	Resistant
	508	Tyrosine [Y]	Asparagine [N]	Artemisinin	Resistant
	527	Proline [P]	Threonine [T]	Artemisinin	Resistant
	533	Glycine [G]	Serine [S]	Artemisinin	Resistant
	537	Aspargine [N]	Isoleucine [I]	Artemisinin	Resistant
	539	Arginine [R]	Threonine [T]	Artemisinin	Resistant
	543	Isoleucine [I]	Threonine [T]	Artemisinin	Resistant
	553	Proline [P]	Leucine [L]	Artemisinin	Resistant
	561	Arginine [R]	Histidine [H]	Artemisinin	Resistant
	568	Valine [V]	Glycine [G]	Artemisinin	Resistant
	574	Proline [P]	Leucine [L]	Artemisinin	Resistant
	580	Cysteine [C]	Tyrosine [Y]	Artemisinin	Resistant
	584	Aspartic acid [D]	Valine [V]	Artemisinin	Resistant
	612	Glutamic acid [E]	Aspartic acid [D]	Artemisinin	Resistant
	623	Serine [S]	Cysteine [C]	Artemisinin	Resistant

Results

Table 2: Patient characteristics by time period of *P. falciparum* infection

Characteristic	Mean SD* Total (N=243)	2008-2009 (N=75)	2013-2014 (N=79)	2017-2018 (N=89)	p-value
Age, years†	39.2 (18.3)	40.9 (17.1)	38.0 (16.6)	38.9 (20.6)	0.61
Female sex, No (%)‡	66 (28.1)	18 (25.4)	19 (25.0)	29 (33.0)	0.47
Parasitemia, percent, (median, range)§	0.3 (0.01-24.0)	0.3 (0.01-17.8)	0.3 (0.01-12.0)	0.7 (0.01-24.0)	0.1
Region of Acquisition, No (%)¶					
West Africa	81 (33.3)	20 (26.7)	17 (21.5)	38 (42.7)	
East Africa	18 (7.4)	4 (5.3)	8 (10.1)	6 (6.7)	
Africa-other	3 (1.2)	1 (1.3)	1 (1.3)	1 (1.1)	
Africa, not otherwise specified	30 (12.3)	11 (14.7)	12 (15.2)	7 (7.9)	
Caribbean, Dominican Republic	1 (0.4)	1 (1.3)	0 (0)	0 (0)	
Southeast Asia	5 (2.1)	0 (0)	4 (5.1)	1 (1.1)	
South America, Guyana	1 (0.4)	0 (0)	0 (0)	1 (1.1)	
Unknown	104 (42.8)	33 (44)	36 (45.6)	35 (39.3)	0.14

* Except where otherwise noted
† Age was missing for 4 patients in 2008-2009, 1 patient in 2013-2014, and 1 patient in 2017-2018
‡ Sex was missing for 4 patients in 2008-2009, 3 patients in 2013-2014, and 1 patient in 2017-2018
§ Parasitemia was missing for 2 patients in 2008-2009
¶West Africa included Congo, Burkina Faso, Cameroon, Ghana, Guinea, Ivory Coast, Liberia, Nigeria, Togo, Senegal, and Sierra Leone; East Africa included Burundi, Ethiopia, Kenya, Rwanda, Sudan, South Sudan, Tanzania and Uganda; Africa-other included Zambia, Libya, and South Africa; Africa not otherwise specified included travel to multiple regions in Africa, or specific destination within Africa not reported; Caribbean included Dominican Republic; Southeast Asia included Burma and Philippines

Table 3: Mutant genotype frequency across over the three time periods

Gene	2008-2009	2013-2014	2017-2018	p-Value
ATPase A623E	1.30%	1.30%	0.00%	0.55
ATPase S769N	0.00%	0.00%	0.00%	1
CytB Y268SCN	0.00%	0.00%	0.00%	1
DHFR A16V	0.00%	0.00%	0.00%	1
DHFR S108N	89.30%	97.30%	100%	**0.001**
DHFR I164L	1.40%	0.00%	0.00%	0.32
DHFR C50R	0.00%	0.00%	1.30%	**0.01**
DHFR N51I	88.00%	92.30%	92.70%	0.55
DHFR C59R	90.70%	94.90%	93.30%	0.58
DHPS S436F	2.80%	5.00%	0.00%	0.411
DHPS S436FA	36.70%	43.40%	51.70%	0.167
DHPS A437G	88.00%	86.10%	92.00%	0.473
DHPS K540E	13.30%	20.50%	18.00%	0.5
DHPS A581G	9.50%	8.00%	12.50%	0.637
DHPS A613TS	12.20%	18.00%	28.70%	0.029
MDR1 N86Y	42.70%	14.30%	7.50%	**<0.001**
MDR1 Y184F	49.30%	60.30%	53.00%	0.4
MDR1 S1034T	0.00%	0.00%	1.20%	1
MDR1 S1034R	0.00%	0.00%	0.00%	1
MDR1 N1042D	0.00%	0.00%	0.00%	1
MDR1 D1246Y	17.60%	3.80%	3.50%	**0.003**
CRT K76T	56.80%	38.40%	32.80%	0.013
CRT N75E	52.00%	37.50%	26.80%	0.014
CRT M74I	52.00%	37.50%	26.80%	0.014
CRT C72S	1.40%	1.30%	0%	1

Table 4: Mutant allele frequency among wildtype genotypes over the three time periods

Gene	MT allele frequency among WT Median (range) % 2008-2009	2013-2014	2017-2018	p-value
ATPase A623E	0 (0-13.1)	0 (0-28.6)	3.9 (0-18.71)	**<0.001**
ATPase S769N	4.9 (1.6-10.9)	4.4 (0-8.2)	6.0 (1.4-27.4)	**<0.006**
CytB Y268N	0 (0-2.4)	0 (0-8.4)	3 (0-19.0)	**<0.001**
CytB Y268S	1.7 (0-5.0)	1.1 (0.7-2.6)	0.9 (0.37-8.5)	**0.008**
CytB Y268C	0.7 (0-1.5)	0.6 (0-18.8)	0.6 (0-3.7)	**<0.001**
DHFR A16V	0.7 (0.3-4.7)	0.8 (0-19.4)	0.7 (0.5-2.4)	**0.002**
DHFR S108N	6.5 (0-45.5)	21.2 (15.0-27.5)	NA	0.3
DHFR I164L	0.58 (0-96.8)	0 (0-11.1)	1.2 (0-43.5)	0.006
DHFR C50R	0.8 (0-3.4)	0.9 (0-5.4)	0.8 (0-3.2)	**0.008**
DHFR N51I	5.4 (2.1-48.1)	18.7 (0-39.8)	3.0 (0-7.4)	0.1
DHFR C59R	29.8 (16.5-50.0)	20.7 (0.8-25.6)	26.2 (24.2-45.8)	**0.061**
DHPS S436A	1.1 (0-41.5)	1.7 (0.3-39.1)	1.3 (0.5-37.0)	0.483
DHPS S436F	0.4 (0-41.5)	0.4 (0-2.9)	0 (0-2.5)	0.34
DHPS K540E	0.5 (0.2-13.6)	2.4 (0.3-16)	0.5 (0.2-8.9)	**<0.001**
DHPS A581G	0.1 (0-6.3)	2.0 (0-45.8)	0.7 (0-43.5)	**<0.001**
DHPS A613T	2.0 (0-32.0)	2.2 (0-35.8)	0.8 (0-39.8)	0.833
DHPS A613S	3.9 (0.5-7.3)	5.3 (1.3-10.7)	3.3 (0.9-6.3)	**<0.001**
DHPS A437G	0.4 (0-36.8)	1.6 (0-49.0)	0 (0-46.4)	0.984
MDR1 N86Y	2.0 (0.8-39.7)	3.0 (0.6-45.6)	2.5 (0.9-42.2)	0.015
MDR1 Y184F	1.3 (0-36.4)	1.7 (0-45.9)	1.7 (0-44.7)	0.61
MDR1 S1034T	0.9 (0-27.9)	0.8 (0.4-2.1)	1.4 (0.8-46.8)	**<0.001**
MDR1 S1034R	0.5 (0-2.2)	0.7 (0-3.5)	1.1 (0-4.7)	**<0.001**
MDR1 N1042D	4.3 (0-31.7)	5.4 (3.0-9.6)	6.5 (3.3-15.7)	**<0.001**
MDR1 D1246Y	7.4 (4.4-15.9)	8.2 (2.5-34.9)	16.3 (5.0-27.6)	**<0.001**
CRT K76T	15.9 (1.9-47.3)	13.5 (1.4-44.9)	5.8 (1.9-32.8)	**<0.001**
CRT C72S (TGT-AGT)	29.6 (9.2-48.8)	28.7 (8.1-48.8)	16.0 (11.8-30.0)	**<0.001**
CRT C72S (TGT-TCT)	21.16 (3.1-39.1)	23.0 (3.4-36.8)	14.9 (9.3-23.6)	**0.001**
mdr1 copy number	1.1 (0.8-1.4)	1.1 (0.3-2.0)	1.9 (0.7-5.4)	**<0.001**

Conclusions

- Mutant genotypes for various molecular markers of drug resistance were highly prevalent among *P. falciparum* cases imported to Ontario from sub-Saharan Africa (Table 3)
- There was a significant decrease in mutant alleles between 2008-09 and 2017-18 among wildtype genotypes for the following: *atpase* 623; *mdr1* 86 and 1246 (Table 3)
- No mutations were observed at *atpase* 769, *cytb* 268, *dhfr* 16, *mdr1* 1034 and 1042 (Table 3)
- There was a significant increase in mutant alleles between 2008-09 and 2017-18 among wildtype genotypes for the following: *atpase* 623, 769*; cytb* 268N; *dhps* 581*; mdr1* 1034T, 1034R, 1042, 1246 (Table 4)
- An absence of mutations during all time periods within the *k13* gene indicate a lack of resistance to artemisinin in this sample set, but few cases were imported from southeast Asia (data not shown)
- Co-mutations in multiple genes suggested potential resistance to more than one anti-malarial.
- Observation of co-existence of minor genotypes in relatively high frequency (≥20%) confirms the heterogeneous nature of infection, which may lead to differential drug resistance levels and therapeutic responsiveness.

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Epidemiological Update on Fever in Returning Travelers to Ontario from the 'Rapid Assessment of Febrile Travelers' (RAFT) Programme

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Introduction

- Fever in the returned traveler is a common syndrome, occurring in 17% of ill returned Canadian travelers and new immigrants presenting for care after travel[1].
- Although often due to self-limited infections, such as travelers' diarrhea, fever after travel may indicate serious and potentially life-threatening causes, such as malaria, dengue, or typhoid fever, as was the case in 28% of febrile returned Canadian travelers or new immigrants studied recently[1].
- National Canadian guidelines on the assessment of febrile returned travelers have been published and have been adapted into an ED decision-algorithm to standardize the evaluation and disposition of such patients, through creation of the "Rapid Assessment of Febrile Travelers" (RAFT) Program[2,3].
- The RAFT program facilitates the collection of epidemiological data regarding returning febrile travelers, to understand how destination relates to disease risk.

Methods

- From 2016-2018, RAFT patients were seen in the Tropical Disease Unit within 24 hours after referral from the Emergency Room where the Rapid Assessment Algorithm was followed.
- Criteria for RAFT referral include: presentation to participating EDs, reported fever, and travel outside of Canada within the past year. Exclusion criteria include *Plasmodium falciparum* malaria, and fulfillment of admission criteria such as unstable vital signs or major lab derangements.
- Epidemiological data about all returning febrile travelers including demographics, represented geographic regions, and final diagnoses were collected and analyzed using descriptive statistics.

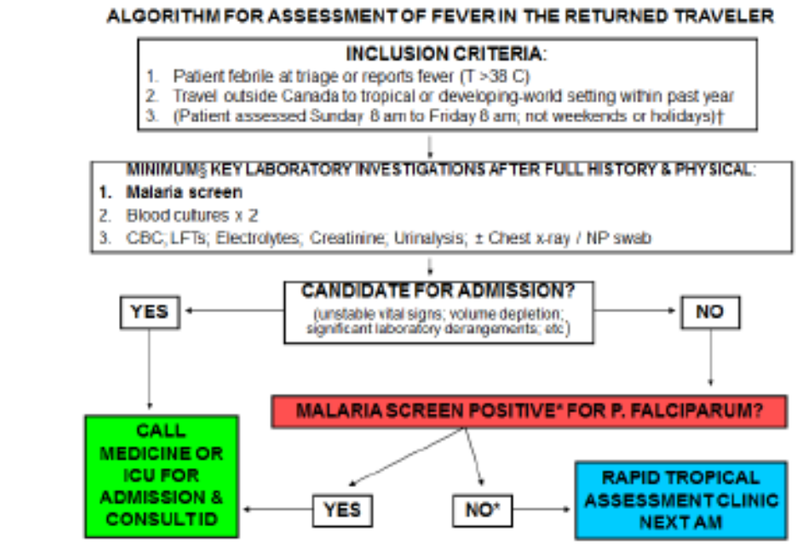

Figure 1. Rapid Assessment Algorithm for Fever in the Returned Traveler
† Between Friday after 8 am and Sunday before 8 am, if the patient does NOT have P. falciparum or otherwise fulfill admission criteria, the patient should STILL be referred to GIM or ID for disposition (as per standard procedure).
§ Additional investigations should be based on clinical judgment.
*If malaria screen is positive for P. vivax, P. ovale, or P. malariae (ie, non-P. falciparum), please initiate chloroquine therapy: 4 tablet loading dose (620 mg base), followed by 2 tablets 6 hours later. If malaria screen is positive for P. vivax & the patient traveled to Papua New Guinea or Indonesia, please initiate Malarone therapy: 4 tablets PO x 1 with food.

Results

Figure 1. Distribution of regions traveled by patients referred to RAFT clinic.

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Results

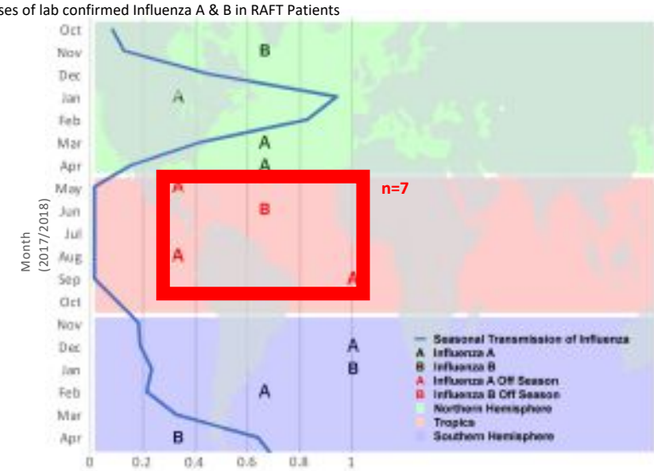
Table 1. Breakdown of diagnoses of patients referred to RAFT clinic, n=482

Diagnosis	#	%
Gastrointestinal Syndrome	**121**	**25.1**
Travelers' Diarrhea	45	9.3
Enteric Fever	21	3.3
Salmonella typhi	15	2.9
Salmonella, non-typhoidal	4	0.8
Post Infectious IBS	12	2.5
Gastritis	8	1.7
Campylobacteriosis	5	1
Cryptosporidiosis	5	1
Escherichia coli	3	0.6
Shigella	3	0.6
Blastocystis hominis	2	0.4
Dientamoeba fragilis	2	0.4
Giardiasis	2	0.4
Helicobacter pylori	2	0.4
Hepatitis A	2	0.4
Choledolithiasis	1	0.2
Clostridium difficile	1	0.2
Clostridium perfringens	1	0.2
Enterococcus faecalis	1	0.2
Food Poisoning	1	0.2
Liver Injury	1	0.2
Norovirus	1	0.2
Strongylodiasis	1	0.2
Ulcerative Colitis	1	0.2
Non Specific Viral Syndrome	**122**	**25.3**
Viral Syndrome	110	22.8
Ebstein-Barr Virus	5	0.8
Mononucleosis-like illness	1	0.2
Cytomegalovirus	4	0.8
Post Infectious Fatigue	3	0.6
Screening	**10**	**2.1**
Schistosomiasis	10	2.1
No Diagnosis	**7**	**1.5**

Diagnosis	#	%
Respiratory Syndrome	**101**	**21**
Viral URTI	39	8.1
Influenza	29	6
Pneumonia	19	3.9
Pharyngitis	9	1.7
Group A Streptococcus	7	1.5
Haemophilus pharyngitis	1	0.2
Sinusitis	2	0.4
Bronchitis	1	0.2
Otitis Media	1	0.2
Legionellosis	1	0.2
Vector-borne	**62**	**14.1**
Flaviviruses	36	0.2
Dengue	27	5.6
Zika	6	1.3
Japanese encephalitis	1	0.2
West Nile Virus	1	0.2
Malaria	10	2.1
P. falciparum	5	1
P. vivax	3	0.6
Non falciparum	2	0.4
Rickettsial infection	7	1.5
African Tick Bite Fever	5	1
Mediterranean Spotted Fever	1	0.2
Chikungunya	3	0.6
Insect Bite	3	0.6
Leishmaniasis	2	0.4
Tick Bite	1	0.2
Fever with Lymphadenopathy	**7**	**1.5**
Tuberculosis	4	0.8
Coccidioidomycoses	1	0.2
Mumps	1	0.2
Parotitis	1	0.2

Diagnosis	#	%
STI / Genitourinary	**18**	**3.7**
Acute HSV-1	5	1
Cystitis	5	1
Pylelonephritis	4	0.8
Bacterial Vaginosis	1	0.2
Chlamydia	1	0.2
Prostatitis	1	0.2
Yeast Infection	1	0.2
Non Specific Skin / Soft Tissue	**22**	**4.6**
Cellulitis	5	1
Rash	3	0.6
Shingles	3	0.6
Myiasis	2	0.4
Staphylococcus aureus	2	0.4
Lice	1	0.2
MRSA cellulitis	1	0.2
Dermatitis	1	0.2
Non TB Mycobacterium	1	0.2
Perianal abscess	1	0.2
Varicella	1	0.2
Cordylobia anthropophaga	1	0.2
Other / Environmetal	**8**	**1.6**
Altitude Sickness	1	0.2
Arthritis	1	0.2
Ciguatera	1	0.2
Drug reaction	1	0.2
Endocarditis	1	0.2
Malaria Insomnia	1	0.2
Mass	1	0.2
Needlestick injury	1	0.2
Bacterial Zoonosis	**4**	**0.8**
Leptospirosis	2	0.4
Brucellosis	1	0.2
Rat Bite Fever	1	0.2

Figure 2. Cases of lab confirmed Influenza A & B in RAFT Patients



Summary and Conclusions

- Understanding the risk of disease and how it corresponds to geographic location travelled is critical in the evaluation of a febrile returned traveler as it will inform decision-making about diagnostic testing and optimizing treatment plans.
- Among the 29 lab-confirmed cases of influenza evaluated in RAFT, off-season transmission accounted for a quarter. Influenza circulates year-round in tropical regions and seasonally in temperate regions with peak transmission from October to March in the northern hemisphere and from April to October in the southern hemisphere[4]. Clinicians should have it on their differential diagnosis and perform nasopharyngeal swabs on returned travelers with influenza-like illness, regardless of month or season.
- Gastrointestinal, nonspecific viral, and respiratory syndromes were well represented clinical diagnoses, with travelers diarrhea and viral upper respiratory tract infections being the top causes of fever in our subset of travelers.
- Assessing the travelers disease risk by geographic region can guide pre-travel counseling and help clinicians recommend appropriate preventative measures and vaccines for at-risk destinations.

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What's New in Environmental Illnesses of Travel: Updated Guidelines from the Wilderness Medical Society

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Introduction

- Global rates of travel have skyrocketed in recent years and with this, travelers have also become increasingly adventurous.
- With this rising trend, travelers are at an increased risk of environmental illness and exposures and travel specialists must develop expertise in evidence-based therapeutics and risk-mitigation of environmental illness.
- The Wilderness Medicine Society (WMS) has recently published updates treatment and prevention guidelines on acute altitude sickness[1], frostbite[2], heat illness[3], and avalanche and non-avalanche snow burial[4].
- With the fast expansion of wilderness medicine research, a summary of the practice guidelines as well as a review of the emerging evidence is provided here.

Objective

- To highlight emerging evidence of clinical significance in wilderness medicine, for the purpose of updating and guiding travel medicine specialists caring for patients at risk of environmental exposures.

Methods

- Environmental illness management procedures were organized and reviewed
- Updated guidelines were compared with previous versions, and the evidence prompting new guidelines were reviewed and highlighted
- A concise literature search was conducted to assess the future direction of environmental-illness research and foresee the projection of future updated guidelines

Results

Table 1: Summary of WMS guidelines updated in 2019

Guidelines	Evidence	Grading
Acute altitude illness		
Inhaled budesonide should not be used for altitude illness prophylaxis.	Though small improvement in spirometry and oxygen saturation was reported, results could not be replicated in more methodologically sound clinical trials.	1C
Acetaminophen should not be used for AMS prevention.	Study used improper control measures.	1C
Hypoxic tents can be used for facilitating acclimatization and preventing AMS, provided sufficiently long exposures can be undertaken regularly over an appropriate number of weeks and other factors, such as sleep quality, are not compromised.	On account of evidence from a placebo-controlled utility study that indicated a lower incident of AMS among users.	2B
Acetaminophen can be used to treat headache at high altitude, but not to treat AMS or HACE.	A randomized controlled trial has found that acetaminophen can be used to alleviate high-altitude headache but there is no evidence of its ability to effectively treat AMS or HACE.	1C
Ibuprofen can be used to treat headache at high altitude, but not to treat AMS or HACE.	A randomized controlled trial has found that ibuprofen can be used to alleviate high-altitude headache but there is no evidence of its ability to effectively treat AMS or HACE.	1C

Results - continued

Guidelines	Evidence	Grading
No recommendation can be made regarding use of continuous positive airway pressure (CPAP) for AMS treatment.	Although feasibility of administering CPAP to treat AMS has been demonstrated, there is a lack of systemic research to support it.	N/A
Nifedipine should be used for HAPE treatment when descent is impossible or delayed and reliable access to supplemental oxygen or portable hyperbaric therapy is unavailable.	According to a prospective, cross-sectional study, addition of nifedipine to descent, oxygen, and rest did not provide additional benefit in terms of time to resolution of hypoxemia and radiographic opacities or hospital length of stay.	1C
CPAP may be considered for treatment of HAPE when supplemental oxygen or pulmonary vasodilators are not available or as adjunctive therapy in patients not responding to supplemental oxygen alone.	Although CPAP may be considered as an alternate or adjunctive avenue of therapy, but is not highly recommended due to lack of systematic evidence and limitations on its overall utility and feasibility	2C
Acetazolamide should not be used for treatment of HAPE.	Acetazolamide should not be used for treatment of HAPE.	1C
Frostbite injuries		
Although further studies are needed to determine the absolute efficacy of tPA for frostbite injury and to compare intra-arterial tPA to IV prostacyclin, monitored administration of IV or intra-arterial tPA within 24 h of injury is recommended at a dosage of a 3 mg bolus followed by a 1 mg/mL infusion, all while administering heparin at 500 units/h.	As of the start of 2019, the recommendation for immediate intravenous or intra-arterial thrombolytic therapy for deep frostbite injuries had been further validated through evidence from one randomized controlled prospective trial (tPA plus iloprost, n=16), 3 retrospective cohort studies (n=59), 8 retrospective case series (n=130), and 3 case reports.	1C
If available, appropriate imaging, including single photon emission computed tomography (CT)/CT, should be used to assess tissue viability and guide timing and extent of amputation.	Kraft et al. demonstrated the ability of single photon emission CT/CT to enable early and precise anatomic localization of nonviable tissue in a study assessing 7 frostbite patients.	1C
Consider iloprost for deep frostbite to or proximal to the proximal interphalangeal joint; within 48 h after injury, especially if angiography is not available; or with contraindications to thrombolysis.	A randomized trial assessing the efficacy of aspirin plus 1) buflomedil; 2) iloprost; or 3) intravenous tPA plus iloprost in 47 frostbite patients found a 0% amputation rate in the iloprost group. From randomized trials to case series, iloprost showed favourable effects if administered within 72 hours of injury, with no serious side effects reported afterwards.	1B
Heat Illness		
Heat syncope patients or individuals at risk for heat syncope to always take caution prior to participating in strenuous exercise, and to seek cardiology diagnostics after a syncopal episode, especially those that are recurrent and inconsistent with exercise-associated collapse.	Case reports have linked acute heat stress to precipitating electrocardiogram changes, symptomatic arrhythmias, and cardiac arrest with features of underlying Brugada syndrome.	2C

Summary of guidelines on Avalanche and non-avalanche accidents[4]:

- the most effective preventative measures include avalanche avoidance, burial avoidance, trauma minimization, and asphyxia avoidance
- Therapeutic protocol involved cardiopulmonary resuscitation and advanced life support based on injury
- Recommendations were based largely on non-peer reviewed publications and data as per research availability on the topic

Discussion

- As new evidence has emerged, prevention and treatment strategies, as well as their respective gradings, have been updated for guidelines on acute altitude sickness, frostbite and heat illness.
- In accord with WMS panel, the following topics can be anticipated in future guideline updates:
  - For acute altitude sickness: systematic evidence on adjunctive therapies and pediatric illness management
  - For frostbite injuries: medical inquiry into prevention medication and therapeutic management of injuries that yield better long-term outcomes
  - For heat illness: creating sound and ethical methodological premises for conducting clinical trials that accurately portray the physiology
  - For avalanche injuries: conducting more accurate simulation trials to assess efficacy of preventative and therapeutic interventions
- Guidelines may also benefit from further exploring the clinical concerns of medical professionals when administering therapeutic interventions for environmental-related illnesses (ex. afterdrop phenomenon[5] as a concern of frost-bite management)

Conclusion

- In 2019, the WMS provided updated guidelines on the prevention, treatment and long-term management of acute altitude illness, frostbite injuries, and heat illness.
- New prevention and treatment guidelines were also provided for avalanche and non-avalanche snow burial.
- These systematically-derived medical recommendations expand the scope of paradigms in travel medicine through informed implementation of recommendations within medical policies and inspiring further research and involvement of clinical expertise.

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An Update on the Role of Imaging in the Care of Patients with Genitourinary Schistosomiasis: A Systematic Review

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Introduction

- Schistosomiasis is infectious disease caused by parasitic worms
- Causes significant morbidity and mortality globally but especially in South America, Asia and Sub-Saharan Africa
- Genitourinary Schistosomiasis is caused by *Schistosoma haematobium* which is endemic in Africa and the Middle East
- Infection leads to severe fibrosis of the urogenital tract and can cause serious lesions in organs like the bladder
- Previous work has been to show the use of imaging as a diagnostic tool for schistosomiasis (ie hepatosplenic)

Objective

- To search available literature regarding the role of imaging in the evaluation of patients with genitourinary schistosomiasis for use of initial risk stratification and management

Methods

- Five databases were searched: Ovid Medline, EMBASE, Cochrane Library of Systematic Reviews, Epistemonikos and LILACs from database inception to December, 2020.
- Titles, abstracts and full-text articles were screened by two reviewers.
- Data extraction was performed by the reviewers.
- Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) was employed

Schistosomiasis		Species		Imaging		Organs
Schistosomiasis	AND	haematobium	AND	CT	AND	Bladder
				(Computed AND tomography)		Ureter
Schisto*		haematob*		Ultraso*		Ureter*
				Sonogr*		Genital
				MRI		Prostate
				(Magnetic AND resonance AND imaging)		Seminal vesicle
				Echo		
						Vas deferen*

Table 1. Search Strategy

Included	Excluded
All study types: Observation (Cohort, case control, cross-sectional), intervention, case studies and case series	Non-human and mouse model studies
Studies including Schistosoma haematobium(active or past infection)	Non-genitourinary schistosomiasis
Utilization of any form of medical imaging	Lab studies
Assessment of ureter, kidney, bladder, genitals	Conference Abstracts

Table 2. Inclusion and Exclusion Criteria

Results

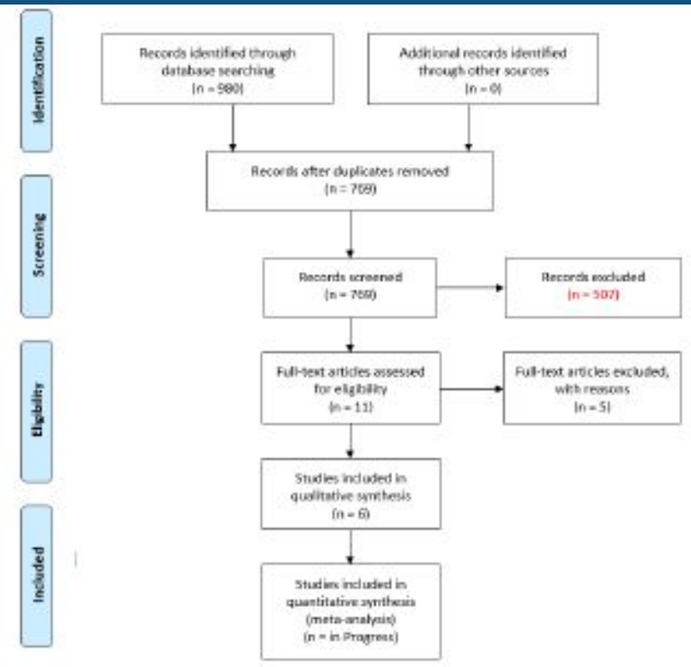

Figure 1. PRISMA Flowchart

Study Name	Baseline Characteristics	Patients diagnosed with Schistosoma haematobium	Imaging used	Patients with Bladder Abnormalities detected by imaging
Tohon (2011)	School age children in 5 endemic villages in Niger	76.8%	Ultrasound	37.7%
Brouwer (2003)	Primary school students from 3 schools in rural Zimbabwe	71%	Ultrasound	27% had bladder masses and thickenings. 50% of infected students had bladder damage
Serieye (1996)	Inhabitants in an established endemic area in Madagascar, older than 5 years who had no prior anti-schistosomal treatment	75.9%	Ultrasound	47%
Deniaud (2020)	Sub-Saharan African migrants who went to health-care consultations in Paris. Most cases were from West Africa	100%	Ultrasound	32.6% of the 86 individuals who underwent ultrasonography
Figueiredo (2013)	Patients at the Urology Service of the Americo Boavida Hospital in Angola aged between 18-75 years	27%	Ultrasound	55.8% with hyper-echogenicity's and 23.1% with bladder masses
Nmorsi (2007)	Volunteers who lived in Nigeria	31.2%	Ultrasound	55.8% wall thickness, 69.8% abnormal shape. 27.9% irregular wall, 23.3% masses, 4.7% had pseudopolyps, 69.8% has echogenic particles and 55.8% had calcifications

Table 3. Preliminary Baseline Characteristics of Included Studies

Discussion

- Ultrasound was able to show abnormalities in the bladder caused by schistosomiasis
- Imaging was able to show bladder masses, hyperechogenicities, general lesions along with calcifications caused by *S. haematobium* infection.
- Therefore, imaging is an important tool for risk stratification & management caused by schistosomiasis
- Synthesizing the current literature on imaging evaluating genitourinary schistosomiasis will strengthen the current body of knowledge as well as translate into clinical recommendations that will improve risk stratification and management of urinary schistosomiasis

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Influence of Host Nutriome on Immunological Control of Trypanosoma cruzi Infection

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Introduction

- Host nutritional status may impact humoral & cellular mechanisms, modulating the immunologic control of parasitic infections
- Insufficient or surplus micronutrients can weaken the immune systems' function, resulting in poor immunologic control of protozoal infections
- Chagas disease, caused by Trypanosoma cruzi, is heavily influenced by the host's immune system, which can be modulated by the host's nutritional status
- To further understand this, we intend to study the relationship between tissue-based protozoal infections & host micronutrient status

Methods

- Combinations of search terms such as Parasite* AND (Immunology OR Immunity OR Immune System OR Immune Function OR Immune Impairment OR Immune Response OR Immune Status) from database inception to February 10, 2020 were searched in five electronic databases
- Screening was performed independently by two reviewers with discrepancies arbitrated by a tertiary reviewer
- A thorough bias assessment will be carried out using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach following screening

Results

Author, Year	Country	Design	Population	Sample Size	Assessment / Intervention	Mean Age ± SD	Sex (F:M)	Outcomes
[1]Junior, 2019	Brazil	Observational Cohort	Overweight adult males with cardiac and indeterminate forms of Chagas Disease	64 Indeterminate (46) Cardiac (18)	Serum vitamin D (via 25(OH)D3), and cathelicidin LL-37	Indeterminate (60) Cardiac (62)	0:46	Patients with the cardiac form had lower levels of serum 25(OH)D3 (p=0.03), however cathelicidin was similar between groups.
[2]Castilhos, 2017	Brazil	Case-Control	Age, sex, & co-morbidity matched Chagas cases and healthy controls	162 Cases (81) Controls (81)	Nutritional status via food frequency questionnaire and diet quality via the BHEI-R	Cases (63 ± 13.5) Controls (66 ± 10.7)	102:60	Chagas group had a lower intake of energy, vitamins A, D, and E, magnesium, and selenium, and a higher intake of lipids consistent with an inflammatory diet (p<0.0001 - p=0.0060). No statistically significant difference in BHEI-R.
[3]da Silva, 2017	Brazil	Randomized Control Trial	Patients > 18 years old previously diagnosed with chronic Chagas cardiomyopathy versus healthy controls	40 Intervention (21) Controls (19)	Omega-3 PUFAs at a dose of 3 g/day or a placebo (corn oil) for 8 weeks	Intervention (58.6 ± 11) Controls (55 ± 9.5)	29:19	The omega-3 PUFAs group demonstrated greater improvements in serum triglycerides (−21.1 vs. −4.1; p = 0.05) and IL-10 levels (−10.6 vs. −35.7; p = 0.01)
[4]Rivera 2002	Brazil	Case-Control	Confirmed positive serology for Chagas disease	170 Cases Rio de Janeiro (122) Belo Horizonte (48) 32 Controls Rio de Janeiro (16) Belo Horizonte (16)	Serum selenium, glutathione peroxidase activity, and thyroid-stimulating hormone concentration, during the progression of chagasic cardiomyopathy	Cases: Rio de Janeiro (49 ± 12) Belo Horizonte (43 ± 10) Controls: Rio de Janeiro (33 ± 8) Belo Horizonte (39 ± 12)	Cases: Rio de Janeiro (65:67) Belo Horizonte (17:31) Controls: Rio de Janeiro (7:9) Belo Horizonte (9:7)	Selenium concentration was significantly lower in chronic disease patients than in healthy adults on all accounts (65 ng/mL versus 72 ng/ mL; P< 0.01).

Table 1. Preliminary Data Extraction of Included Studies
Abbreviations: Brazilian Healthy Eating Index-Revised (BHEI-R), Poly-Unsaturated Fatty Acids (PUFA), Interleukin (IL-10)

Results

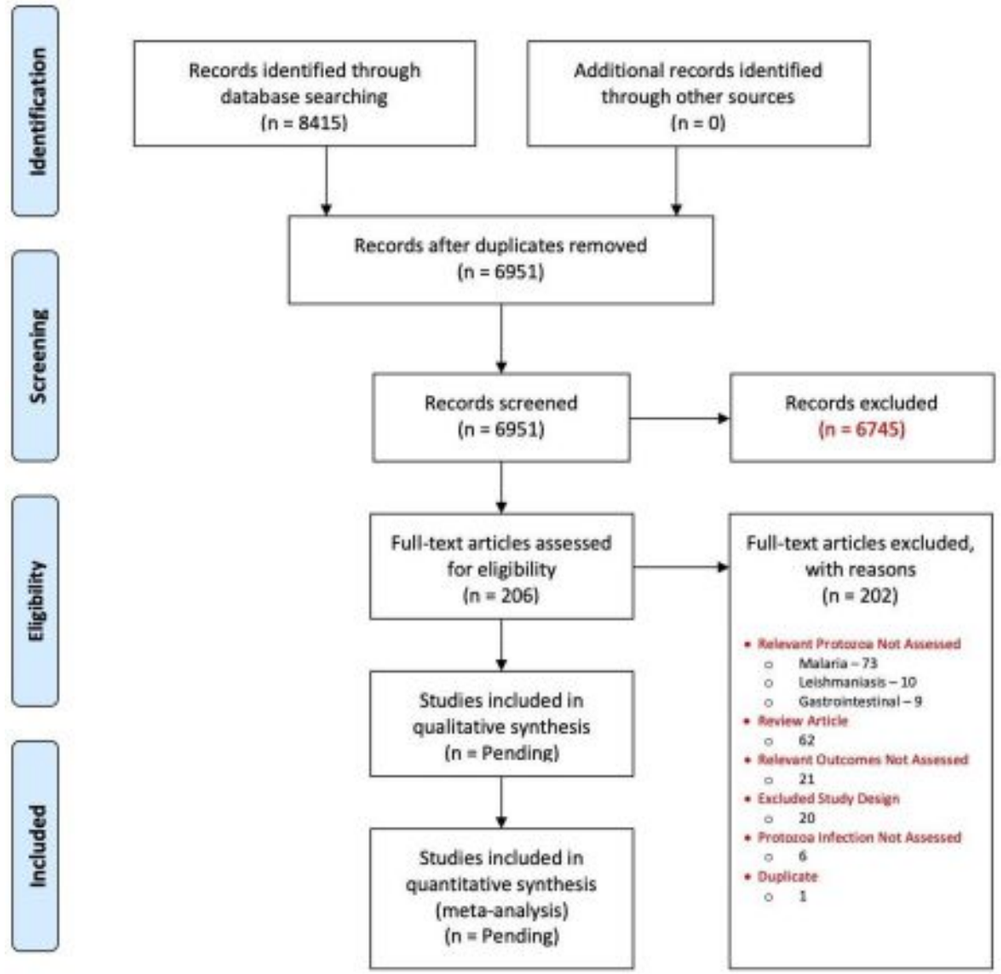

Figure 1. PRISMA Flowchart

Discussion

- Following full-text screening 4 articles remained for inclusion
- A cursory review of relevant articles suggests that the status of magnesium, selenium, omega-3, & vitamins A, D, & E are associated with greater disease severity
- The data collected will be concisely reported to illustrate the findings of published literature regarding the various ways that the function of the immune system in people with Chagas disease alters & deteriorates due to nutrient deficiencies or irregular micronutrient status
- This combined body of information will potentially improve the prognosis of patients with Chagas disease, by informing about possible adjunctive therapies.

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A Systematic Review of Virulence Factors in New World *Leishmania* species

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INTRODUCTION

- Leishmaniasis is a neglected tropical disease divided into three major classifications based on clinical presentation: cutaneous (CL), mucocutaneous (MCL) and visceral (VL)
- Transmitted by the *Lutzomyia* spp. and *Phlebotomus* spp. sandflies, there are up to 2 million cases of Leishmaniasis globally while 350 million people are at risk
- Parasite-determined factors play a complementary role in the pathogenesis of leishmaniasis
- Virulence factors (VFs), or pathogen moieties facilitating disease, can potentiate host cell damage by *Leishmania* spp. by increased expression, host cell invasion, stress tolerance, and modulation of the host immune system
- Due to large eukaryotic genomes in *Leishmania* spp., there is a wide array of VFs which contribute to different aspects of pathogenesis; we aim to synthesize this knowledge by systematically mapping the literature

METHODS

- PubMed (NCBI), MEDLINE (OVID), EMBASE (OVID), Web of Science, and LILACS (VHL) were searched from inception to July 2018 using combinations of the search terms "virulence factor*", "*Leishmania*", and "Leishmaniasis*", while accounting for unique database syntax
- Iterative inclusion and exclusion of search terms was employed to maximize relevant article extraction
- Primarily, molecular and mechanistic pathogenesis studies in various model systems, observational studies, review studies, cohort studies, as well as clinical trials are included
- Synthesis is done by grouping of similar VFs in similar pathogenesis mechanisms, e.g. heat shock
- 760 MEDLINE, 1942 PubMed, 1314 EMBASE, 438 Web of Science, and 8 LILACS records were retrieved for title and abstract screening; after a multi-step de-duplication pipeline, 2620 remained
- All records undergo double-reviewer screening, with tertiary

Virulence Factor	Mechanisms of Pathogenesis
HSP23 HSP60 HSP70 HSP83 HSP90 HSP100 HSP65	• Heat shock proteins • Thermotolerance/survival • Chaperones that facilitate the stabilization of proteins in stressful host environments • Significant expression changes in HSPs as parasite is engulfed in host cells • Aid in adapting from poikilothermic insect vector to a homeothermic mammalian host
LPG	• Lipophosphoglycan • Cell surface anchored molecule • Species-specific sugar component • Required to cause infection in the sandfly hindgut
GP63	• Metalloprotease • Cleaves C3b complement • Halts and hinders innate immunity • Protects parasite from cell lysis
CPB	• Lowered virulence in macrophages • Lowered virulence in mice • Required to cause infection
EF-1alpha	• Elongation factor that is part of the parasite exosome • Blocks Nitric Oxide production • Promotes survival
A2	• Exacerbate parasite-derived immunopathogenesis • Significant in visceral leishmaniasis
MPI	• Catalyze the interconversion of F6P and M6P • Required for glycoconjugates • Loss of MPI leads to loss of surface-anchored VF synthesis, such as leishmanolysin

Species	Virulence Factors
L. mexicana	IPG1, CPB, GP63, LPG, CPC, CHT1, A2, GPI8, ALD1
L. chagasi	G6PD, ARG, GPX, GP46, GP63, HSP70, CPB, A2, HO-1, HSP90
L. amazonensis	GP63, CPB, ICAM-L, KMP-11, LFR1, sAcP, LIT1, LHR1, LIR1, SGL-C, SODA, SMP-3
L. V. braziliensis	FLI1, PGF2S, TXNPX, CPB, GP65, SOD, SST1, HSP20, HSP70, HSP83, MPI, GP63
L. V. panamensis	HSP20, HSP70, HSP83, MPI, GP63
L. V. guyanensis	HSP20, HSP70, HSP83, MPI, GP63, PGPA, MBL2

RESULTS

- Some common parasite-derived pathogenesis mechanisms in *Leishmania* include:
  - Heat shock adaptation to the host environment
  - Evading the immune system
  - Increased expression of survival factors
  - Preventing innate immunity opsonisation
  - Modulation of the host immune system
- Heat shock is mainly directed by heat shock proteins (HSPs):
  - Different HSPs are used preferentially in different species
  - HSP23 can protect against thermal, acidic and oxidative stresses
  - CyP40 is thought to be a co-chaperone that helps the parasite infect macrophages
- Heat shock and resulting thermotolerance is a crucial method by which *Leishmania* species exert their virulence

DISCUSSION

- The ability to comprehensively synthesize all the known literature around parasite-determined virulence factors can open new doors into network-level pathogenesis
- Connecting the dots between virulence factors (if any) to construct a more complete picture of parasite pathogenesis can help better illuminate the underpinnings of different disease manifestations
- Once all parasite-determined VFs are mapped, it can elucidate how they may tie into host-determined immunopathogenesis mechanisms
- Being able to modulate some of these network-level systems can potentially identify novel targets for therapeutics and diagnostics
- This systematic review has implications for painting a fuller picture of parasite-determined *Leishmania* pathogenesis and hence help tie the ends between different VFs, and maybe shed light into host environmental factors

Abstract Presentation Number: 1841

Accuracy of Diagnostics in New World Tegumentary Leishmaniasis: A Systematic Review

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BACKGROUND

- New World Tegumentary Leishmaniasis (TL) is geographically specific to Central and South America and is characterized by cutaneous and mucocutaneous ulcerative skin lesions.
- The species responsible for New World TL include: *Leishmania (L.) amazonensis, L. mexicana, Leishmania Viannia (L.V.) braziliensis, L.V. guyanensis, L.V. panamensis and L.V. peruviana*
- The clinical presentation of New World TL is similar to that of epidemiologically overlapping fungal and mycobacterial infections, thereby necessitating confirmatory diagnostics to inform appropriate treatment[1].
- Laboratory diagnostic techniques for New World TL include the leishmanin skin test (LST); microscopy, culture and molecular assays[1].
- **Our objective was to determine optimal methods to accurately and efficiently diagnose New World TL to improve diagnostic stewardship**

METHODS

- We searched five databases from inception to Oct 2019 including Ovid MEDLINE, Ovid Embase, LILACS, Cochrane Library and Scopus.
- The following search terms were used: ("cut* leish*" OR "muc* leish*" OR "teg* leish*") AND (diagnosis OR diagnostic accuracy OR sensitivity OR specificity OR stard OR test*) AND NOT (viscer*).
- All systematic reviews, diagnostic trials and observational studies were included.
- Titles, abstracts and full-texts are systematically doubled screened by two reviewers with a tertiary arbitrator.
- Full texts were excluded if they did not involve the diagnosis of cutaneous leishmaniasis (CL) and/or mucocutaneous leishmaniasis (ML). Full texts were also excluded if they did not include more than 10 human subjects, specify a reference comparator or use specimens taken from ulcers.
- Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA)[2] and Quality Assessment of Diagnostic Accuracy Studies (QUADAS)[3] will be employed.

RESULTS

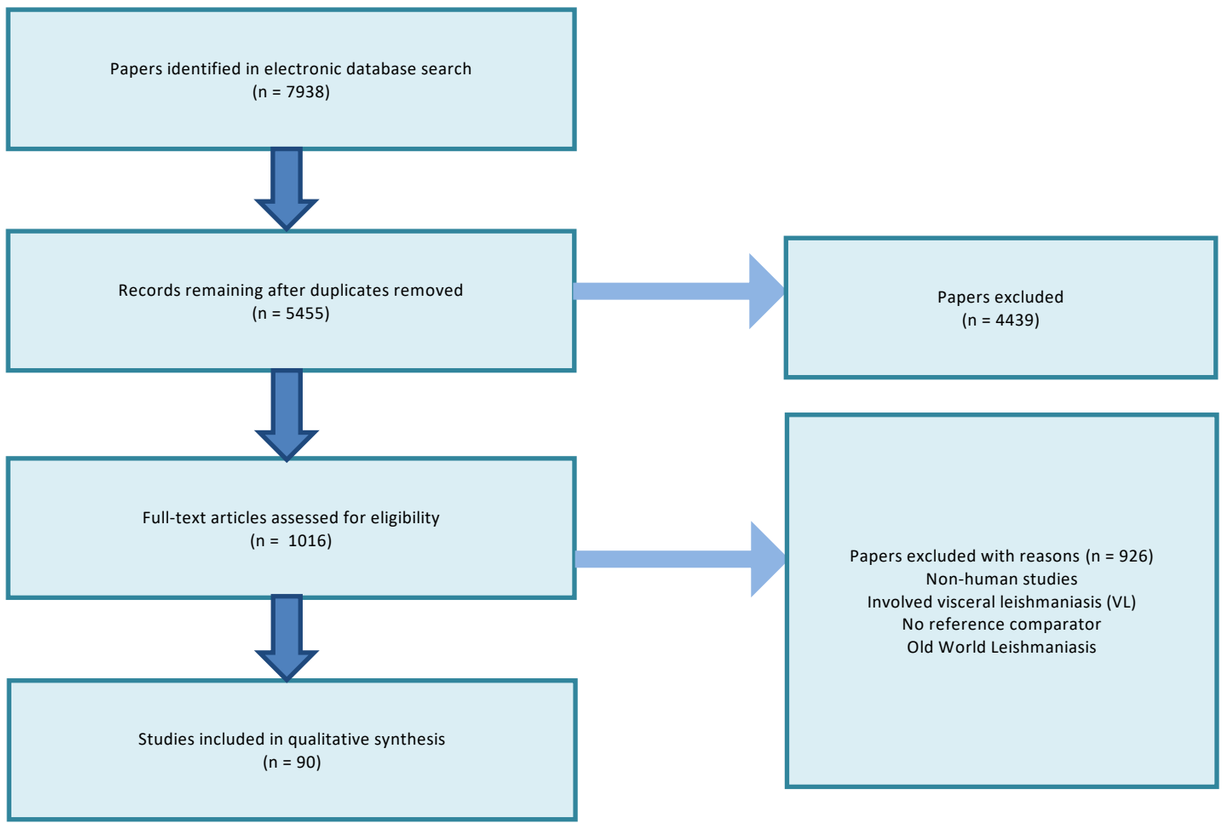

Figure 1. PRISMA flow diagram for database search from inception to October 2019. 90 articles due for extraction in order to perform QUADAS.

Table 1. Descriptive data for eligible full texts including patients with CL, MCL or both in patients with New World Leishmaniasis

Author, Published Year	Lesion Type	# of Patients	Country	Index Tests	Reference Comparator
CordobaLanus et al., 2005	CL and MCL	22	Argentina	PCR-hybridization: 90.5% sensitivity; 100% specificity	Histopathology test: 61.9% sensitivity; 50% specificity
Castillo & Rojas, 1997	CL	61	Colombia	Biopsy sample: dermal scrapings 96.7% positive results using Giemsa staining	Tissue sections: 75.4% positive results using Field staining 50.9% positive results using Wright staining
Gonzalez et al., 2018	CL	49	Panama	kDNA-PCR 46/49: 94% positive results using kDNA-PCR	Biopsy sample taken with Harris punch (Whatman International Ltd.), Tissue sections: Of the 46, 48% positive results using Hematoxylin-Eosin staining
Oliveira et al., 1988	CL and MCL	105	Brazil	Biopsy sample: Montenegro's skin tests 57.1% positive results using Leishman stained inprints of smears	48.6% positive results using parasite cultivation 38.5% positive results using histopathological examination
Schallig et al., 2019	CL	93	Suriname	Loopamp™ Leishmania detection kit : 84.8% sensitivity using microscopy 42.9% specificity using microscopy 91.4% sensitivity using PCR 91.7% specificity using PCR	CL Detect™ Rapid Test: 36.7% sensitivity using microscopy 85.7% specificity using microscopy 35.8% sensitivity using PCR 83.3% specificity using PCR
Wang et al., 2017	CL	16	Ecuador	93.75% sensitivity using ITS1 PCR	56.25% sensitivity using microscopic tissue smear 87.5% sensitivity using Cyt B PCR
Satow et al., 2013	CL and MCL	128	Brazil	kDNA-PCR 112/128: 87.5% positive results using kDNA-PCR	62.8% positive results using Montenegro skin test 61.8% positive results using direct investigation 19.3% positive results using *in vitro* culture

DISCUSSION

- The diagnosis of CL in the study carried out by CordobaLanus et al., 2005 showed that PCR-hybridization had the highest specificity (100%) and sensitivity (90.5%). However, the histology test had the lowest specificity (50%) and lowest sensitivity (61.9%)
- One study also showed that the Giemsa stain was the most specific stain for identifying NW Leishmaniasis in which they report that it identified 96.7% positive results in a sample that was positive for CL
- For the diagnosis of CL and MCL in the study carried out by Satow et al., 2013 it showed that kDNA-PCR identified 87.5% positive results, the Montenegro test following at 62.8% positive results, 61.8% positive results using direct investigation and 19.3% positive results using *in vitro* culture
- Two diagnostic tests were explored in Schallig et al., 2019 which included the use of the CL Detect™ Rapid Test and the Loopamp™ Leishmania detection kit
- Using the CL Detect increased the specificity (85.7%) of microscopy but decreased the specificity (83.3%) of PCR; however, Loopamp Detection kit decreased the specificity (42.9%) of microscopy, but increased the specificity (91.7%) PCR
- Overall, PCR had the highest sensitivity and specificity with histopathology having the lowest sensitivity in the diagnosis of CL and MCL

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Reactivation of New World Tegumentary Leishmaniasis following Iatrogenic Immunosuppression: A Systematic Review of Secondary Prophylaxis

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Introduction

- New World Leishmaniasis is a neglected parasitic disease found in Central and South America caused by a number of protozoal species including *Leishmania (L.) mexicana and L. Viannia (V.)* subgenus complexes
- Recent increases in human migration, travel and urbanization have caused importation into non-endemic areas.
- Immunosuppressive drugs prescribed to this patient population may favor reactivation and dissemination of *Leishmania* spp. and poses a potential problem for rapid diagnosis and treatment for immune-related disorders or solid organ transplants in patients with a prior history of leishmaniasis.[1]

Objective: We aim to synthesize available information to guide clinical management of patients with latent Leishmaniasis undergoing planned iatrogenic immunosuppressive treatment.

Methods

- PubMed (NCBI), Medline (OVID), Embase (OVID), Web of Science (BioSIS) and LILACS (VHL) were searched for between inception to November 15, 2020 with combinations of the search terms “Leishmania reactivation”, “Leishmaniasis” and “Immunotherapy”.
- The systematic review will include case series, case reports, cohort studies, clinical trials and relevant systematic reviews and meta-analyses.
- To assess the quality of the studies reporting therapeutic interventions, the GRADE approach will be utilized.[2]
- LILACS articles will be assessed by Spanish speaking individuals to ensure accurate rating of the inclusion and exclusion criteria.

Results

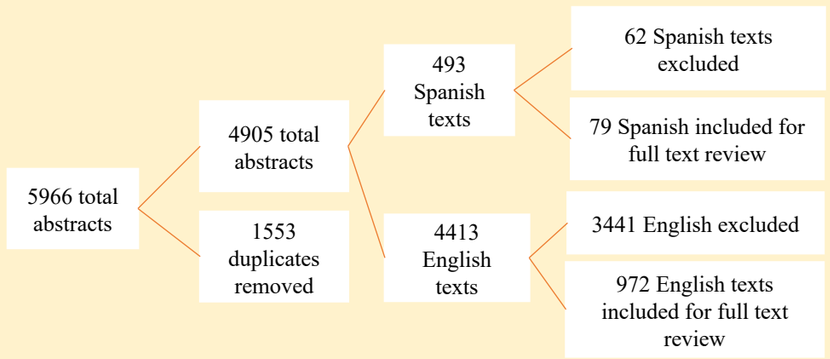

Figure 1: Workflow highlighting current screening progress .

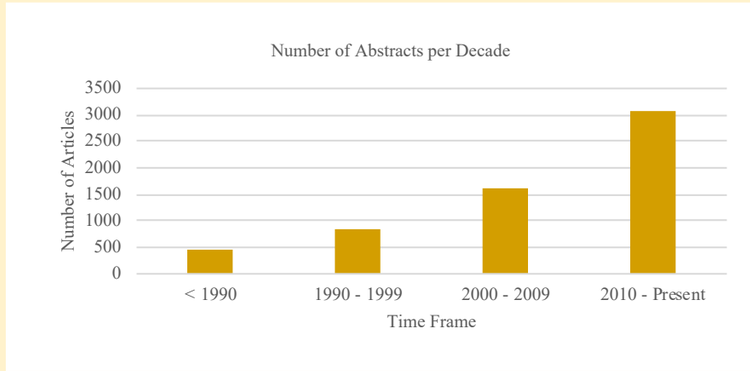

Figure 2: Abstracts by time frame.

Discussion & Conclusions

- Of the 4905 abstracts screened, 1051 have progressed to full text review and 3503 have been excluded for not meeting inclusion criteria. Currently, full text screening for English text will begin with pending completion of Spanish abstract screening. (Figure 1 and 2).
- Alterations to immunological control of latent protozoal infections through immunosuppression may lead to worse health outcomes and increased risks of mucosal and visceral disease presentation in those with initially benign *Leishmania* infections.
- Synthesizing current evidence on the effects of immunosuppressive treatments on active or latent Leishmaniasis can advance our understanding of the management of patients who are undergoing emergency or planned immunosuppression.

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Lifestyle Interventions for Neuropathic Pain: Evaluation of the HEALM Quality Assessment Tool

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Introduction

- The “Grading of Recommendations, Assessment, Development and Evaluations” (GRADE) framework has emerged as a common and transparent approach to evaluating certainty (or “quality”) of evidence for interventional and comparator studies
- However, design elements that are intrinsic to non-RCT lifestyle studies may contribute to poor grading of otherwise high quality and robust trials
- In order to mitigate this bias the “Hierarchies of Evidence Applied to Lifestyle Medicine” (HEALM) framework has been developed
- This framework makes specific considerations for the pitfalls of traditional quality of evidence tools, however, lacks validation against a gold standard assessment tool
- As a result this study seeks to validate the use of HEALM as a strength of evidence tool compared to GRADE for our “Lifestyle Interventions for Neuropathic Pain” systematic review

Methods

- A comprehensive search strategy was conducted using 5 databases from inception to April 2020, that resulted in 7108 articles for screening
- Articles were screened independently by two reviewers and discrepancies were resolved by a tertiary arbitrator during title/abstract, and full-text screening
- A total of 211 articles have been isolated for absolute inclusion / bias assessment
- GRADE and HEALM will be simultaneously implemented to assess their quality of evidence, followed by a comprehensive comparative analysis

Inclusion	Exclusion
Randomized Controlled Trial	Systematic Reviews
Clinical Trials	Reviews
Observational Studies	Conference Abstracts
Cohort Studies	Editorials
Case-control Studies	Animal Studies
Case Series & Reports	In Vitro Studies
Non-English Publications	Trial Descriptions

Table 1. Inclusion and exclusion criteria implemented during title and abstract screening

Database	# Articles
Medline	4128
PubMed	3280
Scopus	106
Embase	66
LILACS	0

Table 2. Number of articles captured by search strategy per database

[1]GRADE	[2]HEALM
Common, transparent, iteratively refined over many years, the gold standard	Very new framework (2019), specifically tailored towards lifestyle medicine
Reviewers make subjective judgements based on individual expertise	Reviewers make objective judgements based on a series of questions and criteria
Certainty / quality of evidence is rated up or down depending on specific considerations	Certainty / quality of evidence is given a grade: **A** (strong/decisive), **B** (moderate/suggestive), **C** (insufficient/inconclusive)
Considerations include: Risk of bias, imprecision, inconsistency, indirection, publication bias (can result in a **lower** rating) Large magnitude of effect, dose-response gradient, residual confounding decreases magnitude of effect (can result in a **higher** rating)	**Questions / criteria focus on:** Mechanisms of action, causality/attribution, generalizability in large populations, considerations of larger time periods (decades, lifetimes, generations) Answers to questions are assigned values (**Yes**=2, **Uncertain**=1, **No**=0) which contribute to the overall score: **A (≥7), B (5-6), C (<5)**
Critiqued for potential bias towards randomized controlled trials over observational studies	Considers common restrictions intrinsic to lifestyle trials Including: cost constraints, adherence challenges, difficulty blinding, limited generalizability (all could potentially result in a lower rating)

Table 3. Comparison of quality of evidence frameworks, GRADE & HEALM

Results

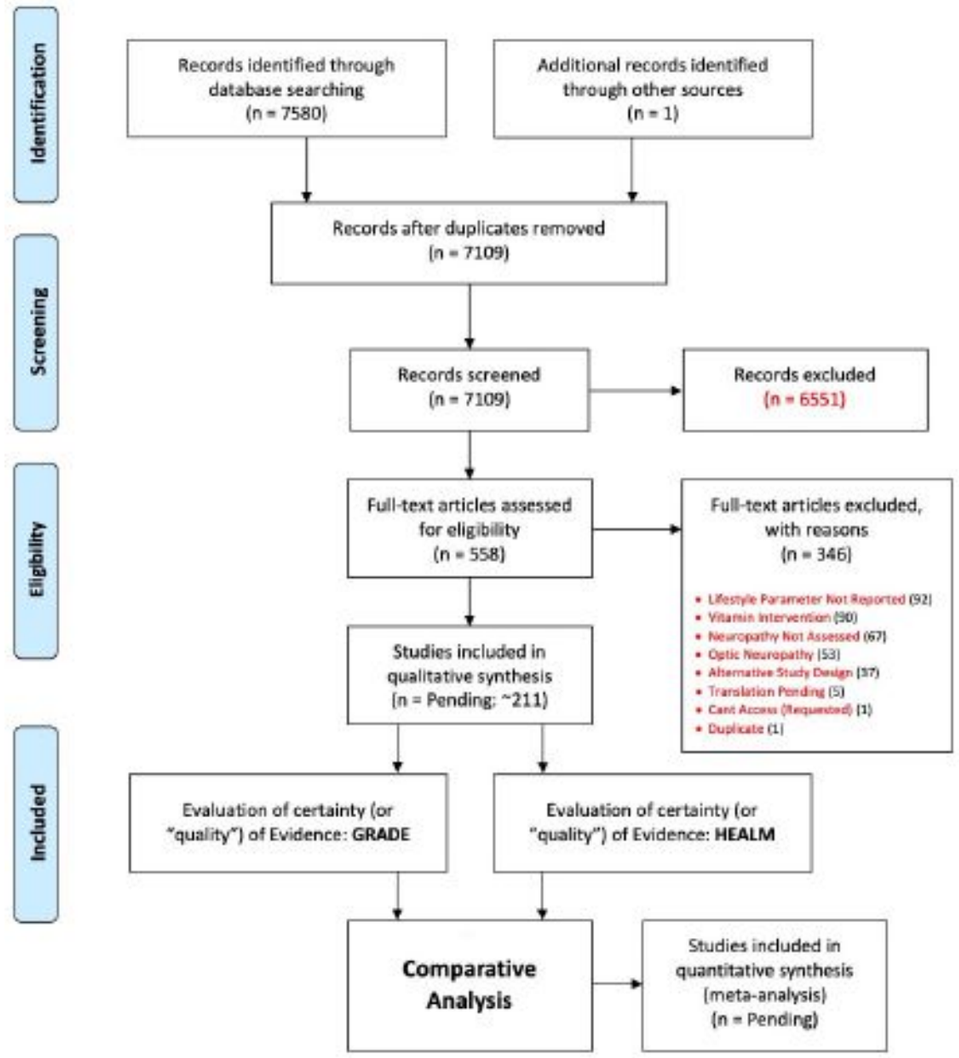

Figure 1. Modified PRISMA Flowchart

Discussion

- The reported quality of evidence for each article will be compared between tools to ascertain HEALM's utility
- It is hypothesized that the quality/certainty of evidence from lifestyle trials will be considered more robust in HEALM vs GRADE due to the intrinsic pitfalls of such research and potential bias within each framework
- Overall this validation project will allow for the succinct organization and dissemination of lifestyle outcomes by public health professionals and clinicians worldwide

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