

**Children's intestinal parasite and nutritional patterns in face of
integrated school garden, nutrition, water, sanitation and hygiene
interventions in central Burkina Faso**

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Summary

Background: Undernutrition is a global public health problem, with over 159 million children under the age of 5 years affected in low- and middle-income countries (LMICs). The determinants of children’s nutritional status are multifactorial. The direct causes of undernutrition in children are insufficient energy and nutrient intake, and recurrent infectious diseases, such as intestinal parasitic infection. Underlying determinants of children’s nutritional status include food insecurity, inadequate child care, weak health systems, and a lack of access to clean water, improved sanitation and adequate hygiene (WASH). To address these challenges, international development organisations are increasingly paying attention to enhancing synergies between agriculture, nutrition and health through multi-sectoral collaboration. Yet, there is a lack of evidence to support the effect of agricultural and health interventions on improving children’s nutritional status, particularly for school-aged children.

To fill this gap, a project entitled “Vegetables go to School: Improving Nutrition through Agricultural Diversification” (VgtS) was developed to address schoolchildren’s nutrition in a multi-pronged approach, through introducing school vegetable gardens and other school-based health, nutritional and environmental interventions. The VgtS project is funded by the Swiss Agency for Development and Cooperation (SDC) and was implemented in five countries, including Burkina Faso, with the overall goal of improving schoolchildren’s nutritional status. This PhD thesis was embedded in the VgtS project in Burkina Faso as operational research study to improve the evidence-base of the interlinked approach as well as to influence the design and implementation of the interventions.

In Burkina Faso, undernutrition, anaemia and diarrhoeal diseases are the leading causes of morbidity in under-5-year-old children. Whilst Demographic and Health Surveys (DHS) and national nutrition surveillance systems have been measuring the height and weight of children under the age of 5 years, there is a paucity of anthropometric data for school-aged children (8-14 years). Likewise, the global burden of disease from polyparasitism of intestinal parasitic infections caused by helminths and intestinal protozoa has not yet been estimated for school-aged children or for the population of any other age group.

Goal and objectives: The overarching goal of this PhD thesis was to assess undernutrition and intestinal parasitic infections among children in rural schools in two regions of Burkina Faso, and to generate evidence on the effects of complementary school garden, nutrition and WASH

interventions on schoolchildren's nutrition and health status. Three specific objectives were pursued:

- (i) to deepen our understanding on undernutrition and associated risk factors among schoolchildren (8-14 years) at a baseline cross-sectional survey before implementing complementary school garden, nutrition and WASH interventions in the two VgtS project regions;
- (ii) to investigate intestinal parasites and its associations with household- and school-level WASH conditions at baseline of the implementation of complementary school garden, nutrition and WASH interventions in the two VgtS project regions; and
- (iii) to assess whether complementary school garden, nutrition and WASH interventions reduce the prevalence of intestinal parasitic infections and improve schoolchildren's nutritional status.

Methods: This study was designed as a cluster-randomised controlled trial, with an equal number of schools randomly allocated to an intervention and to a control group. The intervention group benefited from complementary nutrition and WASH interventions linked to the school garden programme. A baseline cross-sectional survey was conducted between February and March 2015 among 385 children aged 8-14 years in eight randomly selected schools situated in the Plateau Central and Centre-Ouest regions of Burkina Faso. An end-line survey was conducted in the same cohort of children one year after the baseline survey, between February and March 2016.

In both surveys, the same field and laboratory procedures were employed. Schoolchildren's nutritional status was determined by anthropometric measurements. Children were asked to provide single stool and urine samples over two consecutive days, which were examined for infection with helminths and intestinal protozoa. The Kato-Katz method was used to diagnose soil-transmitted helminths (*Ascaris lumbricoides*, hookworm and *Trichuris trichiura*), *Hymenolepis nana* and *Schistosoma mansoni*. The formalin-ether concentration method was employed to detect both, helminths and intestinal protozoa. Urine samples were examined with a urine filtration technique to identify *Schistosoma haematobium* eggs. Prevalence of anaemia was determined by measuring haemoglobin levels in finger-prick blood samples. All children found anaemic or infected with intestinal parasites received treatments according to national guidelines.

Questionnaires were administered to children to determine their knowledge of nutrition and health and their related attitudes and practices (KAP). Children's caregivers/parents were administered a questionnaire to assess basic household sociodemographic and economic characteristics, health KAP and WASH conditions. Water samples from community sources, children's households and children's drinking water cups were analysed for contamination with coliform bacteria and faecal streptococci using a membrane filtration technique.

Results: More than a third (35%) of the children surveyed in the two study regions were undernourished at baseline. The prevalence of undernutrition was higher among children aged 12-14 years compared to their younger peers (8-11 years). Intestinal protozoa were highly prevalent (85%), while faecal-oral transmitted helminths and schistosomiasis showed low prevalence (7% and 4%, respectively) and were mainly of light intensity. Intestinal protozoa were significantly associated with household sociodemographic characteristics. Children from households with freely roaming domestic animals, particularly dogs, showed higher odds of *Giardia intestinalis* infection. Water quality, household drinking water source and storage did not emerge as significant risk factors for intestinal parasitic infections in children. We further observed that undernutrition, anaemia and parasitic infections were strongly associated.

Between the baseline and end-line surveys, the prevalence of intestinal parasitic infections decreased in children from both the intervention and control groups (from 90% to 62%, and from 82% to 72%, respectively) with a significantly stronger decrease in children from the intervention group. Furthermore, adequate handwashing practices before eating and after using latrines at schools increased significantly more among children from the intervention group. Indices of undernutrition and anaemia did not decrease at end-line in the intervention group and water quality remained poor without significant changes.

Conclusions: Undernutrition and intestinal parasitic infections, particularly intestinal protozoa infections, are an important public health concern among schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso. In view of our findings and of the multifactorial aetiology of undernutrition, concerted efforts are needed to concurrently address undernutrition, intestinal parasitic infections and access to WASH among schoolchildren.

By conducting repeated cross-sectional surveys in a cohort of children, this study showed that a combination of nutritional and WASH-based interventions linked to a school garden programme and delivered through a school platform, holds promise for improving schoolchildren's health

and nutritional status. Our findings call for sustaining the achievements made and for increased public health measures tailored to school-aged children, through multi-sectoral school-, household- and community-based programmes.

List of Abbreviations

ALU	Albert-Ludwigs-Universität Freiburg
AVRDC	World Vegetable Center
BMI	Body mass index
CGIAR	Consultative Group on International Agricultural Research
CLTS	Community-led total sanitation
DALY	Disability-adjusted life year
DHS	Demographic and Health Survey
EED	Environmental enteric dysfunction
FEC	Formalin-ether concentration
GBD	Global Burden of Disease
HAZ	Height-for-age z-score
HIC	High-income countries
JMP	Joint Monitoring Programme for Water Supply and Sanitation
KAP	Knowledge, attitudes and practices
LMICs	Low- and middle-income countries
MDA	Mass drug administration
MNDs	Micronutrient deficiencies
NFSI	Nutrition Friendly School Initiative
NTD	Neglected tropical disease
PCR	Polymerase chain reaction
PEM	Protein-energy malnutrition
PhD	Doctor of Philosophy
RCT	Randomised controlled trial
SD	Standard deviation
SDC	Swiss Agency for Development and Cooperation
SDG	Sustainable Development Goal
SSA	Sub-Saharan Africa
STH	Soil-transmitted helminth
Swiss TPH	Swiss Tropical and Public Health Institute
UNICEF	United Nations Children's Fund
VgtS	Vegetables go to School: Improving Nutrition through Agricultural Diversification
WASH	Water, sanitation and hygiene

WAZ	Weight-for-age z-score
WHO	World Health Organization
YLDs	Years lived with disability
YLLs	Years of life lost

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1 Thesis outline

This PhD thesis is organised into eight chapters, including three peer-reviewed publications and one manuscript currently under review. It begins with an introduction (Chapter 2), which provides a succinct definition of undernutrition and its multifactorial aetiology, with a particular emphasis on intestinal parasitic infections and inadequate water, sanitation and hygiene (WASH) conditions as risk factors (Figure 1.1). A summary of interventions and their impacts on nutrition in children is also provided in this chapter, with a review of the literature on nutrition, WASH, and agricultural interventions. Further, this chapter highlights the identified research needs and introduces the collaborative framework of the “Vegetables go to School: Improving Nutrition through Agricultural Diversification” (VgtS) project. In the following chapter (Chapter 3), the objectives of this PhD thesis are presented.

Chapter 4 describes the innovative study design of this PhD research, including the methodology, the study sites, and the sampling procedure. The study is designed as a cluster-randomised controlled trial, with a baseline and end-line survey. The corresponding manuscript was published in *BMC Public Health* (March, 2016) under the title “Complementary school garden, nutrition, water, sanitation and hygiene interventions to improve children’s nutrition and health status in Burkina Faso and Nepal: a study protocol”.

Chapters 5 and 6 consist of a manuscript each, presenting the findings from the cross-sectional baseline survey. They are respectively outlined as follows: one manuscript on schoolchildren’s nutritional status and associated risk factors accepted for publication at *Infectious Diseases of Poverty* (September, 2016); and one manuscript on schoolchildren’s intestinal parasitic infection status and its associations with household- and school-level WASH conditions published in *Parasites and Vectors* (October, 2016). Chapter 7 presents key results from the end-line survey, assessing the effects of complementary school garden, nutrition and WASH interventions on schoolchildren’s health and nutritional status. This manuscript is currently under review at *Environmental Health Perspectives*.

The last chapter (Chapter 8) provides an overview of the main results of this research. These are discussed in accordance with the PhD study objectives, highlighting the key findings, lessons learned and some methodological limitations to our study. Subsequently, public health implications are outlined, followed by a summary of how this PhD work contributes to the main pillars of Swiss TPH in the field of public health. Finally, a set of conclusions, research needs, and recommendations are provided to authorities in Burkina Faso and the broader international research and development community.

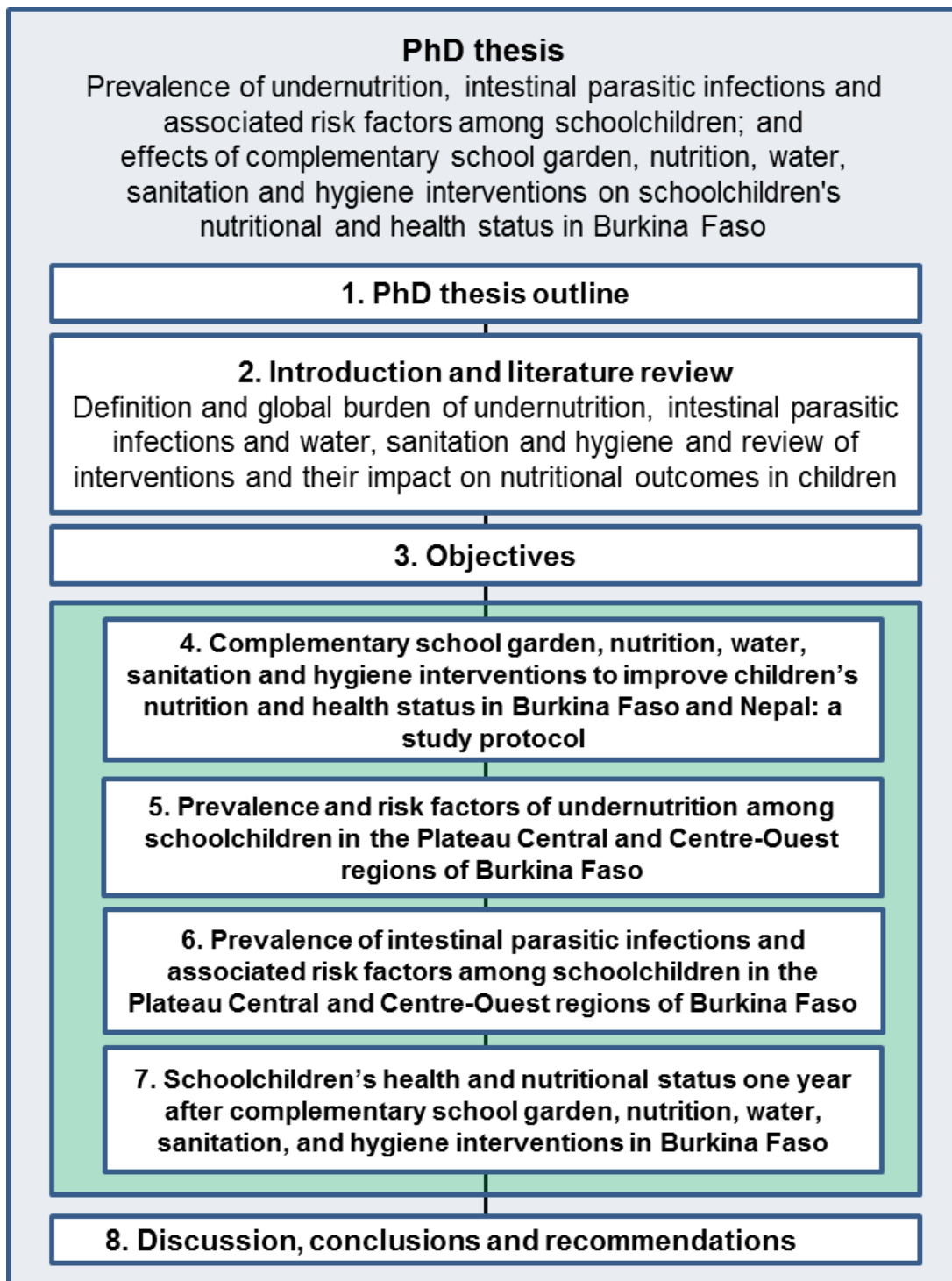


Figure 1.1: Schematic PhD thesis structure

2 Introduction

2.1 Definition of undernutrition

Undernutrition is defined as a state of nutritional insufficiency resulting from a lack of food or from the inability of the body to convert or absorb certain nutrients [1, 2]. Undernutrition encompasses stunting, wasting, and deficiencies of essential vitamins and minerals (collectively referred to as micronutrients), while malnutrition encompasses both, undernutrition and overnutrition (i.e. overweight and obesity due to over-consumption of specific nutrients) [2, 3]. Undernutrition in children mainly develops during maternal pregnancy (in utero) and until the age of 24 months [4].

The four most commonly used anthropometric indices to describe undernutrition in children are: (1) height-for-age (also referred to as stunting, growth stunting or faltering), (2) body mass index-for-age (BMI, thinness), (3) weight-for-height (wasting or thinness), and (4) weight-for-age (underweight) [5-7]. For school-aged children (5-14 years), particularly height-for-age and BMI-for-age are used to characterise their nutritional status, as weight-for-age and weight-for-height are only used for children under the age of 10 and five, respectively [7].

2.2 Aetiology of undernutrition

The causes of undernutrition in children are multifactorial [3, 8, 9]. The determinants of optimum growth and development of children consist of factors operating at different levels of causation [8]. First, the proximal factors (immediate causes) of undernutrition are: (i) inadequate dietary intake and (ii) disease. Second, these proximal factors are influenced by the following underlying causes: (iii) household food insecurity, (iv) inadequate child care, feeding practices, and inadequate health care services, and (v) contaminated household environments (e.g. with helminth, bacteria, protozoa), partially due to inadequate WASH conditions [9] (Figure 2.1). Third, the determinants of children's optimum growth and development also consist of more distal socioeconomic and political factors (basic causes), such as income poverty and lack of capital [8]. This thesis will particularly focus on the proximal factors of undernutrition, investigating the role and contribution of intestinal parasitic infections (helminths and intestinal protozoa) and WASH conditions as risk factors for undernutrition in schoolchildren, which will be introduced in the following chapters.

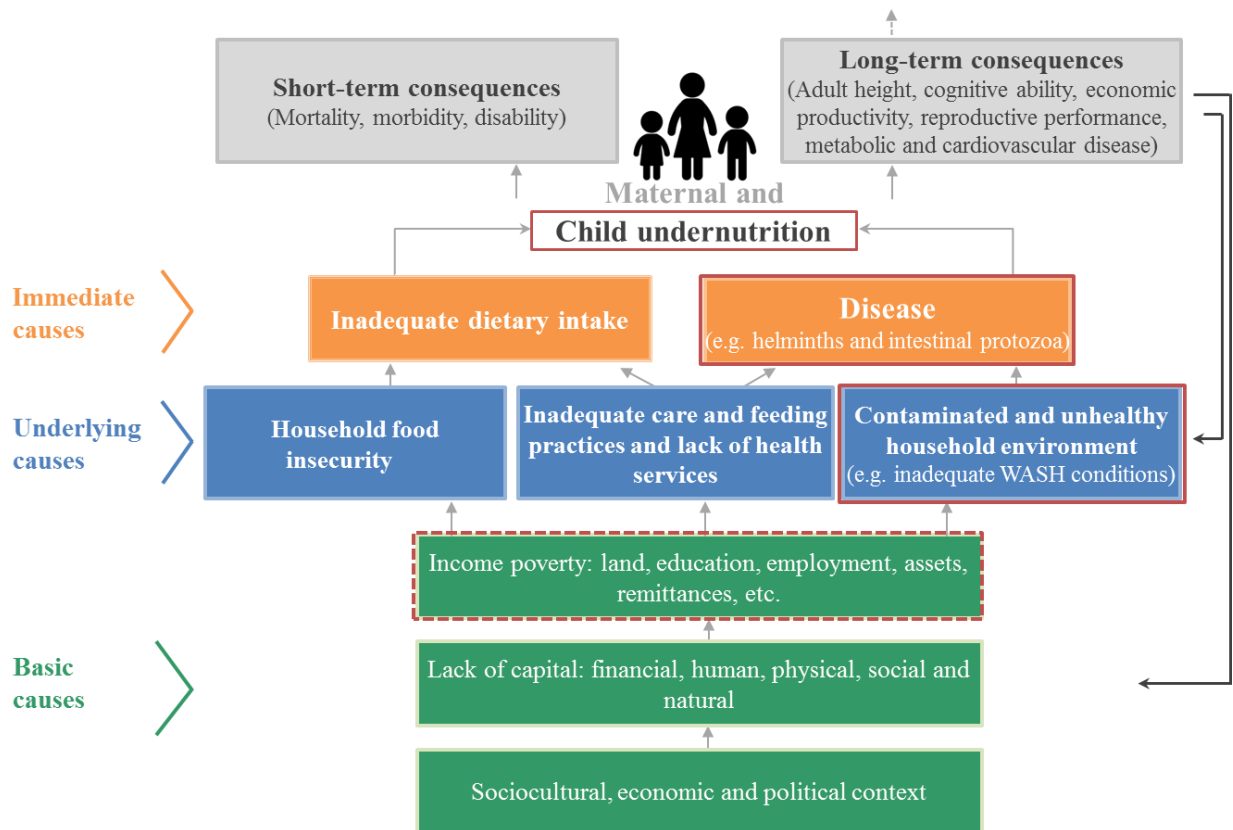


Figure 2.1: Determinants of child undernutrition

The black arrows show that the consequences of undernutrition can feed back to the underlying and basic causes of undernutrition, continuing the cycle of undernutrition, inequities and poverty. Source: adapted from UNICEF (1990 and 2013) and Black et al. (2013). The boxes circled in red highlight the focus of this PhD thesis.

2.3 Inadequate dietary intake

2.3.1 Nutritional deficiencies

Inadequate dietary intake manifests in complex nutritional deficiencies, frequently involving calorie and protein (macronutrients) and various micronutrient deficiencies (MNDs, often referred to as vitamins and minerals) [10, 11]. Vitamin A, folate, iron, iodine, and zinc are the most common MNDs; however, several other MNDs and associated disorders are also of concern (e.g. vitamin E, vitamin B₆, copper, and selenium) [11]. A lack of adequate nutrients (macro- and micronutrients) can lead to impaired growth and development in children [12, 13]. Most important clinical nutritional disorders resulting from inadequate dietary intake are protein-energy malnutrition (PEM) and disorders related to MNDs [12], with iron deficiency as the most common form [13, 14]. Furthermore, undernutrition due to insufficient intake of energy and macronutrients and/or due to specific MNDs impairs important immune functions (e.g. the adequate functioning of immune cells and protecting them from oxidative stress) that are

fundamental to host protection from infectious agents, such as bacteria, viruses or intestinal parasites [12, 13, 15].

2.3.2 Prevalence and burden of undernutrition in low- and middle-income countries and situation in Burkina Faso

Undernutrition is a common public health problem in low- and middle-income countries (LMICs) [16, 17]. In 2011, about 28%, or 159.7 million children under the age of five in LMICs were stunted, 17% (99.0 million) were underweight and 9% (50.3 million) were wasted. Stunting and underweight caused about 14% of total deaths of children under the age of 5 years, while the proportion for wasting was 13% [8]. Particularly in East and West Africa, and South-Central Asia, children under the age of 5 years have the highest prevalence estimates for stunting of the UN sub-regions with 42% (East Africa) and 36% (West Africa and South-Central Asia) [8]. As for many LMICs, undernutrition in children under the age of 5 years in Burkina Faso is highly prevalent [18, 19]; according to the 2016 Global Nutrition Report, 33% of children under the age of 5 years were stunted and 11% were wasted [20]. These global and national estimates are based on data from surveys in the WHO database and other population-based data, such as Demographic and Health Surveys (DHS), which primarily focus on children under the age of 5 years or on adolescents over 15 for sexual and reproductive health issues [8]. These estimates show that there is a lack of anthropometric data for schoolchildren [21-23]. Nevertheless, the Global Burden of Disease Study (GBD) presents data and estimates on the burden of diseases in children aged 5-14 [24], including nutritional deficiencies (PEM, iodine, vitamin A and iron deficiency anaemia). According to the newest estimates of the GBD 2015, the highest burden of the nutritional deficiencies investigated was attributable to iron-deficiency anaemia, causing 4.7% of disability-adjusted life years (DALYs, combining years of life lost (YLLs) and years lived with disability (YLDs)) in children of this age group in Burkina Faso [24].

Although data on schoolchildren's nutritional status (anthropometrics) is scant, schoolchildren still face a considerable burden of nutritional disorders and are often at high risk of acquiring intestinal parasitic infections, particularly helminths in sub-Saharan Africa (SSA) [13, 25, 26]. The interaction between inadequate dietary intake, undernutrition and disease-related consequences is therefore of great importance and will be further highlighted in the following sections.

2.4 Intestinal parasites

2.4.1 Biology and lifecycle

Soil-transmitted helminthiasis

The soil-transmitted helminths (STH) are a group of parasitic nematode worms. The most common species are *Ascaris lumbricoides*, hookworms of the genera *Ancylostoma* and *Necator*, and *Trichuris trichiura*. Human infection is caused without any intermediate host directly through contact with parasite eggs (*A. lumbricoides* and *T. trichiura*) or larvae (in the case of hookworm) that thrive in the warm and moist soil [27]. Once eggs of *A. lumbricoides* and *T. trichiura* are ingested from contaminated foods (raw vegetables in particular) or water, larvae develop and migrate to their final habitat in the intestine. As adult worms, STH can live for years in the human gastrointestinal tract and their eggs are excreted with faeces of infected persons. While *A. lumbricoides* and *T. trichiura* feed on their host's intestinal food content, hookworms suck blood and fluids from grasping and cutting gut tissue [27, 28].

Schistosomiasis

Schistosomiasis, or bilharzia, is an infectious parasitic disease. It is caused by trematode flukes of the genus *Schistosoma*, of which three main species infect humans; *S. mansoni* and *S. japonicum* cause intestinal schistosomiasis, whilst *S. haematobium* infection involves the urinary tract [29, 30]. Schistosomiasis is a water-based disease [31]. Excreted eggs in fresh water hatch and release motile miracidia, which infect a suitable intermediary snail host (i.e. snails from the genus *Bulinus* for *S. haematobium* and snails from the genus *Biomphalaria* for *S. mansoni*). The parasite undergoes asexual replication in the snail (production of cercariae), which are shed back into the water as free-swimming larval stages [30]. Infection of humans occurs through contact with fresh water bodies infested with cercariae, which penetrate the skin of potential hosts. Female and male adult worms settle within the portal veins of their human host, where they mate and produce fertilised eggs. The eggs are excreted in the environment through faeces or urine where they may reach fresh water sources, while some remain trapped in the host tissues where they induce inflammation before dying [30, 32-34].

Intestinal protozoa infections

Numerous protozoa inhabit the human intestinal tract. Most of these protozoa are non-pathogenic commensal, or causing only mild disease [35]. In terms of disease burden and prevalence, the following sections will focus on two common intestinal protozoa; i.e. *Giardia intestinalis*, also often referred to as *G. lamblia* or *G. duodenalis*, causing giardiasis and *Entamoeba histolytica*, causing amoebiasis [36]. The life cycle of both intestinal protozoa species are simple consisting of a cyst stage (long-lived infective stages) and a motile

trophozoite stage. Once ingested, cysts transform to the trophozoite stages, during which they take up nutrients and undergo asexual replication, while some develop into cysts again. Cysts are characterised by a resistant wall and once excreted in stool, they maintain the life cycle by further faecal-oral spread [37, 38]. Both, *E. histolytica* and *G. intestinalis* are transmitted through contaminated water and food, however the latter is relatively uncommon for *G. intestinalis* [39].

2.4.2 Global epidemiology of intestinal parasites and situation in Burkina Faso

Soil-transmitted helminth infections

Latest estimates of the WHO indicate that over 800 million school-age children live in areas where STH are endemic [40]. STH are very common in SSA. Of the estimated 181 million school-aged children in SSA in 2005, almost one-half (89 million) were either infected with *A. lumbricoides*, *T. trichiura* and hookworm, or with a combination of the three [41, 42]. Findings from a systematic review and geostatistical meta-analysis conducted in 2015 showed that the prevalence of overall STHs among school-aged children in Burkina Faso from 2000 onwards was predicted at 10.7%, of which 9.9% for hookworm and 0.4% for both *A. lumbricoides* and *T. trichiura* [43].

Schistosomiasis

Estimates from 2012 suggest that around 163 million people in SSA were infected with one of two *Schistosoma* spp. prevalent in SSA (*S. haematobium* and *S. mansoni*), 57 million (35%) of whom were school-aged children [44]. School-aged children are also particularly affected by schistosomiasis [42]. Before the implementation of national control programmes in Burkina Faso in 2004, a combined prevalence of *S. haematobium* and *S. mansoni* was found in 30 to 50% of school-aged children. Two years after treatment through the national control programme, the prevalence was still between 7 and 13% among school-aged children [45].

Intestinal protozoa infections

Giardia intestinalis is the most frequently reported intestinal parasite worldwide, and has an estimated prevalence range of between 20 to 30% in LMICs [46]. Information on the prevalence of *E. histolytica*/*E. dispar* or *G. intestinalis* infections is scarce and little data are available from SSA and Burkina Faso [47, 48], yet findings from several cross-sectional surveys conducted between 1990 and 2012 in Burkina Faso reported prevalence rates between 23 and 39% for *E. histolytica*/*E. dispar* and 5 and 46% for *G. intestinalis* among all age-groups [49-51].

2.4.3 Intestinal parasitic infections and their contribution to undernutrition in children

STH infections and schistosomiasis cause significant morbidity in LMICs, but are rarely lethal [52-57]. Chronic infections with these parasites can contribute to growth stunting by causing a decline in food intake (loss of appetite), diarrhoea (defined as the passage of three or more loose or liquid stools per day), malabsorption and/or an increase in nutrient wastage for the immune response, all of which lead to nutrient losses and deficiencies, and to further damage of the immune mechanisms [58-60].

Giardia intestinalis and *E. histolytica* are responsible for considerable rates of morbidity and mortality in LMICs. Most cases of *G. intestinalis* and *E. histolytica* infections are asymptomatic. In addition, *E. dispar* infection can coexist with *E. histolytica* and is harmless. However, by colonizing the human small and/or large intestine, infection with *G. intestinalis* can cause persistent and acute diarrhoea (> 14 and < 14 days, respectively) [61, 62]. Infection with *E. histolytica* can cause amoebic colitis (when trophozoites become invasive and cause damage to intestinal mucosa or blood vessels, provoking inflammation, abdominal pain, watery or bloody diarrhoea), and in only rare cases can lead to the development of liver abscesses [63, 64]. Both infection with *G. intestinalis* and *E. histolytica* are associated with underweight, retardation of growth and development in children [62-66].

Intestinal parasitic infections are highly prevalent in school-aged children in SSA and in Burkina Faso more specifically. According to the GBD 2015, intestinal infectious diseases, infections with nematodes, and schistosomiasis are estimated to contribute to 5.9%, 0.2% and 0.1% of total DALYs in children aged 5-14 in Burkina Faso, respectively [24]. Considering their high prevalence and their contribution to the burden of disease in children of this age group, investigating the association between undernutrition and intestinal parasitic infections in school-aged children make this an important area of research [25].

2.5 Water, sanitation and hygiene (WASH)

2.5.1 WASH terminology

WASH refers to the collective term “water, sanitation and hygiene”. Due to their interdependent nature, WASH includes strategies to improve access to an adequate amount of safe water (e.g. water quality, quantity, and distance to water source), adequate sanitation (e.g. access to improved latrines, such as all sanitation facilities that hygienically separate human excreta from human contact to prevent risk of environmental contamination and exposure to faeces harbouring infectious STH eggs or intestinal protozoa cysts), and hygiene practices (e.g. handwashing with soap before eating and after defecation) [67-69].

2.5.2 Global WASH conditions and situation in Burkina Faso

According to the 2015 update report from WHO/UNICEF entitled ‘Joint Monitoring Programme for Water Supply and Sanitation’ (JMP), 30% of the population in SSA had access to an improved sanitation facility, while 27% used unimproved sanitation facilities and 23% practiced open defaecation. In addition, the joint WHO/UNICEF report showed that 32% of the population in SSA had no access to improved drinking water sources [70]. Current levels of handwashing with soap are especially low in SSA, where coverage is at most 50% (of 38 countries for which data were available) [70].

For Burkina Faso, the 2015 WHO/UNICEF/JMP data showed that 8% of the population had no access to improved drinking water sources (3% in urban and 24% in rural areas); 20% of the population lacked access to improved sanitation, whilst 55% practised open defaecation (9% in urban and 75% in rural areas). In 2010, 90% of the population did not have a handwashing facility with water and soap at home [70].

2.5.3 WASH and undernutrition

Safe WASH conditions are a critical underlying determinant of children’s health (Figure 2.1) [9, 67, 71, 72]. Three biological mechanisms, in particular, have been described that plausibly link poor WASH conditions to undernutrition: (1) via STH infections (caused by *A. lumbricoides*, *T. trichiura*, and hookworm) and schistosomiasis [28, 73-75]; (2) via repeated episodes of diarrhoea [76-78]; and (3) through a condition called environmental enteric dysfunction (EED), a subclinical disorder characterised by diminished intestinal absorptive capacity, reduced barrier integrity, and gut inflammation [79-82]. The commonality between these three mechanisms is that chronic exposure to a contaminated environment due to unsafe WASH conditions (e.g. to faeces contaminated with protozoan cysts or oocysts, helminth eggs and viral or bacterial pathogens) causes symptomatic (diarrhoea) or asymptomatic infection (EED) [82]; which in turn can lead to loss of nutrients, malabsorption, impaired digestion and ultimately to decline of childhood growth [73, 83, 84].

Poor WASH conditions are estimated to be responsible for about 50% of child undernutrition in LMICs [85]; and to contribute to 11.5% of total DALYs (105,013 DALYs) in children aged 5-14 in Burkina Faso in 2015, primarily attributed to intestinal infectious diseases (5.7% of total DALYs) and diarrhoeal diseases (4.9% of total DALYs) [24]. In recognition of these estimates, inadequate WASH conditions in Burkina Faso may pose a considerable risk to undernutrition in schoolchildren [86].

2.6 Interventions to address undernutrition in children

2.6.1 Categories of interventions

There are two main categories of interventions to address undernutrition in children: a) nutrition-specific interventions, and b) nutrition-sensitive interventions [87, 88]. While nutrition-specific interventions aim to address the immediate causes of undernutrition (inadequate dietary intake and disease), the objective of nutrition-sensitive interventions is to target the underlying determinants of undernutrition. Key nutrition-sensitive interventions draw on the complementary sectors of WASH and agriculture, as they bear great potential to affect underlying determinants of undernutrition (e.g. access to safe and hygienic environments and to diverse diets) [88].

The focus of interventions aiming to improve the nutritional status of children lies primarily on women and children under the age of five [87]. This is due to women's nutritional status at the time of conception and during pregnancy being important for foetal growth and development. Along with children's nutritional status in the first two years of life, these factors are important determinants for childhood undernutrition [8]. However, children in mid-childhood and early adolescence are the target group of school-based interventions. While primarily aiming at improving school enrolment, attendance, and cognitive outcomes, several school-based interventions have been implemented to address children's health. For example, micronutrient supplementation, school feeding and anthelmintic treatment programmes have been instigated [87, 89]. Table 2.1 presents a summary of the nutritional effects from selected key nutrition-specific and nutrition-sensitive interventions, including school-based programmes.

Table 2.1: Summary of interventions and their impacts on nutrition

Category	Settings/interventions	Findings
Nutrition-specific interventions		
Overall nutrition-specific interventions	Lancet review “ <i>Evidence-based interventions for maternal and child undernutrition</i> ” (2013) [87] <ul style="list-style-type: none"> • Low- and middle-income countries (LMICs) • Children aged under 5 • Interventions: maternal nutrition during pregnancy, infant and young child feeding practices, micronutrient supplementation and management of acute malnutrition) 	Modelling of ten evidence-based nutrition-specific interventions at 90% coverage from the studies included in the Lancet review [87]: <ul style="list-style-type: none"> • 15% reduction in mortality • 20% reduction in stunting • 60% reduction in wasting
School feeding programmes	Cochrane review “ <i>School feeding for improving the physical and psychosocial health of school children</i> ” (2007) [90] <ul style="list-style-type: none"> • LMICs and high-income countries (HICs) • Children aged over 5 • 18 studies included with various interventions: breakfast and food supplements (milk, meat or energy supplements), or school milk only (excluding studies on fortification). 	Small effects on anthropometric indices: <ul style="list-style-type: none"> • Significant weight gain (0.39 kg over 11 months and 0.71 kg over 19 months) and change in WAZ in LMICs [91-93] • Small but non-significant height gain in LMICs [91-93]
Micronutrient supplementation and fortification	Review on the “ <i>Effects of daily iron supplementation in primary-school-aged children</i> ” (2013) [94] <ul style="list-style-type: none"> • 32 studies included, 31 in LMICs • Children aged 5-12 • Intervention: daily iron supplementation 	Improvements of anthropometric indices: <ul style="list-style-type: none"> • Improvement in HAZ [94] and WAZ among anaemic children [95]
	Review on “ <i>can multi-micronutrient food fortification improve the micronutrient status, growth, health and cognition of schoolchildren?</i> ”(2011) [96] <ul style="list-style-type: none"> • Children aged 6-18 in LMICs • Interventions: multiple micronutrients provided via fortification (between 6 and 14 months) 	Out of seven studies including anthropometric measurements: <ul style="list-style-type: none"> • Four studies found a significant beneficial effects on weight and BMI [97-100] • Two studies found a significant beneficial effect on height gain [98, 99]
Anthelmintic treatments	Review on “ <i>mass deworming and child nutrition</i> ” (2016) [101] <ul style="list-style-type: none"> • Children aged 1-19 in LMICs • 22 studies included (four new ones to the previous review) • Intervention: anthelmintic treatments (multiple doses) 	Positive benefits on weight gain found [101]: <ul style="list-style-type: none"> • In areas below 20% prevalence: 0.13 kg • In areas with over 20% prevalence: 0.15 kg • In areas with over 50% prevalence: 0.18 kg
	Review on “ <i>deworming drugs and its effects on nutritional indicators</i> ” (2015) [102] <ul style="list-style-type: none"> • Children aged 1-19 in LMICs • 45 studies included, of which nine RCTs • Intervention: anthelmintic treatments (single and multiple doses) 	Mixed effect on anthropometric indices [102]: <ul style="list-style-type: none"> • <i>Single dose for infected children</i>: 0.75 kg • <i>Single dose for all children in endemic areas</i>: little (less than 0.04 kg) or no effect found • <i>Multiple doses (11 studies)</i>: little (0.08 kg) or no effect on average weight and height gain found
Nutrition-sensitive interventions		
WASH interventions		
WASH interventions	Review on “ <i>Interventions to improve water quality and supply, sanitation and hygiene practices</i> ” (2013) [86] <ul style="list-style-type: none"> • Children aged under 5, only one study included children aged over 5 in LMICs • 14 studies included, of which five RCTs in meta-analysis • WASH interventions: specifically solar disinfection of water, provision of soap, and improvement of water quality 	Meta-analysis including the five RCTs found mixed effects on anthropometric indices [86]: <ul style="list-style-type: none"> • Modest impact on HAZ (greater effect in children aged under 2) • No impact on WAZ and WHZ
	Studies on the “ <i>Effects of sanitation interventions on stunting</i> ” [103-107] (2013-2016) <ul style="list-style-type: none"> • Children aged under 5 in LMICs (Mali, India, Indonesia) • Sanitation interventions: community-led total sanitation, large-scale sanitation programmes 	Mixed effects on anthropometric indices: <ul style="list-style-type: none"> • Two studies reported a significant effect on HAZ [103, 104] • Three studies showed no effect on HAZ [105-107], on WAZ [106, 107], or on BMIZ [107]
	Studies on “ <i>WASH interventions to reduce enteric infections and improve nutritional status</i> ” [108-110] (2013-2015) <ul style="list-style-type: none"> • Children aged under 5 in LMICs (Mali, India, Indonesia) 	Studies currently underway, evaluating biological markers of environmental enteric dysfunctions and anthropometrical indices.
Agricultural interventions		
Agricultural interventions	Lancet review “ <i>Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition?</i> ” (2013) [88]. <ul style="list-style-type: none"> • Women and children aged under 5 in LMICs • 5 literature reviews on agricultural interventions [111-115] • Interventions include: homestead food production systems, small livestock, dairy development and biofortification 	Insufficient evidence on improvements of children’s nutritional status (anthropometric indices and micronutrient status) found.
School garden programmes	Review on “ <i>the health and well-being impacts of school gardening</i> ” (2016) [116] <ul style="list-style-type: none"> • OECD countries (Australia, UK, USA and Portugal) • School-aged children up to 18 years old • 40 studies included, 5 RCTs, 14 non-randomised trials, 16 qualitative studies and 3 mixed methods • Interventions: cultivation of any kinds of plants, with in combination with educational, cooking or tasting activities 	Lack of evidence on anthropometric indices: <ul style="list-style-type: none"> • Two studies reported BMI to reduce obesity [117, 118], positive effect on reducing BMI in one study found [118] • Improved preferences towards fruits and vegetables but limited evidence on children’s fruit and vegetable intake

2.6.2 Nutrition-specific interventions

The 2013 *Lancet* series on “Maternal and child undernutrition” reviewed interventions which affect maternal and child undernutrition. In this comprehensive review, the potential effects of nutritional interventions for 34 countries that have 90% of stunted children were modelled [87]. The authors suggested that if populations can access ten evidence-based nutrition-specific interventions (including maternal nutrition during pregnancy, infant and young child feeding practices, micronutrient supplementation and management of acute malnutrition) at 90% coverage, the current total of deaths in children under the age of five could be reduced by 15%, while 20% of global stunting cases and 60% of wasting cases could be averted [87]. Given the modest effect on the reduction of stunting through these nutrition-specific interventions, the authors highlighted the importance of concurrently addressing the more distal and underlying contributing factors of undernutrition (e.g. WASH, education, food insecurity) to accelerate progress on reducing child undernutrition [87].

School feeding programmes are a type of nutritional interventions, yet they are often considered as social protection measures (conditional in-kind transfer) aiming to provide incentives for school enrolment. Evidence of nutritional benefits of school feeding programmes is scarce [87, 88]. A Cochrane review [90] of 18 relevant studies on the effectiveness of school feeding programmes in LMICs and high-income countries (HICs) reported beneficial effects on weight gain, but showed inconclusive results for height gain in schoolchildren.

Otherwise, micronutrient supplementation (provision of individual or mixtures of nutrients separately from the diet) and fortification (addition of one or more essential micronutrients to a food or drink) targeting schoolchildren have primarily focused on cognitive outcomes, showing positive impacts on school performance, but only small benefits on linear growth [94, 96].

Considering other nutrition-specific interventions, recent evidence from two meta-analyses suggest that anthelmintic treatments have a small benefit for children’s nutritional status, particularly when children were found infected with STH (detected by screening) [101, 102]. Of note, several studies also showed benefits on weight gain and growth among school-aged children after deworming in settings with chronic schistosomiasis [75, 119].

This sub-chapter highlights four main issues. Firstly, nutrition-specific interventions alone cannot sufficiently address wasting and stunting, and are not apt to change the conditions that contribute to child undernutrition [87]. Secondly, evidence on the effects of nutrition-specific interventions on school-aged children’s nutritional status, including school feeding programmes, is scarce and showed limited beneficial effects [90, 94, 96]. Thirdly, anthelmintic treatments for children are an important strategy to reduce the burden of disease, but the benefits for children’s nutritional status are small. Therefore and fourthly, there is growing understanding

that in order to achieve a sustained reduction of undernutrition in children, it is crucial to address its underlying determinants, combining multi-sectoral activities through nutrition-sensitive interventions [8, 87, 88].

2.6.3 WASH interventions

This section reviews WASH interventions targeting the three plausible biological mechanisms, i.e. STH infections, diarrhoea and EED, and their association with undernutrition. Considering the first mechanism, two recent systematic reviews conducted by Ziegelbauer and colleagues (2012) and Strunz and colleagues (2014) revealed that WASH access and safe practices are generally associated with lower odds of STH infections [67, 73]. There was no additional empirical evidence that links WASH improvements to reductions in STH infections and improvement in nutritional outcomes.

As to the second mechanism, it has long been shown that infectious and diarrhoeal diseases increase the risk of undernutrition in children [76, 120], and that WASH interventions reduce childhood diarrhoea [121, 122]. Yet, the potential effects that diarrhoea control programmes (i.e. WASH interventions) could have on undernutrition remains unresolved [79]. Dangour and colleagues (2013) reviewed 14 WASH intervention studies in LMICs and their effects on anthropometric outcomes in children aged under the age of 5 years [86]. Their findings revealed a very modest impact on stunting (with a greater effect on height growth in children aged under two) and no impact on underweight or wasting. The five cluster-RCTs generating this evidence investigated water disinfection and hygiene (handwashing with soap) interventions. However, this review did not identify any water supply or sanitation interventions. Moreover, these interventions were of rather short duration (nine to 12 months) [86]. More recent and subsequently published results of five trials have, however, described the effects of sanitation interventions on stunting among children aged under the age of 5 years; two of them reported a beneficial effect after implementing a community-led total sanitation (CLTS) programme in Mali and India [103, 104], while three did not find any effect [82, 105-107]. These three studies (conducted in Indonesia and in India) showing no effects of the sanitation interventions on stunting reported a very small uptake and compliance, with a low use of newly established toilets [82, 105-107].

With regards to the third mechanism, several studies have found that reductions in clinical presentations of diarrhoea were not associated with improvements in nutritional status, particularly stunting. These studies highlighted the importance of other pathways in improving nutritional outcomes other than diarrhoea [103, 123-125]. The hypothesis evolved that exposure to poor sanitation and hygiene causes EED [81, 126]; and that EED, rather than diarrhoea, is the primary pathway for poor sanitation and hygiene to lead to stunting [79]. Therefore, the

plausible question arising is whether improvements in WASH prevent or mitigate EED, and have a positive impact on growth in children. The current understanding of EED and its possible consequences for health is still limited [8, 126-128]. So far, evidence is constrained to observational studies [129-131]. Several large intervention studies are however ongoing [108-110], for example the SHINE - Sanitation, Hygiene, Infant Nutrition - project in Zimbabwe, investigating both the independent effect of WASH interventions and the combined effect of WASH and food supplementation interventions on childhood stunting [110]. These three trials include biological markers of EED to assess whether improvements in WASH can reduce EED and to what extent the effects of WASH on stunting are mediated by this subclinical condition [108-110].

Hence, current evidence suggests that WASH interventions reduce exposure to and infections with STH [67, 73], other enteric pathogens [79], and reduce childhood diarrhoea [121, 122]. Yet, key findings from literature reviews showed little effects of WASH interventions on childhood undernutrition, with scant evidence for school-aged children [86].

2.6.4 Agricultural interventions

The persistence of undernutrition as a global public health concern and the recent recognition that growing more food is necessary, but does not automatically translate into better nutrition and health, has led to the question of what kind of actions might be required for agriculture to contribute most effectively to improved nutritional outcomes [88, 132].

As part of the 2013 *Lancet* series on maternal and child undernutrition, Ruel and Aldermann (2013) [88] reviewed evidence of nutritional effects from agricultural interventions, particularly home garden and homestead food production systems, and the biofortification of staple crops¹. In brief, key findings of five selected literature reviews analysed (2001-2012) were largely consistent [111-115], showing a lack of evidence on the effectiveness of agricultural interventions on child nutrition (anthropometry or micronutrient status). For the few studies reporting a beneficial impact, the effects on child anthropometry were generally found to be small [88]. In one of the reviews included in the 2013 *Lancet* series, which was conducted by Girard and colleagues (2012) [114], it was noted that nutritional benefits are more likely when agricultural interventions include the production of foods rich not only in sources of micronutrients, but also in energy and/or protein. For example, studies on crop production strategies, including orange sweet potato [134], and animal production and dairy systems [135, 136], reported improved growth outcomes for children. In contrast, strategies promoting only improved varieties of fruits and vegetables, such as home gardening interventions [137], were

¹ Biofortification refers to the process by which the nutritional quality of food crops is improved through agronomic practices, conventional plant breeding, or modern biotechnology [133].

not found to impact child growth [114]. Ruel and Aldermann concluded that the lack of nutritional effects of agricultural interventions is less attributed to the specific type of intervention investigated, but mostly due to the poor quality evaluations (weak study designs with sample sizes often too small to draw on conclusions) [88].

Of note, school gardens are cultivated areas around or near schools, tended partly by students with the purpose of producing and facilitating access to fruits and vegetables in school-based settings [138]. Hence, school gardens can be considered as a small-scale agricultural intervention, which in combination with educational and awareness raising activities, also aim at increasing knowledge on healthy foods and promote increased vegetable and fruit consumption [138]. Evaluations of school garden programmes are thus far mostly restrained to HICs. Findings from a recent systematic review (2016) on the impacts of school garden programmes (in USA, Australia, Portugal, and UK) showed positive effects on schoolchildren's preferences for vegetables and fruits, but limited impacts on their vegetable and fruit intake [116]. Only two studies reported anthropometric measures [117, 118], and only one statistically significant difference in BMI was reported from a non-randomised controlled study, with the objective of reducing obesity among schoolchildren [118]. Hence, school-gardening interventions are promising approaches to improve health outcomes of schoolchildren, but evidence is currently restrained to HICs and is of limited scientific quality (i.e. non-randomised studies, self-reported outcomes measures) [116].

Two major issues emerge from this sub-chapter. First, there is little evidence of the nutritional benefits from agricultural interventions, including school gardening programmes, and second; evidence on the nutritional effects of agricultural interventions is particularly scarce for school-aged children [88, 116].

2.7 Background of the PhD thesis

2.7.1 Identified research needs

Schools are an ideal entry point for linking agriculture, nutrition and WASH interventions [139]. Apart from being an obvious place to educate children on healthy diets, schools can promote practical and positive changes in personal hygiene, nutrition and health by: (1) increasing food availability and diversity with school gardens [138]; (2) offering well-balanced and nutritious meals through a school feeding programme (in which parts of the garden produce could be used) [140]; and (3) promoting handwashing with soap and safe sanitary behaviours [139]. In addition, given that schoolchildren are the main reservoir of worm loads in a population [88], mass anthelmintic drugs can be included as part of a larger school health programme [140].

However, as highlighted in previous chapters, there is a lack of, or inconclusive evidence on the effects of nutritional (including school feeding), WASH, and agricultural (including school gardens) interventions on nutritional outcomes of schoolchildren. There is also insufficient evidence of combined approaches across the nutrition, health, agriculture, education, and WASH sectors addressing proximal and underlying determinants of undernutrition in children [88, 141, 142].

To address these research gaps, this PhD thesis examines the effects of combined school garden, nutrition, health, education and WASH interventions with a focus on schoolchildren's nutritional and health outcomes. These interventions are summarized in this PhD thesis as complementary (i) school garden, (ii) nutrition, and (iii) WASH interventions. The PhD research is conducted in the frame of a school garden programme, entitled "Vegetables go to School: Improving Nutrition through Agricultural Diversification" (VgtS) (Figure 2.2).

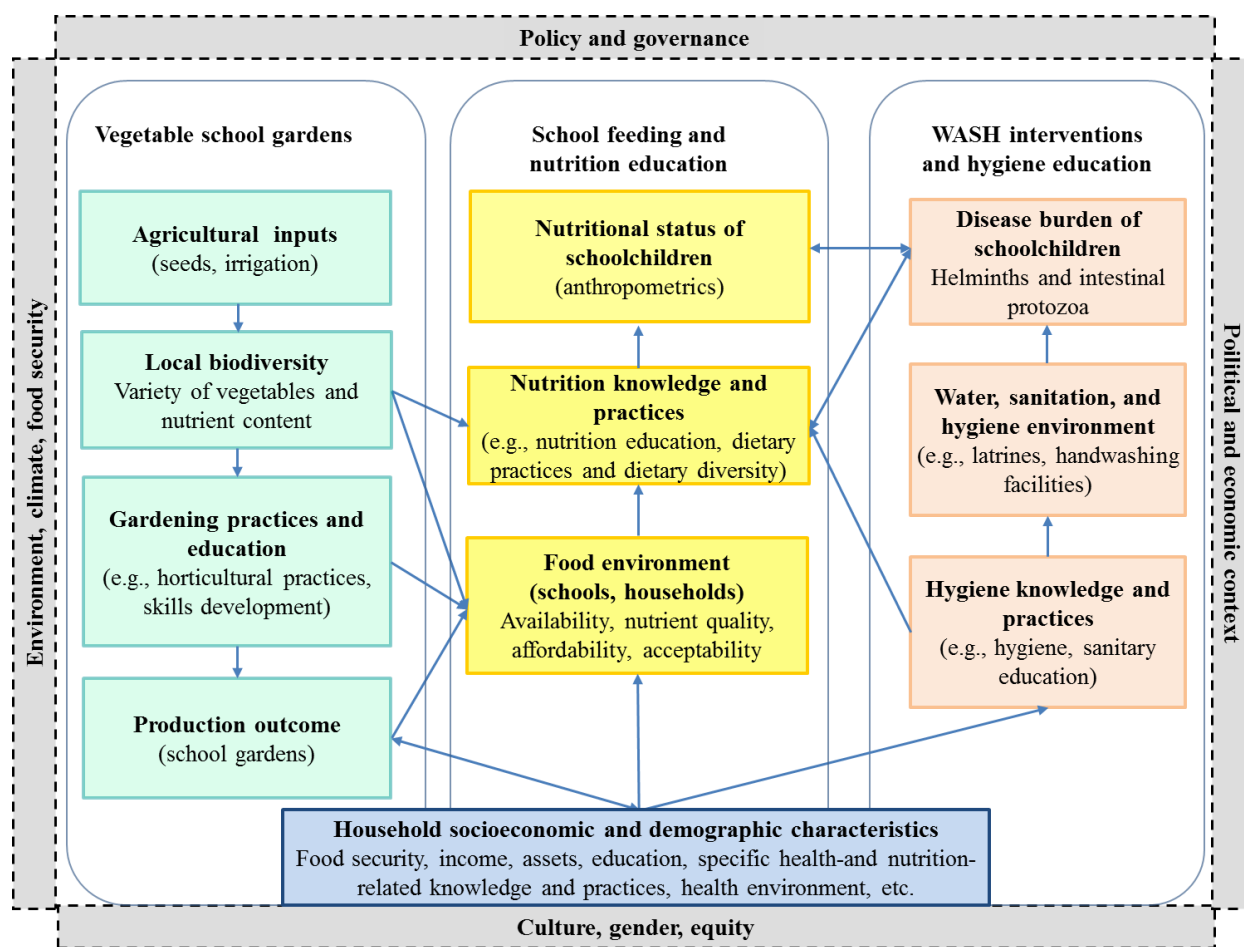


Figure 2.2: Conceptual framework on the potential contributions of school garden, nutrition and WASH interventions to address nutritional and health outcomes of schoolchildren.

Source: the framework is adapted from Hawkes et al. (2012) and Turner et al. (2013), incorporating the “Vegetables go to School” project interventions.

2.7.2 Collaborative framework, the “Vegetables go to School: Improving Nutrition through Agricultural Diversification” project

This PhD thesis is part of the VgtS project which is funded by the Swiss Agency for Development and Cooperation (SDC). The VgtS project was launched in 2012 in six target countries (Bhutan, Burkina Faso, Indonesia, Nepal, the Philippines and Tanzania) and was implemented by country teams composed of members of different ministries (i.e., education, agriculture and health), in collaboration with the World Vegetable Center (AVRDC) in Taiwan, the University of Freiburg (ALU) in Germany and the Swiss Tropical and Public Health Institute (Swiss TPH) in Switzerland as academic partners. This joint operational research project pursued the following three objectives:

- (1) to improve capacity in the target countries to successfully implement school gardens;
- (2) to implement school gardens and encourage consumption of a diversity of vegetables by schoolchildren; and

(3)to increase knowledge on how school gardens linked to complementary WASH interventions contribute to improved nutrition and health of schoolchildren.

This PhD work is embedded in the third objective, which is described in detail in Chapter 4.

The VgtS project was funded over a period of five years (from 2012 to 2017) in two distinct phases. The first research-oriented phase (from 2012 to 2016) intended to test the feasibility and to pilot school gardening programmes in the 6 target countries. The second phase, from July 2016 until June 2017, aims to develop a policy roadmap/framework (policy briefs) based on the research findings in order to sustain and scale up the interventions in each of the target countries.

2.8 References

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3 Goal and objectives of the thesis

The overarching goal of this PhD thesis is to deepen the understanding of undernutrition, intestinal parasitic infections and associated risk factors in schoolchildren (8-14 years) in primarily rural schools of two regions in Burkina Faso, and to generate evidence on the effects of complementary school garden, nutrition and WASH interventions on schoolchildren's nutrition and health status.

Three particular objectives are linked to this overarching goal:

Objective 1: To investigate schoolchildren's nutritional status and associated risk factors at a baseline cross-sectional survey before implementing complementary school garden, nutrition and WASH interventions in the two VqtS project regions. To determine the prevalence of undernutrition, in particular stunting, thinness and underweight, among schoolchildren (8-14 years) and its association with intestinal parasitic infections, anaemia, socioeconomic and demographic factors, and health knowledge, attitudes and practices.

Objective 2: To examine the prevalence of intestinal parasitic infections in schoolchildren and its association with household- and school-level WASH conditions at baseline of the implementation of complementary school garden, nutrition and WASH interventions in the two VqtS project regions. To assess the prevalence of helminths and intestinal protozoa among schoolchildren (8-14 years) and to investigate the association with children's hygiene practices, household WASH conditions and school-, household-, and community-level drinking water contamination.

Objective 3: To elucidate whether complementary school garden, nutrition and WASH interventions reduce the prevalence of intestinal parasitic infections and improve schoolchildren's nutritional status. This objective includes the investigation of the effects of the integrated intervention package in relation to aforementioned key parameters of schoolchildren's nutritional and health status, health knowledge and practices one year after a baseline cross-sectional survey.

4 Complementary school garden, nutrition, water, sanitation and hygiene interventions to improve children's nutrition and health status in Burkina Faso and Nepal: a study protocol

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4.1 Abstract

Background: Malnutrition and intestinal parasitic infections are common among children in Burkina Faso and Nepal. However, specific health-related data in school-aged children in these two countries are scarce. In the frame of a larger multi-stakeholder project entitled “Vegetables go to School: Improving Nutrition through Agricultural Diversification” (VgtS), a study has been designed with the objectives to: (i) describe schoolchildren’s health status in Burkina Faso and Nepal; and to (ii) provide an evidence-base for programme decisions on the relevance of complementary school garden, nutrition, water, sanitation and hygiene (WASH) interventions.

Methods/Design: The studies will be conducted in the Centre Ouest and the Plateau Central regions of Burkina Faso and the Dolakha and Ramechhap districts of Nepal. Data will be collected and combined at the level of schools, children and their households. A range of indicators will be used to examine nutritional status, intestinal parasitic infections and WASH conditions in 24 schools among 1,144 children aged 8-14 years at baseline and a 1-year follow-up. The studies are designed as cluster randomised trials and the schools will be assigned to two core study arms: (i) the ‘complementary school garden, nutrition and WASH intervention’ arm; and the (ii) ‘control’ arm with no interventions. Children will be subjected to parasitological examinations using stool and urine samples and to quality-controlled anthropometric and haemoglobin measurements. Drinking water will be assessed for contamination with coliform bacteria and faecal streptococci. A questionnaire survey on nutritional and health knowledge, attitudes and practices (KAP) will be administered to children and their caregivers, also assessing socioeconomic, food-security and WASH conditions at household level. Focus group and key-informant interviews on children’s nutrition and hygiene perceptions and behaviours will be conducted with their caregivers and school personnel.

Discussion: The studies will contribute to fill a data gap on school-aged children in Burkina Faso and Nepal. The data collected will also serve to inform the design of school-based interventions and will contribute to deepen the understanding of potential effects of these interventions to improve schoolchildren’s health in resource-constrained settings. Key findings will be used to provide guidance for the implementation of health policies at the school level in Burkina Faso and Nepal.

Trial registration: ISRCTN17968589 (date assigned: 17 July 2015)

Keywords: Burkina Faso; Malnutrition; Nepal; Parasitic infections; School-aged children; Study protocol; Water, sanitation, and hygiene (WASH)

4.2 Background

Malnutrition, intestinal parasitic infections and diarrhoeal diseases are common public health problems in children in low- and middle-income countries (LMIC) [1-8]. In many countries, Demographic and Health Surveys (DHS) and national nutrition surveillance systems have been measuring height and weight of children below the age of 5 years, starting in the early 1990s. However, there is a paucity of anthropometric data for school-aged children (5-14 years) [9-11]. Additionally, there are currently no estimates neither for school-aged children, nor the entire population, on the global burden of diseases from polyparasitism of intestinal parasitic infections caused by helminths and intestinal protozoa [7]. Data on the burden of disease caused by intestinal protozoa is scarce, partially explained by the lack of diagnosis at the periphery [12-15]. Although no estimates on the burden of diseases caused by helminth infections for school-aged children exist, an estimate for the burden of disease of sub-groups of helminth infections is available (e.g. schistosomiasis and soil-transmitted helminthiasis) [4, 7, 16]. Estimates from the Global Atlas of Helminth Infection (GAHI; <http://www.thiswormyworld.org/>) showed that, in 2010, 1.01 billion school-aged children lived in areas where prevalence of any soil-transmitted helminth (STH) was above 20% [7]. Furthermore, in 2013, diarrhoeal diseases were responsible for an estimated 7% of deaths in school-aged children in LMICs, with more than 96% attributable to unsafe water, inadequate sanitation and hygiene (WASH) [4, 5].

Burkina Faso and Nepal are both low-income countries that face an array of similar health challenges. Whilst health data among under 5-year-old children, such as nutritional indicators, anaemia or *Plasmodium* prevalence, are collected during national surveys, statistics on school-aged children in these two countries are typically unavailable [17, 18]. Malnutrition, anaemia and diarrhoeal diseases are highly prevalent in under 5-year-old children. Indeed, according to the 2010 and 2011 DHS in Burkina Faso and Nepal, respectively, 35% and 41% of children were stunted; almost 15% of children in both countries reported diarrhoea 2 weeks prior to a DHS; and 88% of the children in Burkina Faso and 46% in Nepal were anaemic [17, 18]. Both countries also face considerable ill-health due to inadequate WASH conditions. For example, according to data from the 2013 Global Burden of Disease Study (GBD) and the World Health Organization (WHO)/United Nations Children's Fund (UNICEF) 'Joint Monitoring Programme for Water Supply and Sanitation', 7% and 8% of deaths in children aged 5-14 years in Burkina Faso and Nepal, respectively, were caused by diarrhoeal diseases, with over 96% in both countries attributed to inadequate WASH conditions as primary risk factor [4, 19]. Table 4.1 provides an

overview of selected health and WASH indicators in Burkina Faso and Nepal for the years 2010 to 2013.

Table 4.1: Overview of health and WASH indicators of Burkina Faso and Nepal

Indicator	Burkina Faso	Nepal
Health	<i>DHS 2010</i>	<i>DHS 2011</i>
Stunting (<5 years)	35%	41%
Wasting (<5 years)	16%	11%
Underweight (<5 years)	26%	29%
Diarrhoea (<5 years)	15%	14%
Anaemia (<5 years)	88%	46%
Mortality (a) and morbidity [DALYs] (b)	<i>GBD 2013</i>	<i>GBD 2013</i>
Diarrhoeal diseases (5 to 14 years old)	7% (a), 5% (b)	8% (a), 4% (b)
Iron-deficiency anaemia (5 to 14 years old)	1% (a), 6% (b)	1% (a), 15% (b)
Intestinal infectious diseases (5 to 14 years old)	4% (a), 2% (b)	10% (a), 4% (b)
Water, sanitation and hygiene (WASH)	<i>DHS 2010 (a) and WHO Progress Report on Drinking-Water and Sanitation 2012 (b)</i>	<i>DHS 2011 (a) and WHO Progress Report on Drinking-Water and Sanitation 2012 (b)</i>
Improved latrines	15% (a), 19% (b)	38% (a), 37% (b)
Non-improved latrines	6% (a), 17% (b)	43% (a), 6% (b)
Open defaecation (bush/field, no latrines)	62% (a), 57% (b)	36% (a), 40% (b)
Soap and water for hand washing available	14% (a)	48% (a)

(a) Mortality rate among children aged 5 to 14 years old

(b) Disability-adjusted life year (DALYs) as indicator of morbidity among children aged 5 to 14 years

Malnutrition, intestinal parasitic infections and inadequate WASH conditions are intricately linked. First, inadequate WASH conditions are important risk factors for both, malnutrition and intestinal parasitic infections [2, 4, 15, 20, 21]. The pathogenic agents associated with poor WASH conditions are viral pathogens, bacterial pathogens, protozoan cysts or oocysts and helminth eggs found in faeces and transmitted through the faecal-oral pathway and can lead to diarrhoea and undernutrition, whereby exposure to one increases vulnerability to the other [22-27]. Second, malnutrition can render a child more susceptible to infection. An inadequate dietary intake leads to weight loss, weakened immunity, invasion by pathogens and mucosal damage, and impaired growth and development in children [28-30]. Third, parasitic infections also contribute to growth stunting by causing a decline in food intake (loss of appetite), diarrhoea, malabsorption and/or an increase in nutrient wastage for the immune response, all of which lead to nutrient losses and further damage to the defence mechanisms, causing a vicious cycle [28-30]. Moreover, it is well documented that infections with intestinal parasites may cause internal

bleeding, leading to a loss of iron and anaemia [31, 32]. Intestinal parasitic infections can go unnoticed for years due to delayed onset of symptoms, which can exacerbate the effects on malnutrition, and hence compromise the development of their cognitive abilities in their formative years [30]. It is therefore crucial to consider the strong interlinkages of malnutrition, parasitic infections, diarrhoeal diseases and WASH for preventive actions and sustainable programmes.

“Vegetables go to School: Improving Nutrition through Agricultural Diversification”

A multi-country and multi-stakeholder project entitled “Vegetables go to School: Improving Nutrition through Agricultural Diversification” (VgtS in short) was developed and is funded by the Swiss Agency for Development and Cooperation (SDC) to address schoolchildren’s nutrition in an interdisciplinary approach through the implementation of school vegetable gardens and other school-based health, nutrition and environmental interventions. The VgtS project was launched in 2012 in six target countries (Bhutan, Burkina Faso, Indonesia, Nepal, the Philippines and Tanzania) and is implemented by country teams composed of members of different ministries, (i.e. education, agriculture and health), in collaboration with the World Vegetable Center (AVRDC; headquartered in Taiwan), the University of Freiburg in Germany and the Swiss Tropical and Public Health Institute (Swiss TPH) in Switzerland as academic partners.

The objectives of the VgtS project are threefold: (i) to encourage agricultural production at the unit of the school and to increase the availability and access to a wide diversity of vegetables in order to favour a balanced and nutritious diet; (ii) to link the school garden to an educational programme that covers basic topics of agriculture, nutrition and WASH (overall project approach in all the countries); and (iii) to link the school garden programme to complementary nutrition and WASH interventions. In this context, the VgtS project embeds two intervention studies in Burkina Faso and Nepal, which include intervention schools benefitting from a complementary school garden, nutrition and WASH intervention package and control schools without any intervention. Here, we present the research protocol for the studies in Burkina Faso and Nepal.

4.3 Methods/design

Goal

The overarching goal of the studies within the frame of the VgtS project in Burkina Faso and Nepal is to address the current data gap on schoolchildren (aged 8-14 years) and to assess the effects of complementary school garden, nutrition and WASH interventions on schoolchildren's health status, as assessed by a baseline and a 1-year follow-up survey through a range of previously identified nutrition, WASH and health indicators (Table 4.2).

Table 4.2: Selected indicators for the two studies in Burkina Faso and Nepal

Objective	Indicator	Methods and tools	Survey module
<i>Individual level of child</i>			
To assess schoolchildren's nutritional status at baseline and follow-up	Nutritional status (BMIZ, HAZ, WAZ and clinical signs of malnutrition)	Digital scale, height measuring board and clinical examination	Nutritional survey (module 1)
To assess the prevalence of anaemia in schoolchildren at baseline and follow-up	Anaemia based on haemoglobin levels < 11.5 g/dl for children aged 7 to 11 years and < 12 g/dl for those aged 12 to 14 years	HemoCue Hb 201 ⁺	Nutritional survey (module 1)
To assess the prevalence of intestinal parasitic infections in schoolchildren at baseline and follow-up	Presence and intensity of intestinal and urinary parasitic infections	Kato-Katz and formalin-ether concentration method for stool samples and centrifugation method for urine samples	Parasitological survey (module 2)
To assess schoolchildren's nutrition and health knowledge, attitudes and practices (KAP) at baseline and follow-up	KAP related to nutrition and health	Questionnaire survey with schoolchildren Focus group discussions with schoolchildren In-depth interviews with school directors and teachers	Children's health KAP (module 3)
<i>Environmental indicators</i>			
To assess drinking water quality of children's drinking water recipients at baseline and follow-up	Presence of thermotolerant coliform bacteria and faecal streptococci	Portable DelAgua field kit and RAPID E. COLI 2 AGAR (coliform bacteria, <i>Escherichia coli</i>) and Bile Esculine Azide AGAR (faecal streptococci) tests	Water quality testing (module 4)
<i>Household level</i>			
<i>Demographic and socio-economic data</i>			
To assess basic household socio-demographic and economic characteristics at baseline and follow-up	Caregiver's age, educational level, occupational status, assets, food security	Household questionnaire	Household questionnaire survey (module 5)
<i>Household nutrition and health -related knowledge, attitudes and practices data</i>			
To assess caregivers' nutrition and health knowledge, attitudes and practices at baseline and follow-up	Caregiver's knowledge, attitudes and practices related to nutrition and health	Household questionnaire Focus group discussions with schoolchildren's caregivers	Household questionnaire survey (module 5) Caregivers' health KAP (module 6)
<i>Socio-environmental conditions data</i>			
To assess household WASH conditions at baseline and follow-up	Drinking water source and distance to it, water storage, improved/non-improved latrine, location of kitchen, available hand washing facilities and soap, presence of domestic animals	Household living condition and information related to hygiene Direct observation	Household questionnaire survey (module 5)
<i>Environmental indicators</i>			
To assess drinking water quality at schoolchildren's households at baseline and follow-up	Presence of thermotolerant coliform bacteria and faecal streptococci	Portable DelAgua field kit and RAPID E. COLI 2 AGAR (coliform bacteria, <i>Escherichia coli</i>) and Bile Esculine Azide AGAR (faecal streptococci) tests	Water quality testing (module 4)
<i>School and community level</i>			
<i>Socio-environmental conditions data</i>			
To assess the WASH conditions at schools at baseline and follow-up	Available drinking water, available improved/non-improved toilet/latrine, available hand washing facilities and soap	In-depth interviews with school directors and teachers Direct observation	WASH survey (module 7)
<i>Environmental indicators</i>			
To assess drinking water quality at schools and	Presence of thermotolerant coliform bacteria and faecal	Portable DelAgua field kit and RAPID E. COLI 2 AGAR (coliform	Water quality testing (module 4)

community sources at baseline and follow-up	streptococci	bacteria, <i>Escherichia coli</i>) and Bile Esculine Azide AGAR (faecal streptococci) tests	
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Study sites and school selection

The studies will be conducted in Burkina Faso and Nepal. The study sites are located within the VgtS project sites, which were selected by the local VgtS country teams, following a set of criteria: (i) accessibility from the capital; (ii) availability of land for the school garden and continuous access to water at schools; (iii) coeducation of boys and girls in public schools; and (iv) willingness of the school principals and the community to participate.

In both countries, the study will be implemented in two different regions. In Burkina Faso, these are the Centre Ouest and the Plateau Central regions, both located in proximity to the capital Ouagadougou (30-180 km). The study sites in Nepal are the Dolakha and Ramechhap districts in the eastern part of the country, both located in proximity of the district headquarters Charikot (133 km) and Manthali (131 km), respectively.

The selection of the schools participating in the two studies is based on a three-stage sampling procedure of schools within the overall VgtS project sites. In a first step, about 100 schools fulfilling the aforementioned eligibility criteria were selected. In a second step, from these 100 schools, a sample of 30 schools were randomly chosen to be included in the VgtS school garden implementation and were randomly allocated to three groups, which receive the school vegetable garden interventions in 2014, 2015 and 2016, respectively. In a third step, out of the 30 VgtS project schools, a total of eight schools in Burkina Faso and 16 schools in Nepal were randomly selected to accommodate the sampling needs of the two complementary and slightly different study designs of Burkina Faso and Nepal (Figure 4.1).

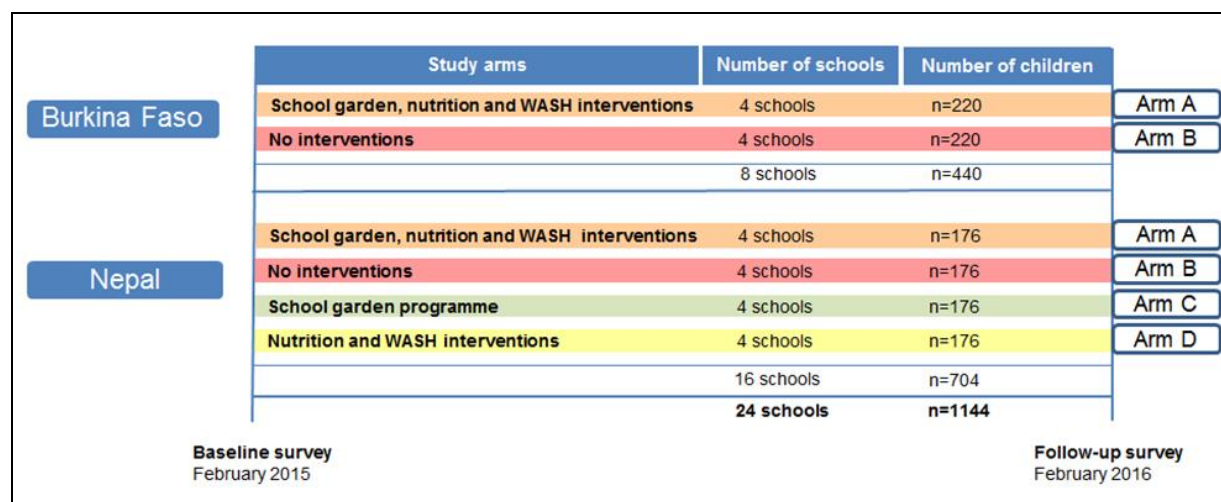


Figure 4.1: Study design for Burkina Faso and Nepal

Study design

The two studies in Burkina Faso and Nepal are designed as cluster randomised trials with an equal number of schools randomly allocated to two core study arms (A, B) and with a cohort of children followed in two consecutive surveys, at baseline and 1-year follow-up. Two additional study arms are included in Nepal (C, D). The four study arms are designed as follows:

- arm A: school garden programme and complementary nutrition and WASH interventions;
- arm B: no interventions, i.e. controls;
- arm C: school garden programme without nutrition and WASH interventions; and
- arm D: nutrition and WASH interventions without the school garden programme.

Each arm comprises four schools. Figure 4.1 shows the study design with the different study arms for Burkina Faso and Nepal. In both countries, schools of arm A will receive the complementary school garden, nutrition and WASH intervention package starting in March 2015. In Nepal, the interventions from arms C and D will be implemented over the same period. In both countries, the control schools of arm B will benefit from the school garden intervention in the year following the interventions.

Sample size

Two separate sample size calculations were conducted for the two study designs of Burkina Faso and Nepal. For the sample size calculation of the study in Burkina Faso, the prevalence of intestinal parasitic infection in children aged 8-14 years was selected as the primary outcome in the comparison between high- and low-risk of intestinal parasitic infection in children. The power calculation was based on the assumption of:

- an average intestinal protozoa and helminth infection rate across schools of 40% [33];
- a coefficient of variation of 10% across schools; and
- a proportion of high risk children of 25%.

A Monte Carlo simulation with 5,000 iterations shows that a total of 400 children from eight schools (i.e. 50 children per school) would provide 85% power for detecting a significant difference in infection rates between high- and low-risk children at the usual level of 5% under these assumptions and for a true odds ratio of 2. Recruitment will be increased by 10% to account for drop-outs or non-participation, which leads to an optimal sample size of at least 440 children.

The sample size calculation for the study in Nepal was also based on the prevalence of intestinal parasitic infections in children aged 8-14 years as the primary outcome. The power calculation was based on the assumption of:

- the prevalence rate of intestinal protozoa and helminth infection is 30% [34] and this rate will remain in steady state in the absence of any intervention;
- probability of new intestinal protozoa and helminth infection at the 1-year follow-up will be reduced by at least 10% through the implementation of the complementary nutrition and WASH intervention package; and
- a coefficient of variation of 10% across schools.

A Monte Carlo simulation with 5,000 iterations shows that a total of 640 schoolchildren from 16 schools (i.e. 40 children per school) would provide 80% of power for detecting a significant difference in infection rates between the four study arms. Recruitment will be increased by 10% accounting for drop-outs and non-participants, which leads to an optimal sample size of at least 704 children.

Eligibility and selection criteria of study participants

In both Burkina Faso and Nepal, children enrolled in school are eligible to participate in the baseline survey if they are between 8 and 14 years old, have signed a written informed consent by their parents, guardians or teachers, and themselves assented orally.

Data collection procedures

Four weeks prior to the study, district and village authorities, school directors and children's parents/guardians will be informed about the forthcoming survey activities by the local survey team. They will be re-informed about the purpose and procedures of the study shortly before the start of the survey activities. Written informed consent (signed or fingerprint) will be obtained from children's parents or legal guardians, whilst children will assent orally. It will be emphasised that participation is voluntary and that children and parents/guardians can withdraw anytime without further obligation.

In each school, a random selection of children aged 8-14 years will be enrolled until at least 55 in Burkina Faso and 44 in Nepal are reached. Moreover, at the follow-up survey, the same children will be re-assessed, who by then will be 9-15 years old. Each child will be attributed a unique identification code (ID) for the different assessments at the onset of the study. A separate household ID connected to the schoolchild's personal ID will be given to children's

households in order to link children's clinical data and nutritional and health knowledge, attitudes and practices (KAP) with the household characteristics. Children will thereafter be invited to provide stool and urine samples, to take anthropometric and haemoglobin (Hb) measurements and to participate in the KAP survey. In Burkina Faso, stool and urine samples will be collected on two consecutive days. In Nepal, a single stool sample will be collected, while urine samples will not be collected as urogenital schistosomiasis is not endemic. Infected, anaemic or undernourished children in all schools will be subjected to clinical, parasitological and nutritional examinations, and will be treated according to national policies.

After these assessments with children at the schools, the same enumerators in Burkina Faso will visit children's households and will invite children's caregivers to respond to a household questionnaire during the two survey days. In Nepal, due to the scattered locations and geographical constraints, additional enumerators will visit the children's households during the same survey period. In both countries, trained and experienced enumerators will conduct the questionnaire surveys in local languages.

Collection of stool and urine samples

The sampled children at the schools will be asked to provide a fresh mid-morning, post-exercise stool sample. The stool samples will be processed and analysed each day (at mid-day the latest) by experienced laboratory technicians and medical microbiologists as follows: first, stool samples will be visually examined for macroscopic appearance of adult worms, also checking the stool consistency and the presence of blood and mucus. Second, a single Kato-Katz thick smear, using 41.7 mg templates, will be prepared on a slide and examined under a microscope for the presence of eggs of *Schistosoma mansoni*, hookworm, *Ascaris lumbricoides*, *Trichuris trichiura* and *Hymenolepis nana*, adhering to standard operating procedures [35, 36].

Third, a formalin-ether concentration technique will be used to enhance sensitivity for the diagnosis of helminths and to detect intestinal protozoa (*Blastocystis hominis*, *Chilomastix mesnili*, *Endolimax nana*, *Entamoeba coli*, *Entamoeba histolytica/Entamoeba dispar*, *Entamoeba hartmanni*, *Giardia intestinalis* and *Iodamoeba bütschlii*) [37]. Approximately 1-2 g of stool will be placed in 15 ml Falcon tubes filled with 10 ml of 5% formalin and will be examined by experienced laboratory technicians for the presence of helminths and intestinal protozoa, using an ether-concentration technique, adhering to an SOP [38]. Additionally, in Nepal, 20 mg of stool will be prepared on a single slide with the saline wet mount concentration for the microscopic detection of the same intestinal protozoa and helminths, according to SOPs [39,

40]. Furthermore, the intensity of infection will be calculated as the number of eggs per 1 g of stool (EPG) and categorised according to the WHO standard classification [41].

In Burkina Faso, children will also be asked to provide fresh, mid-morning and post-exercise urine samples, collected at the same time as the stool samples. Urine samples will be analysed for microhaematuria (biochemical marker and proxy for *Schistosoma haematobium*), using reagent strips (Hemastix; Siemens Healthcare Diagnostics GmbH; Eschborn, Germany) [42], and for the presence and number of *S. haematobium* eggs in 10 ml of urine using a urine centrifugation technique and microscopy [43]. *S. haematobium* infection will be grouped into light (< 50 eggs/10 ml of urine) and heavy (\geq 50 eggs/10 ml of urine) [44].

In order to achieve a higher sensitivity in diagnostics, in selected schoolchildren stool and urine samples will be obtained on two consecutive days in Burkina Faso [45, 46]. For quality control, 10% of all processed stool samples will be re-read under a microscope by independent laboratories [47]. Slides identified with discrepant results will be re-examined by the Institut de Recherches en Sciences de la Santé (IRSS) laboratory and Kirnetar Health Centre team until agreement has been reached.

Collection of anthropometric indicators and measuring Hb levels

Selected schoolchildren will be subjected to anthropometric measurements according to SOPs, as described by WHO, using a digital scale and a height measuring board with a precision of 0.1 kg and 0.1 cm, respectively [48]. Individual z-score will be computed using the new WHO growth reference values for children and adolescents [49]. The nutritional status of schoolchildren will be classified as follows: a z-score within the interval of -3 standard deviation (SD) < z < -2 SD will be used to classify body-mass-index-for-age (BMIZ, thinness), height-for-age (HAZ, stunting) and weight-for-age (WAZ, underweight) as moderate undernutrition, and a z-score < -3 SD to define severe undernutrition. WAZ will only be used for children aged 8-10 years as reference data are not available beyond the age of 10 years [49]. Overweight will be classified as BMIZ >1.0 SD and obesity as BMIZ >2.0 SD [50].

The Hb level will be measured to determine anaemia prevalence. A finger-prick capillary blood sample will be taken, and Hb concentration measured using a HemoCue® 201+ testing device (HemoCue Hb 201+ System; HemoCue AB, Ängelholm, Sweden). Age-specific criteria will be used to identify anaemic children: Hb <11.5 g/dl for children aged 8-11 years and Hb <12 g/dl for children aged 12-14 years [51].

Additionally in Nepal, trained health care professionals will conduct clinical examinations for detecting clinical signs of nutritional deficiencies (e.g. dermatitis, bitot's spot, dry and infected cornea, oedema, enlargement of liver, loss of peripheral sensation, angular stomatitis, pale conjunctiva, red inflamed tongue, swelling of the thyroid gland and bowed legs) [52].

Drinking water quality assessment

In Burkina Faso, drinking water samples will be collected in sterile 250 ml bottles at the selected schools and community sources, children's households and from their drinking water recipients to assess drinking water quality at source and point of use. Water samples will be randomly taken in 20% of participating children's households and in five community sources per study site (always including the school source). Water samples from children's drinking water recipients brought to school will be randomly collected in 30% of the children. Before analysis, the water samples collected will be preserved in cool boxes at 4°C, and transferred to a nearby laboratory. The water samples will be analysed by membrane filtration for the presence/absence (PA) of thermotolerant faecal coliforms (TTC) as colony forming units per 100 ml of water (CFU/100 ml). Furthermore, *E. coli* and faecal streptococci as indicators for faecal contamination will be assessed by the use of the RAPID E. COLI 2 AGAR (coliform bacteria and *E. coli*; Bio-Rad Laboratories, Hercules, USA) and the Bile Esculine Azide AGAR (faecal streptococci; Bio-Rad Laboratories, Hercules, USA) tests according to WHO drinking water standards [53].

In Nepal, drinking water samples will be collected in 250 ml sterile bottles from the school drinking water source and children's drinking water recipients, household and community water sources. Water samples will be collected at every school (n=16) and every child's household (n=440). For the community sources, one water sample per study site will be taken from the principal water distribution channel of the community source (n=16). The water samples will be analysed *in situ* at the schools and households for turbidity, pH and chlorine residual using the *DelAgua* kit (Oxfam-DelAgua; Guildford, UK) using readily available SOPs [54]. If the concentration of free chlorine residual is greater than 0.2 mg/l (0.2 ppm) and the turbidity less than 5 turbidity units, the water samples will not be analysed for TTC [54]. If the results do not meet these criteria, water samples will be transported in cool boxes to the laboratory in Kirnetar Health Centre and stored in a refrigerator at 4°C before analysis using the *DelAgua* kit. The water samples will be tested for CFU/100 ml according to WHO drinking water standards [53].

Quality control will be conducted with 10% of all water samples collected by independent laboratories.

Questionnaire survey with schoolchildren and their caregivers

The KAP survey was established with the guidelines and KAP manual provided by FAO, using standardised questions and amendments by the Swiss TPH research team [55]. Children's caregivers will also be invited to respond to a questionnaire investigating sociodemographic, economic, health and food security issues. The questionnaire surveys for children and their caregivers will be tablet-based using the Open Data Kit software [56].

Focus group discussions and in-depth interviews

Focus group discussions (FGDs) will be conducted with 6 to 10 randomly selected caregivers from sampled children in each school to better understand the caregivers' perceptions on nutrition and health. The school director and teachers will be interviewed with a semi-structured questionnaire to record characteristics of children's health challenges, and to discuss children's nutrition and health education in the curricula, school health activities, school water and sanitary installations and, if existing, the school feeding programmes.

Data management and analysis

Quantitative data from stool and urine examinations, anthropometrics and Hb measurements will be entered in Microsoft Excel 2010 (Microsoft; Redmond, USA). A double data entry system will be used to ensure data quality. Data will be evaluated for discrepancies and validated after removing inconsistencies. The z-score values for height-, weight- and BMI-for-age relative to the WHO 2007 reference will be calculated using WHO AnthroPlus (WHO; Geneva, Switzerland). Statistical analyses will be carried out with Stata version 13 (StataCorp; College Station, USA). Analysis of baseline data will be conducted to describe the prevalence of malnutrition, intestinal parasitic infections, WASH conditions, KAP and basic socioeconomic characteristics. Logistic regression models will be employed to estimate associations of malnutrition, intestinal parasitic infections and anaemia with risk factors.

FGDs and in-depth interviews (IDIs) will be transcribed, translated into English by bilingual research assistants and entered as Microsoft Word documents into MAXQDA version 11 (VERBI GmbH 2012; Berlin, Germany) for data coding and analysis. Main themes will be identified and coded in order to categorise explanations and descriptions of nutritional and health related perceptions and issues.

Longitudinal analysis will be conducted to evaluate the intervention effects of the complementary interventions under study. The results from the different study arms will be compared at the end of the 1-year intervention period.

Data storage and handling

All data files will be stored in a secure server at Swiss TPH. ID codes and name-linked information on participants will remain confidential and will be used only to communicate clinical results to participants for their respective treatments.

Ethical considerations

The two research protocols for Burkina Faso and Nepal were reviewed by (i) the Institutional Research Commission of Swiss TPH (reference number FK 116; date of approval 30 October 2014); (ii) the “Ethikkommission Nordwest- und Zentralschweiz” (EKNZ) in Switzerland for the Nepal study protocol (reference no. UBE-15/02; date of approval 12 January 2015); (iii) the EKNZ in Switzerland for the Burkina Faso study protocol (reference no. 2014-161; date of approval 19 January 2015); (iv) the “Comité d’Ethique pour la Recherche en Santé, Ministère de la Recherche Scientifique et de l’Innovation, et Ministère de la Santé” in Burkina Faso (reference no. 2014-5-058; date of approval 20 May 2014); (v) the “Institutional Review Committee of Kathmandu University School of Medical Sciences, Dhulikhel Hospital”, Nepal (reference no. 86/14, date of approval 24 August 2014); and the (vi) Institutional Review Committee, Health Research Council, Nepal (reference no. 565; date of approval 11 November 2014).

The two studies have been registered under a single trial registration number at the International Standard Randomised Controlled Trial Number Register ISRCTN17968589 (date assigned: 17 July 2015; <http://www.isrctn.com/ISRCTN17968589>).

4.4 Discussion

Malnutrition and intestinal parasitic infections are a major burden on children's health globally and particularly in LMIC, including Burkina Faso and Nepal. Inadequate WASH conditions play an important role in the high burden of communicable diseases [21, 57]. The morbidity due to malnutrition, intestinal parasitic infections and diarrhoeal diseases in Burkina Faso and Nepal continue to be considerable [4]. Given the global persistence of malnutrition and ill-health, the research and international development communities are increasingly paying attention to enhancing nutrition and health as the primary goals and outcomes of food production and delivery systems [58-60]. Agriculture as a source of nutritious food and well-being has recently been recognised and is addressed in the new Sustainable Development Goals (SDGs), particularly in SDG 2: "End hunger, achieve food security and improved nutrition and promote sustainable agriculture" [61]. There is, however, an insufficient evidence-base which supports these agriculture, nutrition and health linkages [58-60]. Indeed, according to Masset et al. (2011), who undertook to date the largest systematic review on agricultural intervention to improve the nutritional status of children, there is "no evidence of the impact [of agricultural interventions] on prevalence rates of stunting, wasting and underweight among children under five" [62]. Even though agriculture interventions were beneficial in promoting consumption of nutritious foods, evidence of improved nutritional indicators was not consistent [62-64]. However, according to Webb (2013), the lack of evidence on the impact of agricultural interventions on nutrition and health outcomes should not be attributed to the inefficacy of these interventions, but rather to insufficient statistical power (small sample size), lack of rigorous counterfactual analysis, inadequate selection of outcome indicators for the kind of interventions considered, and few accounted for the heterogeneity of impacts even when they were positive [60, 62, 64, 65]. Furthermore, school-aged children are moving into the focus of recent initiatives by governments, bilateral and multilateral organisations, and other development actors; which have recognised the benefits of good health and nutrition of schoolchildren to contribute to educational achievement, growth and development [3, 66-70].

The two studies in Burkina Faso and Nepal within the frame of the overall VgtS project that we describe here will support the reinforcement of this recent attention on schoolchildren's nutrition and health by focusing on schools as an entry point for health promotion, infection control and life-skills education. Moreover, the studies will contribute to fill existing data gaps on schoolchildren in these two countries, concurrently identifying their nutritional and health

challenges and needs. The data collected will serve to inform the design of appropriate and tailored school-based interventions with close participation of the local community, school teachers and directors, as well as with the local research and VgtS project team. The precise set of interventions will be developed after the baseline survey in Burkina Faso and Nepal. The interventions will be designed with a multidisciplinary team of educators, epidemiologist, nutritionist, parasitologists and WASH experts in order to improve water quality, sanitary and hygiene environments and to translate the nutritional and health risk factors into effective educational messages, thereby encouraging schoolchildren to change their behaviour.

The two studies also aim to address the scientific research gap by conducting rigorous intervention studies and quantifying the possible effects of complementary school garden, nutrition and WASH interventions. With the two particular study designs as suggested in Burkina Faso and Nepal, we will be able to analyse the different types of school garden, nutrition and WASH intervention packages. While in Burkina Faso the focus will be on the comparison of integrated and complementary school garden, nutrition and WASH interventions (arm A) as compared to the control schools with no interventions (arm B); in Nepal, we will additionally be able to conduct comparisons between these two study arms to the school garden intervention schools (arm C) and the nutrition and WASH intervention schools (arm D).

Several considerations underscore the relevance for the two concerted and complementary study designs. First, with the same research methods and questionnaire tools applied, data collected from Burkina Faso and Nepal will be used for comparative analysis. Second, the two similar study designs will offer strategies for comparing different public health approaches with an emphasis on schoolchildren's health and will provide opportunities for discussing the long-term sustainability of these programmes, especially in areas where the targeted diseases are highly prevalent.

Taken together, the overarching goal of the two studies is to assess the potential of suitable interventions to improve health of school-aged children in resource-constrained settings. The insights gained will contribute to estimate the burden of malnutrition and intestinal parasitic infections in schoolchildren and may provide guidance for future research activities, for the implementation of health policies at the school level, as well as for future public health recommendations and health policy planning.

Competing interests

The authors declare no conflict of interest.

Authors' contributions

All listed authors contributed to the development of the study design, essential study documents and standard operating procedures to be employed for the two intervention studies. According to their different areas of expertise, the authors critically revised specific parts of this manuscript (clinical aspects: SD, PO, JU, GC; data management: SE, AK, JG, CS; diagnostic techniques: SD, PO, JU, GC; methodology: SE, AS, AK, PO, JU, GC; study country-specific issues: AS, SD, SS). SE, in collaboration with AS and RH, wrote the first draft of the manuscript. All authors read and approved the final version of the paper prior to submission.

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5 Prevalence and risk factors of undernutrition among schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso

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5.1 Abstract

Background: Multiple factors determine children’s nutritional status, including energy and nutrient intake, recurrent infectious diseases, access (or lack thereof) to clean water and improved sanitation, and hygiene practices, among others. The “Vegetables go to School: improving nutrition through agricultural diversification” (VgtS) project implements an integrated school garden programme in five countries, including Burkina Faso. The aim of this study was to determine the prevalence of undernutrition and its risk factors among schoolchildren in Burkina Faso before the start of the project.

Methods: In February 2015, a cross-sectional survey was carried out among 455 randomly selected children, aged 8–14 years, in eight schools in the Plateau Central and Centre-Ouest regions of Burkina Faso. Nutritional status was determined by anthropometric assessment. Helminth and intestinal protozoa infections were assessed using the Kato-Katz and a formalin-ether concentration method. A urine filtration technique was used to identify *Schistosoma haematobium* eggs. Prevalence of anaemia was determined by measuring haemoglobin levels in finger-prick blood samples. Questionnaires were administered to children to determine their knowledge of nutrition and health and their related attitudes and practices (KAP). Questionnaires were also administered to the children’s caregivers to identify basic household socio-demographic and economic characteristics, and water, sanitation and hygiene (WASH) conditions. To determine the factors associated with schoolchildren’s nutritional status, mixed logistic regression models were used. Differences and associations were considered statistically significant if *P*-values were below 0.05.

Results: Complete datasets were available for 385 children. The prevalence of undernutrition, stunting and thinness were 35.1%, 29.4% and 11.2%, respectively. The multivariable analysis revealed that undernutrition was associated with older age (i.e. 12–14 years compared to < 12 years; adjusted odds ratio (aOR) = 3.45, 95% confidence interval (CI) 2.12–5.62, *P* < 0.001), multiple pathogenic parasitic infections (aOR = 1.87, 95% CI 1.02–3.43, *P* = 0.044) and with moderate and severe anaemia in children (aOR = 2.52, 95% CI 1.25–5.08, *P* = 0.010).

Conclusions: We found high prevalence of undernutrition among the children surveyed in the two study regions of Burkina Faso. We further observed that undernutrition, anaemia and parasitic infections were strongly associated. In view of these findings, concerted efforts are needed to address undernutrition and associated risk factors among school-aged children. As

part of the VgtS project, WASH, health education and nutritional interventions will be implemented with the goal to improve children's health.

Trial registration: ISRCTN17968589 (date assigned: 17 July 2015)

Keywords: Anaemia, Burkina Faso, Intestinal parasitic infections, School garden, Undernutrition, Water, sanitation, and hygiene (WASH)

5.2 Background

In Burkina Faso, undernutrition, anaemia and diarrhoeal diseases are the leading causes of morbidity in children under the age of five. The most recent Demographic and Health Survey (DHS) of 2010 showed that 88% of children under five were anaemic, 35% were undernourished and 15% suffered from diarrhoea in the two weeks preceding the DHS [1]. While DHS and national nutrition surveillance systems in Burkina Faso have routinely measured the height and weight of children under the age of five since the early 1990s, there is a lack of national anthropometric data for school-aged children (5–14 years) [2-4].

The determinants of children’s nutritional status are multifactorial [5-7]. The direct causes of undernutrition in children are insufficient energy and nutrient intake, and recurrent infectious diseases (e.g. intestinal parasitic infection, malaria and diarrhoea) [7]. Factors that affect children’s nutritional status indirectly include a lack of access to clean water and improved sanitation, inadequate hygiene, a paucity of health education and, importantly, inappropriate agricultural practices and insufficiently healthy and diverse diets [5-9]. Low socio-economic and sanitary conditions prevail in Burkina Faso and together contribute to the burden of infectious diseases in children [1, 10, 11], further compromising their nutritional status [5-9, 12].

To address these challenges, research institutions and international development organisations are paying increased attention to enhancing synergies among agriculture, nutrition and health. The Sustainable Development Goals (SDGs) have recognised agriculture as a source of nutrition and well-being, as addressed in SDG 2: “End hunger, achieve food security and improved nutrition and promote sustainable agriculture” [13]. Yet, there is a dearth of evidence to support the effect of agricultural and health interventions on improving children’s nutritional status [14, 15]. To fill this research gap, a multi-country and multi-stakeholder project entitled “Vegetables go to School: improving nutrition through agricultural diversification” (VgtS), was developed to address schoolchildren’s nutrition in an interdisciplinary way, through introducing school vegetable gardens and other school-based health, nutritional and environmental interventions. The VgtS project is active in five countries in Africa and Asia (Bhutan, Burkina Faso, Indonesia, Nepal and the Philippines), with the overall goal of improving schoolchildren’s nutritional status [16]. Under the VgtS project, two intervention studies were implemented in Burkina Faso and Nepal. These studies assessed schoolchildren’s nutritional and health status at baseline and at 12 months follow-up, using a set of selected qualitative and quantitative indicators. The findings from these studies guided the development of complementary nutrition

and water, sanitation and hygiene (WASH) interventions to operate alongside the school garden programme. Details of the study design and procedures have been described elsewhere [16].

The Burkina Faso setting provided an opportunity to understand the complex interactions among agriculture, undernutrition, intestinal parasitic infections and WASH conditions. Agriculture is a major source of livelihoods in the country and inadequate WASH conditions are well known risk factors for both undernutrition and intestinal parasitic infections [11, 17-20]. In this article, we report findings from a cross-sectional baseline survey carried out in Burkina Faso as part of the intervention component of the VgtS project.

5.3 Methods

Study area

We conducted a cross-sectional baseline study in February 2015. The schools participating in the VgtS project in Burkina Faso are located in the Plateau Central and the Centre-Ouest regions. The Plateau Central region is situated in the north-east, approximately 30–120 km from the capital, Ouagadougou. The Centre-Ouest region is located in the south-west, some 40–180 km from Ouagadougou (Figure 5.1). The two regions are located in the semi-arid North-Sudanian zone, characterised by fields, bushes and scattered trees and a Sudano-Sahelian climate (a short wet and a long dry season, with annual precipitation of 600–1000 mm).

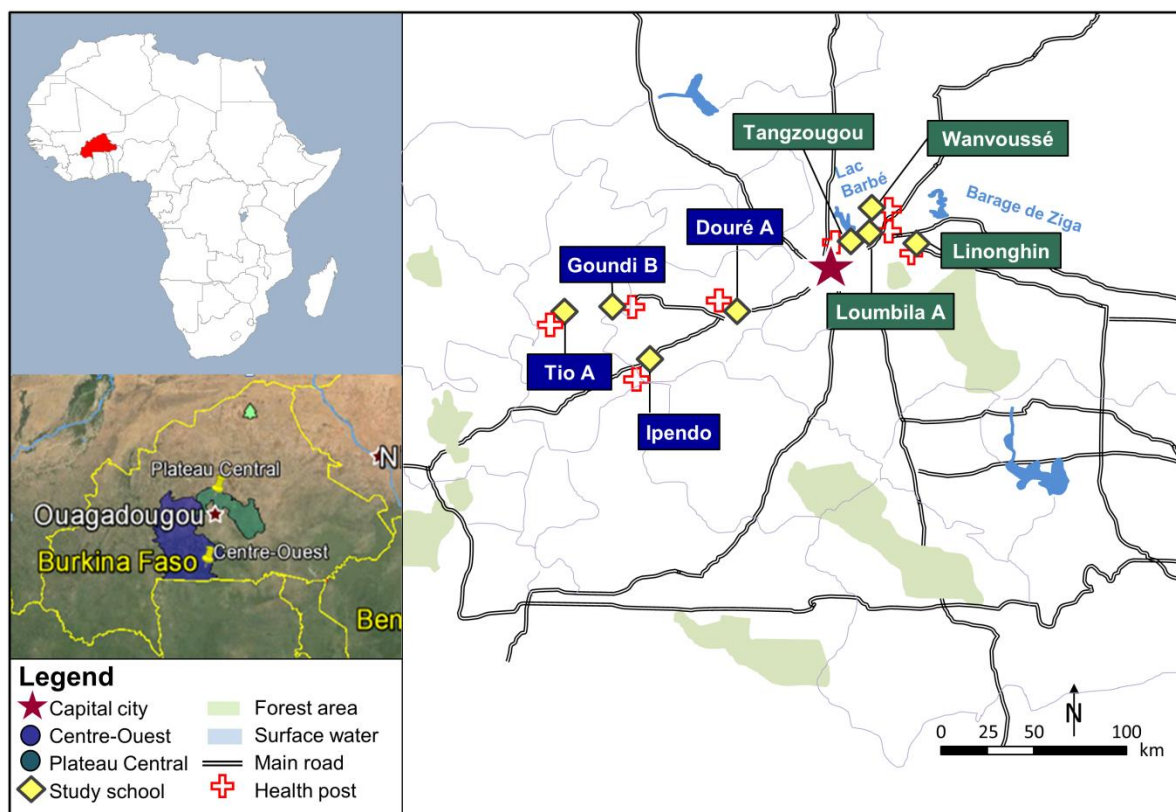


Figure 5.1: Study sites of the cross-sectional survey in Burkina Faso, February 2015

Sample size and sampling method

Our sample size calculation targeted the association between the prevalence of intestinal parasitic infection and the degree of risk among children, aged 8–14 years. We assumed a minimum prevalence of intestinal parasitic infections of 40%, with a coefficient of variation of 10% across schools and a proportion of high-risk children of 25%. We aimed for a power of 85% to detect a difference in infection rates (with $P < 0.05$) between high- and low-risk children at eight schools, for a true odds ratio (OR) of at least 2. A Monte Carlo simulation (5000 iterations) provided a minimal sample size of 400 children (i.e. 50 children per school). Eight of the 30 VgtS project schools in Burkina Faso were randomly selected to participate in the study [16]. In each of the sampled schools, 55–60 children (boys and girls in ratio 1:1) were randomly selected; we assumed that the final sample size would be reduced by 15% due to non-response and missing data [16]. The inclusion criteria for this study were: (i) schoolchildren between the ages of 8 and 14 years; (ii) parents/guardians of the children providing written informed consent; and (iii) children additionally providing oral assent.

Anthropometric survey

Trained field staff collected anthropometric measurements from the children, using a height measuring board and a digital scale (Seca 877; Seca, Germany) with a precision of 0.1 cm and 0.1 kg, respectively and adhering to standard procedures [21]. Anthropometric indices were calculated in accordance with the World Health Organization (WHO) reference, using AnthroPlus (WHO; Geneva, Switzerland) [22, 23]. For children without an exact date of birth or whose age was unknown, school registration lists were consulted. If the exact month or date of birth was unavailable, anthropometric indices were calculated assuming 30 June (mid-year) as the child's date of birth. Three anthropometric indices — height-for-age (HAZ, stunting), body mass index-for-age (BMIZ, thinness) and weight-for-age (WAZ, underweight) — were expressed as differences from the median in z-scores. Children were classified as stunted, thin, or underweight if z-scores of HAZ, BMIZ and WAZ were less than - 2 standard deviations (SD) below the WHO reference median of the standard population. WAZ was only used for children aged 8–10 years, as reference data were not available for children over 10 years [22, 23]. Children were classified as overweight if BMIZ was above 1 SD. We considered children to be malnourished when classified as stunted, thin, underweight or overweight; undernourished children were those classified as stunted, thin or underweight. The categories of stunting, thinness and underweight are not mutually exclusive, as these conditions often overlap; an undernourished child can, for example, be classified as stunted and thin, concurrently.

Haemoglobin survey

Haemoglobin (Hb) concentration was determined in finger-prick capillary blood samples, using a HemoCue portable device (HemoCue Hb 201 System; Ängelholm, Sweden) [24]. Children were classified as mildly anaemic if Hb concentration was less than 11.5 g/dl for children aged 8–11 years and less than 12 g/dl for children aged 12–14 years. Children were classified as moderately and severely anaemic if Hb concentration was less than 11g/dl and 8g/dl, respectively [25].

Parasitological survey

Children were asked to provide a fresh morning stool and a mid-morning post-exercise urine sample, collected on two consecutive days. Stool and urine samples were processed the same day by experienced laboratory technicians. From each stool, a single Kato-Katz thick smear was prepared for diagnosis of soil-transmitted helminths (*Ascaris lumbricoides*, hookworm and *Trichuris trichiura*), *Schistosoma mansoni* and other helminths. A formalin-ether concentration

(FEC) technique was also performed on each sample to diagnose helminths and intestinal protozoa (*Blastocystis hominis*, *Chilomastix mesnili*, *Endolimax nana*, *Entamoeba coli*, *Entamoeba histolytica*/*E. dispar*, *Entamoeba hartmanni*, *Giardia intestinalis*, and *Iodamoeba bütschlii*) [26, 27]. Urine samples were examined for microhaematuria using reagent strips (Hemastix, Siemens Healthcare Diagnostics GmbH; Eschborn, Germany). A urine filtration technique was applied to detect the presence and number of *S. haematobium* eggs [28]. Helminth infection intensity was calculated based on criteria established by the WHO [29].

Questionnaire survey

Questionnaires were administered to children to determine their knowledge of nutrition and health and associated attitudes and practices (KAP) and to the caregivers to identify basic household socio-demographic and economic characteristics and WASH conditions. The KAP and household questionnaires were established according to international guidelines, using standardised questions amended by our research team [1, 30, 31]. Both questionnaires were pre-tested in the study area in November 2014, with children and caregivers who did not subsequently participate in the survey (as part of a pilot study carried out in different schools and villages, far away from those schools selected for the present study). Final local adaptations were made prior to the start of the survey in February 2015.

Data entry and storage

Data were double-entered in Excel 2010 (Microsoft; Redmond, USA). After removing inconsistencies, the datasets were combined and the accuracy of the merged database was verified against the original data through random cross-checking. Data were transferred to and stored electronically on a secure and password-protected server at the Swiss Tropical and Public Health Institute (Swiss TPH; Basel, Switzerland).

Statistical analysis

Categorical variables were described by absolute and relative frequencies. Numerical variables were described by their mean and SD if they were normally distributed, and by their median and interquartile range, otherwise. To characterise household socioeconomic status, we conducted a factor analysis. A list of recorded household assets were included, which took into account the construction materials of the house wall, roof and floor [32]. Four factors reflecting four different socioeconomic domains were retained, including; (i) housing wall materials; (ii) roof materials; (iii) floor materials; and (iv) main energy sources used.

To test for associations between undernutrition (including stunting, thinness and underweight) in children as an outcome variable and associated risk factors, we first conducted a univariable mixed logistic regression analysis with random intercepts at the level of the schools. We included random effects for schools in our logistic regression models, as outcomes might vary between schools due to local factors not accounted for in our models. Non-pathogenic, intestinal protozoa infections (*Trichomonas intestinalis* and *E. coli*) were excluded as potential risk factors for undernutrition in univariable and multivariable analysis. A new variable for hygiene behaviour was created using factor analysis with two conceptually similar categorical variables of: (i) mode of handwashing (e.g. handwashing with soap and water, with water only, with ash, and no handwashing); and (ii) handwashing frequency (before eating, after eating, after playing, and after defecation). Children were classified into one of three categories, reflecting poor, moderate or better hygiene behaviours.

Second, we used a multivariable mixed logistic regression model with random school intercepts and including the categorical exposure variables sex, age, project region and household socioeconomic status as additional independent variables. All other variables were added to the core model one by one, and those with a *P*-value < 0.2 (using likelihood ratio test) were included in the final multivariable model. *ORs* were reported to compare relative odds, while differences and associations were considered as statistically significant if *P*-values were below 0.05, and indicating a trend if *P*-values were between 0.05 and 0.1.

Statistical analyses were performed with Stata version 13 (StataCorp; College Station, USA). Maps, including geographical coordinates of the schools, were established in ArcMap™ version 10 (Environmental System Research Institute; Redlands, USA) and with the Google Earth™ mapping software (<https://www.google.com/earth>).

5.4 Results

Study compliance and respondents' characteristics

Overall, 455 schoolchildren from eight schools were enrolled in the study. Figure 5.2 summarises study participation and compliance, from enrolment to the final sample included for statistical analysis. Parasitological, anthropometric, Hb and KAP questionnaire data were linked by means of a unique identification code (ID). Erroneous ID codes or incomplete datasets with at least one of the parameters missing (e.g. anthropometrics, anaemia, urine and stool analyses, and child and household questionnaires) reduced the number of complete datasets from 455 to 424 children's records and 385 corresponding household records for subsequent

analyses. For households with more than one participating child, one child was selected at random for analysis; hence, another 39 children were excluded and our final dataset comprised 385 children from 385 unique households.

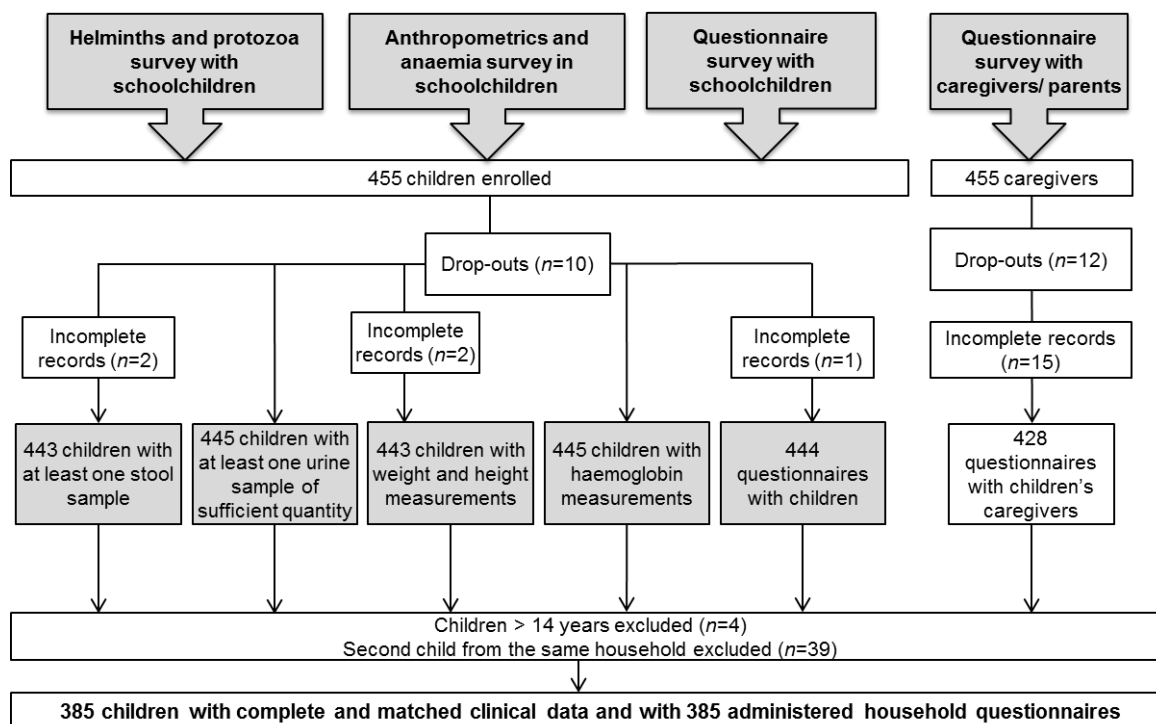


Figure 5.2: Participation in the different study groups of the cross-sectional survey in Burkina Faso, February 2015

The mean age of children interviewed was 11 years (SD 0.7 years, range: 8–14 years). The mean age of the children’s caregivers interviewed was 45 years (SD 14.2 years, range: 20–95 years). Three-quarters of the children’s caregivers had not received any formal education, whereas 59 (15.3%) attended primary school and the remaining 38 (9.9%) received at least a secondary level of education. Almost 90% of children’s caregivers work in the agricultural sector. Respondents’ demographic and economic characteristics are summarised in Table 5.1.

Table 5.1: Characteristics of the study population in the Plateau Central and Centre-Ouest regions, Burkina Faso, February 2015

<i>Children's demographic characteristics</i>		Number	Percent
Age of children*			
Girls		188	48.8
Boys		197	51.2
Age group 1 (8-11 yrs)		251	65.2
Age group 2 (12-14 yrs)		134	34.8
Caregivers' demographic and educational characteristics			
Caregivers' age ⁺			
No formal schooling		288	74.8
Primary education		59	15.3
Secondary or higher education		38	9.9
Main occupation of head of household			
Agriculture		344	89.4
Merchant		8	2.1
Civil service		9	2.3
No employment		2	0.5
Others (housework or retirement)		22	5.7
Socioeconomic domains			
Roof material	Simple (natural and baked clay)	37	9.6
	Metal cover	348	90.4
Wall material	Simple (natural clay)	359	93.3
	Baked or cemented clay	26	6.7
Floor material	Simple (clay, sand, mud, straw)	255	66.2
	Baked or cemented clay	130	33.8
Energy used	Simple (charcoal, firewood)	376	97.7
	Electricity and gas	9	2.3

* = mean age of 11.0 (± 0.7) years+ = mean age of 45.0 (± 14.2) years

Prevalence of malnutrition

Table 5.2 shows the extent of malnutrition, stratified by anthropometric indicators, including age, sex and region. The prevalence of malnutrition and undernutrition in this study were high, at 37.1% and 35.1%, respectively. The prevalence of stunting was 29.4%, while 11.2% of the children were classified as thin. Three out of the 55 children under the age of 10 years were underweight, while eight children were classified as overweight.

Table 5.2: Prevalence of total and specific malnutrition indicators in schoolchildren, Burkina Faso, February 2015

Variable	Malnutrition [n (%)]	Undernutrition [n (%)]	Stunting ^a [n (%)]	Thinness ^a [n (%)]	Underweight ^a [n (%)]	Overweight ^b [n (%)]	Anaemia ^c [n (%)]
Sex							
Female (188)	61 (32.5)	57 (30.3)	47 (25.0)	24 (12.8)	2 (1.1)	4 (2.1)	53 (28.2)
Male (197)	82 (41.6)	78 (39.6)	66 (33.5)	19 (9.6)	1 (0.5)	4 (2.0)	57 (28.9)
Age Group							
8-11 yrs (251)	69 (27.5)	61 (24.3)	47 (18.7)	16 (6.4)	3 (1.2)	8 (3.2)	55 (21.9)
12-14 yrs (134)	74 (55.2)	74 (55.2)	66 (49.3)	27 (20.2)	NA ^d	0 (0)	55 (41.0)
Region							
Plateau Central (198)	69 (34.9)	64 (32.3)	50 (25.3)	19 (9.6)	2 (1.0)	5 (2.5)	53 (26.8)
Centre-Ouest (187)	74 (39.6)	71 (38.0)	63 (33.7)	24 (12.8)	1 (0.5)	3 (1.6)	57 (30.5)
Total	143 (37.1)	135 (35.1)	113 (29.4)	43 (11.2)	3 (0.8)	8 (2.1)	110 (28.6)

^a z-score < - 2^b z-score > 1^c The category of anaemia includes all children classified as anaemic (mild, moderate and severe) based on the concentrations of haemoglobin (Hb) determined in a finger prick blood sample. The cut-offs for anaemia are age-specific: Hb < 11.5 g/dl for children aged 8-11 years, and Hb < 12 g/dl for children aged 12-14 years.^d NA = not available

Intestinal parasitic and *Schistosoma* infections

Table 5.3 shows differences in the prevalence of intestinal protozoa, faecal-oral transmitted helminths and *Schistosoma* infections in children, stratified by sex, age and region. We found that 86.2% of the children were infected with at least one intestinal parasite. Intestinal protozoa infections were highly prevalent (84.7%). *Entamoeba histolytica/E. dispar* was the predominant intestinal protozoon species (66.5%), followed by *E. coli* (37.4%), *G. intestinalis* (28.1%) and *T. intestinalis* (23.4%).

Faecal-oral transmitted helminth infections were found in 7.0% of the children. *Hymenolepis nana* was the most frequently occurring species (6.5%). Only three children were infected with hookworm (0.8%). One child had a dual-species infection with hookworm and *H. nana*. Fifteen children were infected with *S. haematobium* (3.9%), while one child was infected with *S. mansoni* (0.3%).

Co-infections were common, affecting 32.5% of the children, whilst 15.6% and 4.7% suffered from triple and quadruplicate infections, respectively. Infections with *H. nana*, *S. haematobium*, hookworm and *S. mansoni* were of light intensity. The prevalence of intestinal protozoa and faecal-oral transmitted helminth infections differed significantly between schoolchildren in the Plateau Central region and those in Centre-Ouest ($P < 0.05$).

Table 5.3: Prevalence of helminths and intestinal protozoa infections in schoolchildren, Burkina Faso, February 2015

Variable	Trematodes		Total schistosomiasis ^a [n (%)]	Nematodes <i>Hookworm</i> [n (%)]	Cestodes <i>H. nana</i> ^b [n (%)]	Total faecal-oral transmitted helminths ^c [n (%)]	Protozoa					Total protozoa [n (%)]
	<i>S. haematobium</i> ^a [n (%)]	<i>S. mansoni</i> ^a [n (%)]					<i>Entamoeba histolytica/E. dispar</i> [n (%)]	<i>Entamoeba coli</i> [n (%)]	<i>Giardia intestinalis</i> [n (%)]	<i>Trichomonas intestinalis</i> [n (%)]	<i>Balantidium coli</i> [n (%)]	
Sex												
Female (188)	7 (3.7)	0 (0)	7 (3.7)	0 (0)	11 (5.9)	11 (5.9)	131 (69.7)	67 (35.6)	44 (23.4)	39 (20.7)	1 (0.5)	161 (85.6)
Male (197)	8 (4.1)	1 (0.5)	9 (4.6)	3 (1.5)	14 (7.1)	16 (8.1) ^c	125 (63.5)	77 (39.1)	64 (32.5)	51 (25.9)	0 (0)	165 (83.8)
Age group												
8-11 yrs (251)	8 (3.2)	0 (0)	8 (3.2)	2 (0.8)	13 (5.2)	15 (6.0)	163 (64.9)	93 (37.1)	69 (27.5)	51 (20.3)	0 (0)	209 (83.3)
12-14 yrs (134)	7 (5.2)	1 (0.8)	8 (6.0)	1 (0.8)	12 (9.0)	12 (9.0) ^c	93 (69.4)	51 (38.1)	39 (29.1)	39 (29.1)	1 (0.8)	117 (87.3)
Region												
Plateau Central (198)	8 (4.0)	0 (0)	8 (4.0)	1 (0.5)	5 (2.5)	6 (3.0)	110 (55.6)	65 (32.8)	49 (24.8)	55 (27.8)	0 (0)	157 (79.3)
Centre-Ouest (187)	7 (3.7)	1 (0.5)	8 (4.3)	2 (1.1)	20 (10.7)	21 (11.2) ^c	146 (78.1)	79 (42.3)	59 (31.6)	35 (18.7)	1 (0.5)	169 (90.4)
Total (385)	15 (3.9)	1 (0.3)	16 (4.2)	3 (0.8)	25 (6.5)	27 (7.0)	256 (66.5)	144 (37.4)	108 (28.1)	90 (23.4)	1 (0.3)	326 (84.7)

^a *Schistosoma haematobium*, *Schistosoma mansoni*

^b *Hymenolepis nana*

^c The category of total faecal-oral transmitted helminths includes children infected with hookworm and *Hymenolepis nana*. There is one child co-infected with hookworm and *Hymenolepis nana*.

Prevalence of anaemia

The mean Hb concentration was 12.3 g/dl (SD 0.7 g/dl). The prevalence of anaemia in our study sample was 28.6% (Table 5.2). Few children were found to be severely anaemic (0.8%), while 11.2% were found to be moderately anaemic and 16.6% mildly anaemic.

Results from the questionnaire surveys

Key results from children's nutrition and health KAP survey and from the household questionnaire are summarised in Table 5.4. While 79.7% of the children reported using latrines at school for defecation, 22.1% reported washing their hands after defecation. Most children (87.8%) reported washing their hands before eating and 7.3% after playing. Four out of five (79.5%) children reported using soap and water to wash their hands. Combining the mode and frequency of handwashing, children were divided into one of three hygiene categories: 14.6% in the lower, 59.0% in the middle and 26.4% in the better hygiene category. Among the households participating in our survey, 55.3% did not own a latrine, while 23.1% had access to an improved latrine. The majority of children (82.1%) and 22.1% of their caregivers stated that they had never heard of malnutrition. Of the interviewed caregivers, 96.9% indicated that their participating child was breastfed.

Table 5.4: Key findings from children's nutrition and health KAP survey and household questionnaire in Burkina Faso, February 2015

Children (n=385)	Number	Percent
Selected KAP^a indicators:		
Handwashing^b		
Water only	344	89.4
Water and soap	306	79.5
With ash	12	3.1
With mud	1	0.3
Before eating	338	87.8
After eating	55	14.3
After playing	28	7.3
After defecation	85	22.1
Do not wash hands	16	4.2
Hygiene behaviour^c		
Lower category (1)	56	14.6
Middle score (2)	227	59.0
Best category (3)	102	26.4
Sanitary behaviour at school		
Using latrines at school	307	79.7
Open defecation (fields, bush)	71	18.5
Others (at home, teachers home)	7	1.8
Meals (day prior to the survey)		
Breakfast	330	85.7
Lunch	351	91.2
Dinner	358	93.0
Nutritional knowledge		
Heard about malnutrition	69	17.9
Households (n=385)		
Household WASH^d characteristics		
Availability of soap (observational)	118	30.7
Type of latrines used		
Flush toilet (i)	0	0
VIP latrine ^e (ii)	14	3.6
Traditional pit latrine (iii)	83	21.6
EcoSan ^f (iv)	60	15.6
Samplat latrine (v)	15	3.9
No facilities/open defecation (vi)	213	55.3
Total improved ^g (i, ii, iv, v)	89	23.1
Total unimproved ^h (iii, vi)	296	76.9
Nutritional knowledge and practices		
Heard about malnutrition	300	77.9
Participating child was breastfed	373	96.9

^a Knowledge, attitudes and practices

^b Multiple responses occurred for the variables characterising the mode (how) and frequency (when) of handwashing.

^c A new variable for hygiene behaviour was created using factor analysis with two conceptually similar categorical variables of: (i) mode of handwashing (handwashing with water and soap, with water only, with ash, no handwashing); and (ii) its frequency (before eating, after eating, after playing, and after defecation). Children were classified into three categories with lower, middle and better hygiene behaviours.

^d Water, sanitation and hygiene

^e Ventilated improved pit (VIP) latrine is an improved type of pit latrine, which helps remove odours and prevent flies from breeding and escaping. Excreta are collected in a dry pit which has a vent pipe covered with a fly-proof screen at the top.

^f Ecological sanitation (EcoSan) toilets are linked to a closed system that does not need water. The toilet is based on the principle of safely recycling excreta resources to create a valuable resource for agriculture.

^g The total improved sanitation category includes sanitation facilities that hygienically separate human excreta from human contact. In our study, these were: (i) flush toilet, (ii) VIP latrine, (iv) EcoSan toilets, and (v) latrine with slab.

^h The total unimproved sanitation category in our study included: (iii) traditional pit latrines, and (vi) no facilities/open defecation).

Results from the logistic regression analysis

Table 5.5 provides an overview of the associations between undernutrition and all measured helminth and pathogenic intestinal protozoa infections, nutrition and health KAP, caregivers' socioeconomic characteristics and WASH conditions observed in univariable and multivariable regression analyses. The prevalence of undernutrition significantly differed between age groups, with the older age group (12–14 years) showing significantly higher odds of undernutrition (aOR = 3.45, 95% CI 2.12–5.62, $P < 0.001$). Girls showed lower odds of being undernourished, but this association lacked statistical significance in the multivariable analysis. No significant association was observed between undernutrition and study region ($P > 0.05$).

Children infected with multiple pathogenic parasites and those with moderate-to-severe anaemia, were at significantly higher odds of being undernourished (aOR = 1.87, 95% CI 1.02–3.43, $P = 0.044$; and aOR = 2.52, 95% CI 1.25–5.08, $P = 0.010$, respectively).

Overall, children with better hygiene behaviours (third category) did not show lower odds for undernutrition than those in the middle or lower hygiene categories ($P > 0.5$). Relying on traditional pit latrines or having no toilet facility at home was not associated with increased odds for undernutrition in children. Moreover, children who reported not having eaten lunch the day prior to the survey and children who were not breastfed showed higher odds of undernutrition, but these associations were not statistically significant ($P > 0.05$). Neither the level of education of the children's caregivers nor their occupation showed any statistically significant association with undernutrition.

Table 5.5: Results from univariable and multivariable logistic regression analysis with undernutrition as outcome

Undernutrition N=385 / N(cases)=135		Univariable logistic regression ^a			Multivariable logistic regression ^b			
		OR	95% CI	P-value	aOR	95% CI	P-value	
Sex	Male	1.00						
	Female	0.70	0.45- 1.09	0.112	0.72	0.46- 1.14	0.163	
Age group	8-11 yrs	1.00						
	12-14 yrs	3.57	2.20- 5.78	< 0.001	3.45	2.12- 5.62	< 0.001	
Region	Centre-Ouest	1.00						
	Plateau Central	0.89	0.35- 2.27	0.804				
Multiple pathogenic parasites	"yes" vs. "no"	1.94	1.09- 3.47	0.025	1.87	1.02- 3.43	0.044	
Intestinal pathogenic protozoa	"yes" vs. "no"	1.78	1.03- 3.06	0.039	1.71	0.97- 3.03	0.064	
<i>Hymenolepis nana</i>	"yes" vs. "no"	1.42	0.60- 3.36	0.425				
<i>Schistosoma haematobium</i>	"yes" vs. "no"	0.76	0.22- 2.56	0.659				
<i>Giardia intestinalis</i>	"yes" vs. "no"	1.44	0.90- 2.32	0.131	1.46	0.89- 2.40	0.133	
<i>Entamoeba histolytica/E. dispar</i>	"yes" vs. "no"	1.39	0.85- 2.25	0.187	1.41	0.85- 2.34	0.184	
Anaemia	No	1.00						
	Mild	1.59	0.89- 2.85	0.121	1.24	0.67- 2.31	0.486	
	Moderate ^c	2.89	1.48- 5.64	0.002	2.52	1.25- 5.08	0.010	
Hygiene ^d	Middle score (2)	1.00						
	Lower category (1)	1.15	0.59- 2.25	0.676				
	Best category (3)	1.36	0.82- 2.25	0.233				
Sanitary behaviour at school	Open defecation ^e	1.00						
	Using latrines at school	0.97	0.48- 1.95	0.922				
	Others (at teachers')	Na						
Household sanitary conditions	Improved latrines	1.00						
	No latrines/ open defecation	0.96	0.54- 0.54	0.886				
	Traditional latrine	1.18	0.60- 2.29	0.634				
Availability of soap	"yes" vs. "no"	1.14	0.70- 1.84	0.599				
Child's eating habits (day prior to the survey)	Breakfast	"no vs. yes" ^f	0.72	0.38- 1.38	0.326			
	Lunch	"no vs. yes" ^f	1.88	0.89- 4.00	0.100	1.52	0.69- 3.32	0.298
	Dinner	"no vs. yes" ^f	1.30	0.57- 2.99	0.534			
Child "heard about malnutrition"	"no vs. yes" ^f	1.11	0.64- 1.95	0.709				
Caregiver "heard about malnutrition"	"no vs. yes" ^f	1.14	0.67- 1.94	0.618				
"Breastfed child"	"no vs. yes" ^f	2.20	0.41- 11.71	0.354				
Caregiver's education	Never went to school	1.00						
	Primary education	1.30	0.71- 2.37	0.390				
	Secondary education	0.87	0.40- 1.89	0.716				
Caregiver's occupation	Agriculture	1.00						
	Civil service	0.35	0.04- 3.01	0.341				
	Merchant	0.35	0.33- 5.23	0.702				
	Others ^g	0.71	0.28- 1.85	0.487				

^a P-value and odds ratio (OR) based on likelihood ratio test. In univariable logistic regression, the overall P-value of the models is indicated in bold letters.

^b P-value and adjusted (a)OR based on likelihood ratio test of the multivariable regression model. The mixed multivariable logistic regression model with random school intercepts included the categorical exposure variables sex, age group, socioeconomic domains and project region. All risk factors that had a p-value lower than 0.2 in the univariable analyses were included into the multivariable regression analysis (as indicated in the table).

^c The category of moderate anaemia includes the severely anaemic children (n=3).

^d This variable was created with two conceptually similar categorical variables of: (i) mode of handwashing (handwashing with soap and water, with water only, with ash, no handwashing); and (ii) handwashing frequency (before eating, after eating, after playing, and after defecation) where multiple responses were possible. Children were classified into one of three categories, with lower, middle and better hygiene behaviours.

^e Open defecation includes the category of defecating in the bush and behind the latrines.

^f The reference category for the OR is "yes" as compared to "no".

^g 'Others' includes homemakers, retirees and unemployed people.

5.5 Discussion

This paper presents findings from a cross-sectional survey on the prevalence of undernutrition and associated risk factors among schoolchildren, aged 8–14 years, from eight schools in the Plateau Central and Centre-Ouest regions of Burkina Faso. We found that undernutrition was highly prevalent among the surveyed children. Approximately a third of the children were undernourished (35.1%).

According to a study conducted in Ouagadougou in 2008/09 for the WHO’s “Nutrition Friendly School Initiative” (NFSI), the prevalence of stunting in schoolchildren (mean age of 11.5 years) was 8.8%, which is considerably lower than the prevalence of stunting among schoolchildren found in this study (29.4%) [33]. The proportion of thinness in children in our study was 11.2%, which is, however, comparable with the 13.7% found in the NFSI study [33]. Overweight children accounted for 2.1% of all children, with a higher incidence among children aged 8–11 years than among the older age group (3.2% vs. 0%), which is similar to the 2.3% reported in the NFSI study [33].

While few children were classified as thin, a considerably higher proportion of children in our study were stunted. Thinness is often associated with short-term risk factors, like seasonal climatic variations (which cause food scarcity/shortages) and increased occurrence of illnesses [34]. Our study was conducted in the post-harvest (mid-dry) season (February), before the commencement of the dry season (March-June) [35], suggesting that the cause of undernutrition was mainly of a chronic nature, associated with long-term risk factors.

The findings from multivariable mixed logistic regression analyses demonstrated a considerably higher risk of undernutrition among children older than 12 years of age. These results are in accordance with other studies, showing a higher prevalence of stunting in older children in low-income countries in Asia and Africa [36-38]. Moreover, children with moderate and severe anaemia (combined category) and with multiple helminths and intestinal pathogenic protozoa infections (“multiple pathogenic parasites”) showed significantly higher odds for undernutrition. Undernutrition and intestinal parasitic infections are intrinsically linked. While undernutrition and inadequate dietary intake lead to weight loss and weakened immunity and render a child more susceptible to infections, parasitic infections contribute to growth stunting by causing a vicious cycle of reduced food intake (loss of appetite), diarrhoea, malabsorption and/or increased nutrient wastage [39-41]. The observed association was statistically significant in our study, reinforcing evidence of the frequent coexistence of these conditions among children [40]. Moreover, while anaemia contributed to higher odds of undernutrition among children in our

study, the aetiology of anaemia is multifactorial and can result from nutritional deficiencies and parasitic infections, among other things, which have been closely connected to the nutritional status of African schoolchildren [42-45].

Our questionnaire survey revealed important inadequacies in nutrition- and health-related knowledge and practices, but no clear association between undernutrition and WASH conditions or nutritional and health KAPs.

Our study has three main limitations. First, the findings presented here cannot be generalised for all of Burkina Faso. Despite the random selection of schools with a sample size large enough for children in this age range, the results are only representative of two regions. Second, the anthropometric survey has certain limitations with respect to the inaccuracy of children's dates of birth. Indeed, we noted that a considerable number of children had their birthdays either on 31 December or on 1 January, according to the existing school records. Upon further probing in the interview, the children often did not know their exact date of birth. Hence, for these children, we took a mid-year point as the date of birth [46]. Third, only one single Kato-Katz thick smear and FEC from two stool samples from two consecutive days were examined for each participant. Our results may therefore underestimate the true prevalence of parasitic infections, due to the low sensitivity of the Kato-Katz technique and urine concentration method [47, 48].

Despite these limitations, our findings highlight a number of important issues. First, undernutrition in schoolchildren in this part of Burkina Faso is highly prevalent. We therefore suggest giving greater attention to the overall nutritional status of school-aged children. So far, comprehensive population-based data, such as the DHS, focus on adolescents over the age of 15 years for sexual and reproductive health issues or on children under five years of age, as they are more vulnerable and prone to disease, illness and death [1, 49-51]. Children under five are often the primary focus of strategies and actions to address malnutrition [7, 52, 53]. Despite the increased odds of survival for children after the age of five (they generally have a lower prevalence of infections when compared to children under the age of five), school-aged children have increased nutritional needs to support the adolescent growth spurt, requiring diets rich in energy and micronutrients and sufficient in both quantity and quality [54]. It is therefore crucial to address the nutritional needs of children in this age group to match their growth requirements [55].

Second, the results of our study highlight the need for a more profound understanding of how helminths and other intestinal parasites mediate pathways to undernutrition. In particular, it is

important to investigate other primary factors related to the burden of undernutrition among school-aged children, such as malaria and other parasitic infections, and the bioavailability and absorption of micronutrients so as to prevent long-term effects of undernutrition [56-58].

To address the factors underlying and contributing to schoolchildren's nutritional status, we support the growing recommendation from several agencies to enhance multidisciplinary strategies and programmes, including nutrition and WASH interventions for school-aged children, in order to ensure optimal health, growth and development continuing after the age of five [59-61]. Such measures should be reflected in the current development of targets and indicators for reaching SDG 2.

5.6 Conclusions

This study provides new insight into the burden of undernutrition and its risk factors among schoolchildren in Burkina Faso, a country that lacks data on the health of children, aged 8–14 years. Our study shows that undernutrition is highly prevalent in the eight schools of the Plateau Central and Centre-Ouest regions (32.3% and 38.0%, respectively) of Burkina Faso. We also observed that undernutrition, anaemia and parasitic infections were strongly associated. In view of these findings, concerted efforts are needed to address undernutrition and the associated risk factors among school-aged children. As part of the VgtS project, WASH, health education and nutritional interventions will be implemented with the goal of improving schoolchildren's health.

List of abbreviations

aOR: Adjusted odds ratio

BMIZ: Body mass index-for-age

CI: Confidence interval

DHS: Demographic and Health Survey

EKNZ: Ethikkommission Nordwest- und Zentralschweiz

FEC: Formalin-ether concentration

HAZ: Height-for-age

ID: Identification code

IRSS: Institute for Health Sciences Research

KAP: Knowledge, attitudes and practices

Hb: Haemoglobin

NFSI: Nutrition Friendly School Initiative

SD: Standard deviation

SDGs: Sustainable Development Goals

Swiss TPH: Swiss Tropical and Public Health Institute

VgtS: Vegetables go to School: improving nutrition through agricultural diversification

WASH: Water, sanitation and hygiene

WAZ: Weight-for-age

WHO: World Health Organization

Ethics approval and consent to participate

The study protocol was approved by the “Ethikkommission Nordwest-und Zentralschweiz” in Switzerland (EKNZ, reference no. 2014-161) and by the “Comité d’Ethique pour la Recherche en Santé, Ministère de la Recherche Scientifique et de l’Innovation, et Ministère de la Santé” (reference no. 2015-02-026). The study is registered with the clinical trial registry ISRCTN (identifier: ISRCTN17968589). Community and school awareness-raising activities entailed holding two meetings in a classroom at each school; one four weeks prior to the study and one on the day of the study. The purpose of these meetings was to discuss the objectives, procedures, potential benefits and risks of the study with district educational authorities, school directors, teachers, parents and community representatives. Informed consent (via signature) was obtained from the child’s parents or guardians. For illiterate parents/guardians, a fingerprint was obtained in the presence of a literate witness from the school (principal or teacher), whilst children assented orally. It was emphasised that participation was voluntary and that children could withdraw at any time without further obligation. All data records were anonymised, provided with a personal identifier and kept confidential.

Results were communicated to participants. Those found with mild or moderate anaemia (Hb < 11.5 g/dl for children aged 8–11 years and Hb < 12 g/dl for children aged 12–14 years) were referred to a local health centre and treated with iron supplements for 40 days, free of charge. Children found with severe anaemia (Hb < 8 g/dl) and severely malnourished children were referred to a local health centre for further investigation, following national guidelines [62, 63]. Children infected with any kind of intestinal protozoa or helminth were treated according to national guidelines (i.e., a 15–50 mg/kg single dose of metronidazole for five consecutive days against intestinal protozoa infection, a triple dose of 400 mg albendazole against soil-transmitted helminth infections, a 40 mg/kg single dose praziquantel against schistosomiasis and four tablets of niclosamide of 500 mg in two doses for six consecutive days to treat *Hymenolepis nana*). Trained teachers, in collaboration with our research team, and local health personnel, with close involvement of the parents/guardians of infected children, administered

anti-parasitic medications and carefully observed children for proper medication intake and adverse events. All treatments were provided free of charge.

Competing interests

The authors declare that they have no competing interests.

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Authors’ contributions

All listed authors contributed to the study design. SE, AMK and SD coordinated the field and laboratory work. TG supervised the laboratory technicians and assisted in data collection with BS. SE and AMK supervised the research assistants. SE performed the statistical analysis under the supervision of CS and drafted the manuscript. AMK, SD, PO, JG, AS, CS, JU and GC contributed to the interpretation of the data, manuscript writing and revisions. All authors read and approved the final manuscript.

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Availability of data and materials

The dataset supporting the conclusions of this article will not be shared. The paper is written as part of the academic degree of a PhD and therefore the data will be used exclusively by the author

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6 Prevalence of intestinal parasitic infections and associated risk factors among schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso

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6.1 Abstract

Background: Unsafe drinking water, unimproved sanitation and lack of hygiene pose health risks, particularly to children in low- and middle-income countries. This study aimed to assess the prevalence and risk factors of intestinal parasitic infections in school-aged children in two regions of Burkina Faso.

Methods: A cross-sectional survey was carried out in February 2015 with 385 children aged 8–14 years from eight randomly selected schools in the Plateau Central and Centre-Ouest regions of Burkina Faso. Stool samples were subjected to the Kato-Katz and a formalin-ether concentration method for the diagnosis of helminths and intestinal protozoa infections. Urine samples were examined with a urine filtration technique for *Schistosoma haematobium* eggs. Water samples from community sources ($n = 37$), children's households ($n = 95$) and children's drinking water cups ($n = 113$) were analysed for contamination with coliform bacteria and faecal streptococci. Data on individual and family-level risk factors were obtained using a questionnaire. Mixed logistic regression models were employed to determine factors associated with intestinal parasitic infections in schoolchildren.

Results: Intestinal parasitic infections were highly prevalent; 84.7 % of the children harboured intestinal protozoa, while helminth infections were diagnosed in 10.7 % of the children. We found significantly lower odds of pathogenic intestinal protozoa infection (*Entamoeba histolytica*/*E. dispar* and *Giardia intestinalis*) among children from the Plateau Central, compared to the Centre-Ouest region ($P < 0.001$). Children from households with “freely roaming domestic animals” ($P = 0.008$), particularly dogs ($P = 0.016$) showed higher odds of *G. intestinalis*, and children reporting exposure to freshwater sources through domestic chores had higher odds of *S. haematobium* infection compared to children without this water contact activity ($P = 0.035$). Water quality, household drinking water source and storage did not emerge as significant risk factors for intestinal parasitic infections in children.

Conclusions: Intestinal protozoa but not helminths were highly prevalent among schoolchildren in randomly selected schools in two regions of Burkina Faso. Our findings call for specific public health measures tailored to school-aged children and rural communities in this part of Burkina Faso. It will be interesting to assess the effect of water, sanitation and hygiene interventions on the transmission of intestinal parasitic infections.

Keywords: Burkina Faso; Helminths; Hygiene; Intestinal protozoa; Polyparasitism; Sanitation; Water

Trial registration: ISRCTN17968589 (date assigned: 17 July 2015).

6.2 Background

Parasitic infections remain a major public health problem, particularly among children in low- and middle-income countries (LMICs). Several infectious diseases caused by intestinal protozoa (e.g. amoebiasis and giardiasis) or parasitic worms (e.g. schistosomiasis and soil-transmitted helminthiasis) have been classified as neglected tropical diseases (NTDs), as they primarily persist in socially and economically deprived communities [1, 2]. The lack of access to clean water, improved sanitation and adequate hygiene (WASH) are major contributors to the burden of NTDs [3-5]. Among pathogenic agents associated with lack of WASH, water-borne diseases such as amoebiasis or giardiasis cause substantial gastrointestinal morbidity, malnutrition and mortality [6, 7]. It has been estimated that intestinal amoebiasis caused by *Entamoeba histolytica* led to 11,300 deaths worldwide and was ranked fourth in the most fatal parasite-related diseases in 2013 [6, 8]. The prevalence of *Giardia intestinalis* was estimated at 2–3 % in the industrialized world and 20–30 % in LMICs [1]. Water-based diseases (e.g. schistosomiasis) and other parasitic infections constitute another major public health issue in LMICs [10]. Indeed, soil-transmitted helminths were estimated to infect more than one billion people in 2010 with highest prevalence rates observed in school-aged children [11]. It should be noted that most research on parasitic diseases and related morbidity focuses on single species infections. To date, there are no estimates for school-aged children, nor for the entire population, on the global burden of diseases due to polyparasitism of intestinal parasitic infections caused by helminths and intestinal protozoa [1, 11-15].

In Burkina Faso, where polyparasitism is common [16, 17], a deeper understanding of multiple species parasite infections is key for disease control and the reduction of the burden due to these (co-) infections. Whilst health data among under 5-year-old children are collected during national Demographic and Health Surveys (DHS) in Burkina Faso, such as anaemia and *Plasmodium* spp. prevalence, there is a paucity of national health statistics pertaining to school-aged children [18].

In the frame of a project entitled “Vegetables go to School: improving nutrition through agricultural diversification” (VgtS), an intervention study has been conducted in Burkina Faso with the objective of: (i) assessing schoolchildren’s health status at baseline and 1-year follow-up; and (ii) linking a school garden programme to complementary nutrition and WASH interventions, which are described in more detail elsewhere [1]. The present study is part of the VgtS baseline assessment and aims at determining the extent of parasitic infections among

children aged 8–14 years and risk factors for infection. Emphasis was placed on household- and school-level water and sanitary conditions, individual hygiene behaviours, and demographic, environmental and socioeconomic characteristics in the Plateau Central and Centre-Ouest regions of Burkina Faso.

6.3 Methods

Study design and participants

We conducted a cross-sectional survey in February 2015 as part of the VgtS project (cluster randomised trial) in Burkina Faso. The study design is described in detail elsewhere [1]. In brief, eight schools out of the 30 VgtS project schools in Burkina Faso were randomly selected and a random sample of children was invited to participate in the current study.

Our sample size was calculated with regard to the association between the prevalence of intestinal parasitic infection and level of risk in children aged 8–14 years. We assumed a prevalence of intestinal parasitic infections of at least 40 % [2], with a coefficient of variation of 10 % across schools, and a proportion of high-risk children being 25 %. We aimed at a power of 85 % to detect a difference in infection rates with $P < 0.05$ between high- and low-risk children for a true odds ratio (OR) of at least 2 and a total of eight schools. A Monte Carlo simulation (5,000 iterations) provided a minimal sample size of 400 children (i.e. 50 children per school). In each of the eight schools, 55–60 children (half boys and half girls) were randomly selected, as we assumed that the final sample size would be reduced by 15 % due to non-response and missing data [1]. The inclusion criteria for the study were: (i) children enrolled in school; (ii) age 8–14 years; (iii) parents or guardians providing written informed consent (fingerprint for illiterate parents/guardians); and (iv) children with oral assent to participate in the study. Children's caregivers who were willing to participate and who had written informed consent were invited to participate in a household questionnaire survey.

Study sites

The study was conducted in the Plateau Central and the Centre-Ouest regions of Burkina Faso. The two regions were selected as VgtS project sites by the local authorities of the Ministry of Education with regards to the objectives of the project and the feasibility of implementing project activities in accessible regions located near the capital Ouagadougou. Both regions lie in the Volta Basin. The Plateau Central region is situated approximately 30–120 km north-east from Ouagadougou and the Centre-Ouest 40–180 km to its south-west (Figure 6.1). The climate of

the Plateau Central is Sudano-Sahelian, marked by a long dry season lasting from October to May and a short rainy season between June and September. Precipitation is irregular and scant with an annual average of 600 to 800 mm. Drinking water is mainly supplied by surface waters, which are primarily provided by the National Water and Sanitation Authority (Office National de l'Eau et de l'Assainissement, ONEA) of Ziniaré. The hydrographic network of the region is relatively dense but most rivers are temporary. As for the Centre-Ouest, the climate is Sudano-Sahelian with annual precipitation ranging from 700 to 1,200 mm. The main water sources used for drinking water are groundwater and water extracted from the Mouhoun River. Communities within our study sites had access to boreholes equipped with manual pumps, as well as improved- and non-improved wells [21].

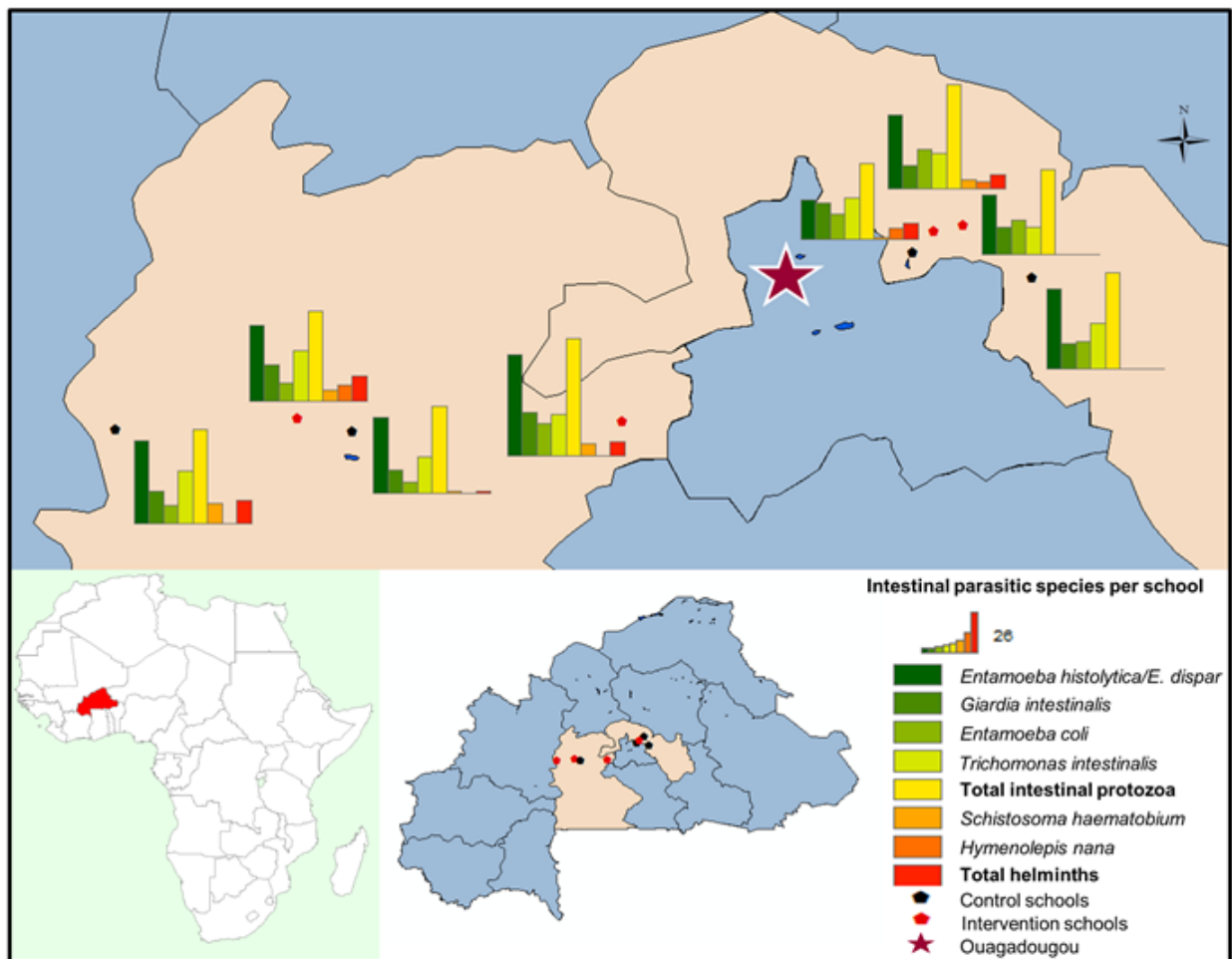


Figure 6.1: Intestinal parasitic infections among schoolchildren in the two regions of Burkina Faso, February 2015

Field and laboratory procedures

A questionnaire was administered to children and their caregivers to identify water sources, sanitary and hygiene knowledge, attitudes and practices (KAP), and exposure to unsafe water and sanitation, including potential confounding factors (e.g. household sociodemographic and economic characteristics). Both questionnaires were established according to international guidelines, using standardised questions and amendments made by our research team [18, 22, 23]. The questionnaires were developed in French, translated orally into the local language by research assistants and pre-tested in November 2014 prior to the survey with children and caregivers who did not otherwise participate in the survey (in different schools and villages). Final local adaptations were done before the start of the survey in February 2015. Research assistants entered data directly into tablet computers (Samsung Galaxy note 10.1 N8010) *via* a data entry mask using Open Data Kit software [24]. Research assistants administered the KAP questionnaire to children at school and visited their caregivers to conduct the household questionnaire at their homes.

Children were asked to provide a fresh morning stool and a mid-morning post-exercise urine sample collected on two consecutive days to assess the presence of soil-transmitted helminths, intestinal protozoa and *Schistosoma* infection. Stool samples were subjected to the Kato-Katz technique (single thick smears, using standard 41.7 mg template), a formalin-ether concentration technique (FECT) for the diagnosis of soil-transmitted helminths (*Ascaris lumbricoides*, hookworm and *Trichuris trichiura*), *S. mansoni*, other helminths and intestinal protozoa (*Blastocystis hominis*, *Chilomastix mesnili*, *Endolimax nana*, *Entamoeba coli*, *E. histolytica/E. dispar*, *E. hartmanni*, *G. intestinalis*, and *Iodamoeba bütschlii*) [25, 26]. Children were considered as positive for a particular infection if at least one of the diagnostic methods revealed a positive result. Urine samples were examined for microhaematuria using reagent strips (Hemastix, Siemens Healthcare Diagnostics GmbH; Eschborn, Germany) and for the presence and number of *S. haematobium* eggs using a urine filtration method [27]. Helminth infection intensity was calculated based on criteria set forth by the World Health Organization (WHO) [28].

Water samples were collected in sterile 250 ml bottles from 30 % of children's drinking water cups ($n = 113$), 20 % of children's households ($n = 95$) and from 4–5 community sources per study site ($n = 37$). Samples were transferred to the laboratory in cooled boxes and stored in a fridge at 4 °C before analysis on the same day.

Three bacterial indicators of faecal contamination, namely *Escherichia coli*, faecal coliforms and faecal streptococci, were determined by a membrane filtration technique [29]. Bacterial cells were concentrated on a 0.2 µm Millipore membrane filter, followed by culture on the chromogenic RAPID E. COLI 2 AGAR (BIO RAD) medium to detect *Escherichia coli* and coliform bacteria, or on the bile-esculine-azide medium to identify faecal streptococci. For *Escherichia coli* and coliform bacteria, incubation was performed at 44.5 °C for 24 h. Colonies of *Escherichia coli* appeared violet to pink, while other coliform colonies stained blue. Faecal streptococci appeared as black stains after 24 h of incubation at 37 °C [29].

Statistical analysis

Kato-Katz thick smear and FECT readings were double-entered into an Excel 2010 spreadsheet (Microsoft; Redmond, USA) and cross-checked. The variable multiple infection was dichotomised in two categories of > 1 , and ≤ 1 infections. Prevalences of intestinal parasitic infections, multiple infections and WASH characteristics were compared according to sex, age group (8–11 years and 12–14 years) and region using univariate mixed logistic regression with random intercepts at the level of schools. Mixed logistic regression models were also applied to investigate associations between dependent variables, namely, infections with *H. nana*; *S. haematobium*; pathogenic intestinal protozoa (*E. histolytica*/*E. dispar* and *G. intestinalis* combined, *E. histolytica*/*E. dispar* and *G. intestinalis*) and 32 independent variables (e.g. sex and age group). Children with non-pathogenic intestinal protozoa infections (*Trichomonas intestinalis* and *Entamoeba coli*) were excluded from logistic regression analysis. A new variable for hygiene behaviour was created using factor analysis with two conceptually similar categorical variables of: (i) mode of handwashing (handwashing with water and soap, with water only, with ash, and no handwashing); (ii) and its frequency (before eating, after eating, after playing and after defaecation). As the median of the factor score had a relative frequency of over 50 %, the hygiene behaviour of children was categorized as poor, moderate or good depending on whether the score was below, at or above the median. Factor analysis was also used to determine household socioeconomic status (SES). From a list of recorded household assets [30], three factors covering four different socioeconomic domains were retained, including (i) housing wall materials; (ii) roof materials; (iii) floor materials; and (iv) main energy sources used. Each factor score was then categorized into tertile classes. Our multivariate core model included a random intercept at the unit of the school and the categorical exposure variables sex, age group, the three categorical SES variables from the factor analysis and project region, which were set, *a priori*, as potential confounders. All the other variables were

assessed one by one and retained for the maximal model if their *P*-value was < 0.2. The final model was then obtained using backward selection with the same level of 0.2. Associations between infections and risk factors are reported as ORs. Differences and associations were considered statistically significant if *P*-values were below 0.05 and as indicating a trend if *P*-values were between 0.05 and 0.1.

To derive estimates of population attributable fractions (PAF), we ran simple Poisson regression models of the infection outcome variables *Y* on binary exposure variables *X*. Estimates of PAF were then obtained via the formula $(RR - 1) \times q / (1 + (RR - 1) \times q)$ where *RR* denotes the relative risk estimate provided by the Poisson regression model and *q* denotes the prevalence of exposure *X*. Confidence limits of the PAF-estimates were obtained using the same formula. Statistical analyses were done using STATA version 13.0 (Stata Corporation; College Station, USA).

6.4 Results

Study participation, demographic and socioeconomic profile

Complete datasets were available for 385 children and their caregivers. Of the final study participants, 48.8 % were girls. The age structure of participating children was as follows: 65.2 % were aged 8–11 years and 34.8 % were aged 12–14 years. There was no statistically significant difference in the number of boys and girls in the two age groups (all *P* > 0.05).

Respondents' demographic and socioeconomic characteristics are summarised in Table 6.1. Mossi was the predominant ethnic group (68.1 %), followed by Gourunsi with 29.6 %. Most Mossi lived in the Plateau Central, while Gourunsi predominantly lived in the Centre-Ouest region. The houses of children's families were mainly made of adobe walls (93.3 %), a tin roof (90.4 %) and a clay or mud-type floor (66.6 %). Only 2.3 % of the households were connected to the power grid using electricity or gas; the remaining households used charcoal and firewood as principal energy source. Almost 90 % of children's caregivers worked in the agricultural sector, while 10.6 % reported non-agricultural sources of income. Domestic animals were kept by 96.4 % of the families, while 63.9 % reported to letting them roam freely within their households. Dogs and goats were particularly common (76.6 and 64.7 %, respectively), followed by cats (39.7 %), swine (28.6 %), cattle (28.3 %), poultry (15.9 %) and sheep (4.4 %). Three-quarters (74.8 %) of the children's caregivers had no formal education, whereas 15.3 % attended primary school and the remaining 9.9 % reached at least a secondary level of education.

Table 6.1: Characteristics of the study population in the two regions of Burkina Faso in February 2015

Children's demographic characteristics (n = 385)		[n (%)]	Plateau Central [n (%)]	Centre-Ouest [n (%)]
Sex				
	Girls	188 (48.8)	97 (49.0)	91 (48.7)
	Boys	197 (51.2)	101 (51.0)	96 (51.3)
Age of children ^a				
	Age group 1 (8–11 years)	251 (65.2)	147 (74.2)	104 (55.6)
	Age group 2 (12–14 years)	134 (34.8)	51 (25.8)	83 (44.4)
Ethnicity				
	Mossi	262 (68.1)	189 (95.5)	73 (39.0)
	Gourunsi	114 (29.6)	1 (0.5)	113 (60.5)
	Others (Dioula, Peulh)	9 (2.3)	8 (4.0)	1 (0.5)
Caregiver's socioeconomic characteristics (n = 385)				
Roof material	Simple (natural and baked clay)	37 (9.6)	12 (6.1)	25 (13.4)
	Metal cover	348 (90.4)	186 (93.9)	162 (86.6)
Wall material	Simple (natural clay)	359 (93.3)	182 (91.9)	177 (94.7)
	Baked or cemented clay	26 (6.7)	16 (8.1)	10 (5.3)
Floor material	Simple (clay, sand, mud, straw)	255 (66.2)	115 (58.1)	140 (74.9)
	Baked or cemented clay	130 (33.8)	83 (41.9)	47 (25.1)
Energy used	Simple (charcoal, firewood)	376 (97.7)	191 (96.5)	185 (98.9)
	Electricity and gas	9 (2.3)	7 (3.5)	2 (1.1)
Possession of domestic animals		371 (96.4)	187 (94.4)	184 (98.4)
Animals roaming freely in household		246 (63.9)	124 (62.6)	122 (65.2)
Caregiver's socio-demographic characteristics (n = 385)				
Caregiver's age ^b				
	No formal schooling	288 (74.8)	142 (71.7)	146 (78.1)
	Primary education	59 (15.3)	28 (14.1)	31 (16.6)
	Secondary or higher education	38 (9.9)	28 (14.1)	10 (5.4)
Main occupation of head of household				
	Agriculture	344 (89.4)	180 (90.9)	164 (87.7)
	Merchant	8 (2.1)	7 (3.5)	1 (0.5)
	Civic service	9 (2.3)	3 (1.5)	6 (3.2)
	Others (housework, retirement and no employment)	24 (6.2)	8 (4.0)	16 (8.6)

^a = mean age of 11.0 (±0.7) years; 10.8 (±0.1) in the Plateau Central and 11.2 (±0.1) in the Centre-Ouest

^b = mean age of 45.0 (±14.2) years; 44.8 (±14.3) in the Plateau Central and 45.2 (±14.1) in the Centre-Ouest

Prevalence of intestinal parasitic infections

The prevalence of intestinal parasitic infections, stratified by sex, age group and region, are summarised in Table 6.2. Over 80 % of the schoolchildren were infected with intestinal protozoa. The predominant species was *E. histolytica/E. dispar* (66.5 %), followed by *Entamoeba coli* (37.4 %), *G. intestinalis* (28.1 %), and *Trichomonas intestinalis* (23.4 %). The total prevalence of helminth infections was 10.7 %. *Hymenolepis nana* was the most frequent species (6.5 %), followed by *S. haematobium* (3.9 %) (Figure 6.1). Three children were infected with hookworm (0.8 %) and one with *S. mansoni* (0.3 %). Infections with *H. nana*, *S. haematobium*, hookworm and *S. mansoni* were all of light intensity.

Table 6.2: Intestinal parasitic infections among schoolchildren in two regions of Burkina Faso in February 2015

Parasite	Prevalence [n (%)]	Sex ^a		Age group ^b		Region ^c	
		F	M	8–11	12–14	PC ^d	CO ^d
Trematodes							
<i>Schistosoma haematobium</i>	15 (3.9)	7 (3.7)	8 (4.1)	8 (3.2)	7 (5.2)	8 (4.0)	7 (3.7)
<i>Schistosoma mansoni</i>	1 (0.3)	0 (0.0)	1 (0.5)	0 (0.0)	1 (0.8)	0 (0.0)	1 (0.5)
Total <i>Schistosoma</i> spp.	16 (4.2)	7 (3.7)	9 (4.6)	8 (3.2)	8 (6.0)	8 (4.0)	8 (4.3)
Nematodes							
Hookworm	3 (0.8)	0 (0.0)	3 (1.5)	2 (0.8)	1 (0.8)	1 (0.5)	2 (1.1)
Cestodes							
<i>Hymenolepis nana</i>	25 (6.5)	11 (5.9)	14 (7.1)	13 (5.2)	12 (9.0)	5 (2.5)	20 (10.7)
Total faecal-oral transmitted helminths^e	27 (7.0)	11 (5.9)	16 (8.1)	15 (6.0)	12 (9.0)	6 (3.0)	21 (11.2)
Intestinal protozoa							
<i>Entamoeba histolytica/E. dispar</i>	256 (66.5)	131 (69.7)	125 (63.5)	163 (64.9)	93 (69.4)	110 (55.6)	146 (78.1)
<i>Entamoeba coli</i>	144 (37.4)	67 (35.6)	77 (39.1)	93 (37.1)	51 (38.1)	65 (32.8)	79 (42.3)
<i>Giardia intestinalis</i>	108 (28.1)	44 (23.4)	64 (32.5)	69 (27.5)	39 (29.1)	49 (24.8)	59 (31.6)
<i>Trichomonas intestinalis</i>	90 (23.4)	39 (20.7)	51 (25.9)	51 (20.3)	39 (29.1)	55 (27.8)	35 (18.7)
<i>Balantidium coli</i>	1 (0.3)	1 (0.5)	0 (0.0)	0 (0.0)	1 (0.8)	0 (0.0)	1 (0.5)
<i>Entamoeba histolytica/E. dispar</i> or <i>Giardia intestinalis</i>	290 (75.3)	144 (76.6)	146 (74.1)	182 (72.5)	108 (80.6)	130 (65.7)	160 (85.6)
Total intestinal protozoa^f	326 (84.7)	161 (85.6)	165 (83.8)	209 (83.3)	117 (87.3)	157 (79.3)	169 (90.4)
Multiple intestinal parasitic infection^g	206 (53.5)	101 (53.7)	105 (53.3)	124 (49.4)	82 (61.2)	103 (48.0)	111 (59.4)

^aSignificant differences in investigated parasite infection prevalence between boys and girls were found for *Giardia intestinalis* ($P = 0.05$)

^b*Trichomonas intestinalis* and multiple parasitic infection prevalence were significantly different between age groups ($P < 0.05$)

^cSignificant regional differences were found for *Hymenolepis nana*, any faecal-oral transmitted helminth, *Entamoeba histolytica/E. dispar*, *Entamoeba coli*, *Trichomonas intestinalis*, *Entamoeba histolytica/E. dispar* or *Giardia intestinalis*, total intestinal protozoa infection, and multiple intestinal parasitic infection ($P < 0.05$)

^dPC, Plateau Central; CO, Centre-Ouest region of Burkina Faso

^eThe category of total faecal-oral transmitted helminths includes children infected with hookworm and *Hymenolepis nana*. There was one child co-infected with hookworm and *Hymenolepis nana*

^fSeveral children were co-infected with intestinal protozoa. The total of this category therefore does not sum up from the separate figures

^gMultiple intestinal parasitic infection was defined as dichotomous variable, classified as > 1 infection vs ≤ 1 infection

Polyparasitism was common; on average, a study participant harboured 1.7 concurrent parasite species. The maximum number of parasite species found in the same host was five. The large majority of children (86.2 %) were infected with at least one intestinal parasite. Dual (32.5 %), triple (15.6 %), and quadruplicate infections (4.7 %) were also recorded (Figure 6.2).

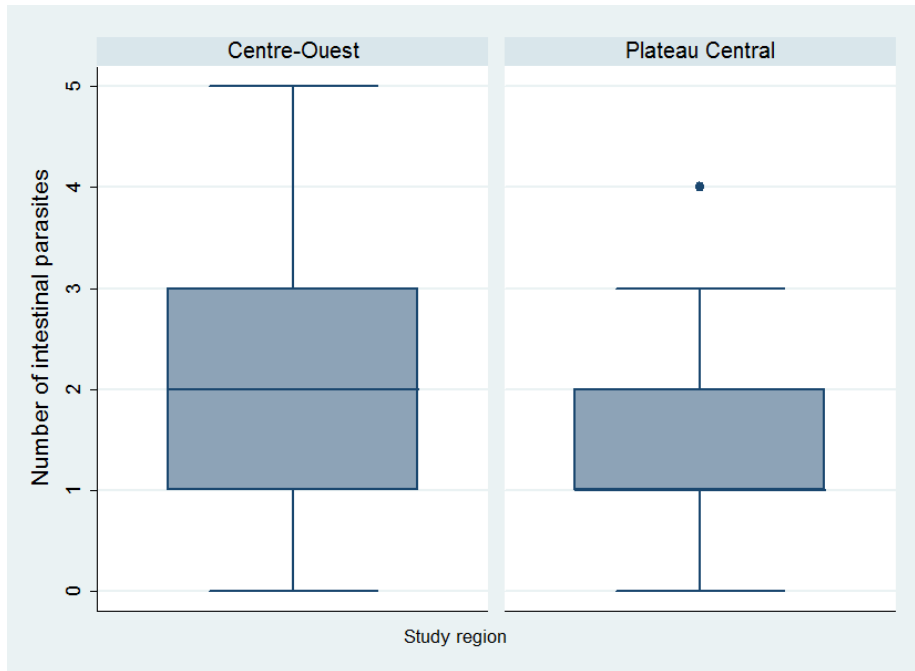


Figure 6.2: Number of concurrent intestinal parasitic infections, stratified by region among 385 schoolchildren in Burkina Faso.

Box plot: boxes illustrate the 25th and 75th percentiles (ptile), while the whiskers indicate the adjacent lower and upper values (values which are within 25th ptile – 1.5 * (75th – 25th ptile) and 75th ptile + 1.5 * (75th – 25th ptile), respectively) and values outside these bounds are plotted individually. The median is shown by the line within the boxes.

Significant regional differences were observed for the total of intestinal protozoa species found ($\chi^2 = 4.68$, $df = 1$, $P = 0.03$). There were considerable differences for multiple intestinal parasitic infection profiles among the two regions. Children from the Centre-Ouest were at higher odds of multiple parasitic infections compared to children from the Plateau Central ($\chi^2 = 4.98$, $df = 1$, $P = 0.03$). The prevalence of infection with *G. intestinalis* was significantly lower in girls compared to boys ($\chi^2 = 9.16$, $df = 6$, $P = 0.05$; see Appendix 9.1.1). *Trichomonas intestinalis* and multiple parasitic infection prevalence were significantly different between age groups, with children aged 12–14 years at higher odds of infection (*T. intestinalis*: $\chi^2 = 3.89$, $df = 1$, $P = 0.05$; multiple parasitic infections: $\chi^2 = 4.85$, $df = 1$, $P = 0.03$).

WASH behaviours

Based on the findings of the questionnaires conducted with children and their caregivers, most children (87.8 %) reported to wash their hands with soap before eating, while only 22.1 % of children reported doing so after defaecation with an even smaller proportion after playing (7.3 %). Almost 20 % of the children reported defaecating in the fields and bushes. Only 23.1 % of the households had access to an improved latrine, while 21.6 % of their families used a traditional pit latrine and 55.3 % did not own a latrine (Table 6.3).

The overall hygiene behaviour, including the modality of handwashing as well as its frequency, and the availability of household latrines was not significantly different across study regions. Yet, statistically significant regional differences were found with regards to children's sanitary practices; children from the Centre-Ouest practised open defaecation (dichotomised variable with the use of any latrines and open defaecation) more frequently than their counterparts from the Plateau Central (31.5 vs 6.1 %; $\chi^2 = 4.67$, $df = 1$, $P = 0.03$).

Children reported both to drink water at school (83.6 %) and to bring water for consumption from home (62.1 %) (multiple responses were possible). Over 60 % of children's families were said to use borehole water as drinking water source in the rainy and the dry seasons, as compared to wells, surfaces or collected rain waters. Most households reported storing their water in an open receptacle (72.2 %). Only 17.9 % said they treated their drinking water before consumption. Statistically significant regional differences were found for reported drinking water treatment; households from the Centre-Ouest treated their water more frequently compared to households from the Plateau Central (25.7 vs 10.6 %; $\chi^2 = 5.53$, $df = 1$, $P = 0.02$). The modality of drinking water storage (open vs closed) and children's water exposure through playing, fishing, domestic chores or making laundry did not significantly differ across regions (all $P > 0.05$).

Associations between children's parasitic infection status and handwashing, sanitary, and hygiene behaviours are summarised in Appendix 9.1.2. Overall, children with both poor and better hygiene behaviours (first and third category) showed lower odds for any intestinal pathogenic protozoa infections than the middle category, however without these differences reaching statistical significance. Children from households with improved latrines and with soap for handwashing available did not show lower odds for any intestinal parasitic protozoa infection. However, children from households with soap for handwashing available showed lower odds for *H. nana* infection ($P = 0.23$) and *S. haematobium* infection ($P = 0.06$). Children reporting to play, fish and to do domestic chores in water, rivers or watersheds showed higher odds for *S. haematobium* infection, but only exposure through domestic chores was statistically

significant ($\chi^2 = 22.65$, $df = 7$, $P = 0.04$). Schoolchildren that reported to drink water from the school source showed significantly lower odds for *S. haematobium* and *H. nana* infection, yet, only the latter was significant in multivariate analysis ($\chi^2 = 5.36$, $df = 7$, $P = 0.02$). No statistically significant association was found between reported drinking water sources and storages and children's intestinal protozoa infection status (all $P > 0.05$).

Among domestic animals held by children's caregivers (cats, cattle, dogs, goats, poultry, sheep and swine), we found a significant association between *G. intestinalis* infection in children and the possession of dogs ($\chi^2 = 14.42$, $df = 7$, $P = 0.016$; Appendix 9.1.1). Domestic animals freely roaming in households contributed to 25.6 % of *G. intestinalis* infection in children (95 % CI 4.0–64.4 %), while dogs contributed to 20.0 % of *G. intestinalis* infection in children (95 % CI 2.6–38.8%). The estimated fraction of *S. haematobium* infection attributable to “any water contact” defined as exposure to freshwater during playing, fishing or doing domestic chores) was 72.0 % infection (95 % CI -45.6–96.1 %).

Drinking water quality

Table 6.3 shows the findings from the drinking water quality analysis. About 90 % of water samples from children's drinking water cups and children's households were contaminated with both faecal coliform bacteria (89.4 and 93.6 %, respectively) and faecal streptococci (89.4 and 92.7 %, respectively). The proportion of samples contaminated with *Escherichia coli* was smaller; 64.2 % of household drinking water and 48.7 % of children's drinking water cups were contaminated. Water samples from community sources were less contaminated with faecal coliform bacteria (35.1 %), faecal streptococci (27.0 %) and *Escherichia coli* (24.3 %).

Significant regional differences were found between water samples contaminated with faecal coliform bacteria from children's drinking water cups (80.7 % in the Plateau Central vs 98.2% in the Centre-Ouest; $\chi^2 = 5.87$, $df = 1$, $P = 0.02$), and water samples contaminated with *Escherichia coli* from both children's drinking water cups (29.8 % in the Plateau Central vs 67.9 % in the Centre-Ouest; $\chi^2 = 15.51$, $df = 1$, $P < 0.001$) and households (48.9 % in the Plateau Central vs 79.2 % in the Centre-Ouest, $\chi^2 = 8.97$, $df = 7$, $P = 0.003$).

In univariate logistic regression analysis, household drinking water contaminated with faecal streptococci was associated with a higher odds of total intestinal pathogenic protozoa infections in children ($P = 0.06$), while this association almost collapsed in multivariate analysis ($P = 0.46$). No significant association was found between water quality of community sources and children's

drinking water cups and their status of infection with total pathogenic intestinal protozoa ($P = 0.79$ and $P = 0.67$, respectively).

Table 6.3: Questionnaire findings and water quality in the two regions of Burkina Faso, February 2015

Children ($n = 385$)	[n (%)]	Plateau Central [n (%)]	Centre-Ouest [n (%)]
Selected KAP indicators^a			
Handwashing^b			
Before eating	338 (87.8)	164 (82.8)	174 (93.1)
After eating	55 (14.3)	25 (12.6)	30 (16.0)
After playing	28 (7.3)	12 (6.1)	16 (8.6)
After defaecation	85 (22.1)	41 (20.7)	44 (23.5)
Do not wash hands	16 (4.2)	15 (7.6)	1 (0.5)
Water only	344 (89.4)	183 (92.4)	161 (86.1)
Water and soap	306 (79.5)	153 (77.3)	153 (81.8)
With ash	12 (3.1)	0 (0.0)	12 (6.4)
With mud	1 (0.3)	0 (0.0)	1 (0.5)
Hygiene^c			
Lower category (1)	56 (14.6)	33 (16.7)	23 (12.3)
Middle score (2)	227 (59.0)	119 (60.1)	108 (57.7)
Best category (3)	102 (26.4)	46 (23.2)	56 (30.0)
Sanitary practices at school*			
Using latrines at school	307 (79.7)	181 (91.4)	126 (67.4)
Open defaecation (fields, bush)	71 (18.5)	12 (6.1)	59 (31.5)
Using latrines at home/ at teachers' house	7 (1.8)	5 (2.5)	2 (1.1)
Drinking water^d			
Drinking water from school	322 (83.6)	174 (87.9)	148 (79.1)
Bringing drinking water from home	239 (62.1)	112 (56.6)	127 (67.9)
Quality of water in children's drinking cups ($n = 113$)			
Coliform bacteria*	101 (89.4)	46 (80.7)	55 (98.2)
<i>Escherichia coli</i> *	55 (48.7)	17 (29.8)	38 (67.9)
Faecal streptococci	101 (89.4)	50 (87.7)	51 (91.1)
Safe to drink without prior treatment	3 (2.7)	3 (5.3)	0 (0.0)
Households ($n = 385$)			
Household WASH characteristics^e			
Type of latrines used			
Flush toilet (i)	0 (0.0)	0 (0.0)	0 (0.0)
VIP latrine ^f (ii)	14 (3.6)	12 (6.1)	2 (1.1)
Traditional pit latrine (iii)	83 (21.6)	65 (32.8)	18 (9.6)
EcoSan ^g (iv)	60 (15.6)	33 (16.7)	27 (14.4)
Samplat latrine (v)	15 (3.9)	13 (6.6)	2 (1.1)
No facilities / open defaecation (vi)	213 (55.3)	75 (37.9)	138 (73.8)
Total improved ^h (i, ii, iv, v)	89 (23.1)	58 (29.3)	31 (16.6)
Total unimproved ⁱ (iii, vi)	296 (76.9)	140 (70.7)	156 (83.4)
Preferred source of drinking water during the rainy season			
Private tab	1 (0.3)	1 (0.5)	0 (0.0)
Shared tab	1 (0.3)	1 (0.5)	0 (0.0)
Public tab	25 (6.5)	18 (9.1)	7 (3.7)
Improved source	4 (1.0)	4 (2.1)	0 (0.0)
Un-improved source	8 (2.1)	0 (0.0)	8 (4.3)
Borehole water	249 (64.6)	161 (81.3)	88 (47.1)
Collected rain water	1 (0.3)	1 (0.5)	0 (0.0)
Surface water	3 (0.8)	1 (0.5)	2 (1.1)
Wells	87 (22.6)	14 (7.1)	73 (39.0)
Others	6 (1.5)	1 (0.5)	5 (2.7)

Preferred source of drinking water during the dry season			
Private tap	1 (0.3)	1 (0.5)	0 (0.0)
Shared tap	2 (0.5)	2 (1.0)	0 (0.0)
Public tap	25 (6.5)	18 (9.1)	7 (3.7)
Improved source	4 (1.0)	0 (0.0)	4 (2.1)
Un-improved source	9 (2.4)	0 (0.0)	9 (4.8)
Borehole water	261 (67.8)	168 (84.9)	93 (49.7)
Surface water	0 (0.0)	0 (0.0)	0 (0.0)
Wells	81 (21.0)	8 (4.0)	73 (39.0)
Others	2 (0.5)	1 (0.5)	1 (0.5)
Household drinking water storage			
Open	278 (72.2)	141 (71.2)	137 (73.3)
Pot or canary	290 (75.3)	146 (73.7)	144 (77.0)
Basin or bowl	16 (4.2)	2 (1.0)	14 (7.5)
Canister (plastic jerrican)	59 (15.3)	38 (19.2)	21 (11.2)
Others	18 (4.7)	11 (5.6)	7 (3.7)
No storage	2 (0.5)	1 (0.5)	1 (0.5)
Household drinking water treated prior to consumption ^l *	69 (17.9)	21 (10.6)	48 (25.7)
Water quality of household drinking water (n = 95)			
Coliform bacteria	89 (93.7)	42 (89.4)	47 (97.9)
<i>Escherichia coli</i> [*]	61 (64.2)	23 (48.9)	38 (79.2)
Faecal streptococci	88 (92.6)	42 (89.4)	46 (95.8)
Safe to drink without prior treatment	0 (0.0)	0 (0.0)	0 (0.0)
Water quality of community sources (n = 37)			
Coliform bacteria	13 (35.1)	4 (22.2)	9 (47.4)
<i>Escherichia coli</i>	9 (24.3)	0 (0.0)	9 (47.4)
Faecal streptococci	10 (27.0)	2 (11.1)	8 (42.1)
Safe to drink without prior treatment	22 (59.5)	12 (66.7)	10 (52.6)

^aKnowledge, attitudes and practices

^bMultiple responses were possible for the variables characterising the mode (how) and frequency (when) of handwashing.

^cA new variable for hygiene behaviour was created using factor analysis with the mode and frequency of handwashing. Children were classified into three categories with poor, middle and good hygiene behaviours

^dMultiple responses were possible for the variables characterising the child's drinking water consumption at school

^eWater, sanitation, and hygiene

^fVentilated improved pit (VIP) latrine is an improved type of pit latrine, which helps remove odours and prevent flies from breeding and escaping. Excreta are collected in a dry pit which has a vent pipe covered with a fly-proof screen at the top

^gEcological sanitation (EcoSan) toilets are linked to a closed system that does not need water. The toilet is based on the principle of safely recycling excreta resources to create a valuable resource for agriculture

^hThe improved sanitation category includes all sanitation facilities that hygienically separate human excreta from human contact; i.e. pit latrine with slab, VIP and EcoSan toilets.

ⁱThe unimproved sanitation category includes traditional pit latrines and no facilities (open defaecation)

^jHouseholds having reported to treat their drinking water through filtration and sedimentation

*Significant regional differences were found for children's sanitary practices (dichotomised variable classified as using latrines vs. open defaecation, $\chi^2 = 4.67$, $df = 1$, $P = 0.03$), water quality of children's drinking water cups (coliform bacteria, $\chi^2 = 5.87$, $df = 1$, $P = 0.02$; *Escherichia coli*, $\chi^2 = 15.51$, $df = 1$, $P < 0.001$); household water treatment ($P = 0.02$); and water quality of household drinking water (*Escherichia coli*, $\chi^2 = 8.97$, $df = 7$, $P = 0.003$) using mixed logistic regression models with random intercepts at the level of schools

6.5 Discussion

The findings of the present cross-sectional survey conducted in eight schools in the Plateau Central and the Centre-Ouest regions of Burkina Faso in February 2015 showed that 86.2 % of the participating children aged 8–14 years harboured at least one species of intestinal parasite. Intestinal protozoa were most commonly found; the two predominant pathogenic intestinal protozoan species in the two study regions under investigation were *E. histolytica/E. dispar* (66.5 %) and *G. intestinalis* (28.1 %). Interestingly, we found a significant association between domestic animals roaming freely within households compared to households where domestic animals were kept outside and the prevalence of *G. intestinalis* among schoolchildren. There was a significant association between *G. intestinalis* infection in children and the presence of dogs at the unit of the household. A number of studies have demonstrated *G. intestinalis* as prevalent in both humans and dogs worldwide and have postulated the occurrence of anthroponotic, zoonotic and animal-specific cycles of transmission [31, 32]. The risk of dogs as potential reservoirs would need molecular confirmation [31]. Nevertheless, this finding illustrates the importance of the household environment and highlights the potential role of freely roaming animals, particularly dogs, in the transmission of *G. intestinalis* (PAF of 20.0%) [31, 33].

Hymenolepis nana was the predominant helminth species, however, the overall prevalence was relatively low (6.5 %). Of note, *H. nana* was also the main helminth species found in previous studies in Burkina Faso [16, 34], and is most often found in countries in which sanitation and hygiene are inadequate [35, 36]. We did, however, not find a significantly lower prevalence of *H. nana* in schoolchildren with better hygiene behaviours as would have been anticipated. There was a tendency for lower odds of *H. nana* infections for schoolchildren from families reporting to drink water from borehole sources; yet, these associations lacked statistical significance in multivariate analysis. However, schoolchildren that reported to drink water from the school source showed significantly lower odds for *H. nana* infection in multivariate analysis. It is conceivable that unsafe drinking water contaminated with soil or faeces could act as a carrier of infectious *H. nana* eggs. Yet, the normal mode of transmission is ingestion of the eggs in food contaminated with faeces rather than ingestion of contaminated drinking water [33]. In our study, we did not analyse drinking water for the presence of helminth eggs. Therefore, the association between drinking water source and *H. nana* infection has limited biological plausibility, and cannot be inferred.

The findings from univariate and multivariate mixed logistic regression analyses demonstrated a considerably higher risk of *S. haematobium* infection among children reporting exposure to freshwater sources through domestic chores. This result is in accordance with previous studies, showing a higher prevalence *S. haematobium* infection in children observed to play, work or swim in open water bodies that may contain infected snails [37, 38]. Moreover, children from the Plateau Central showed higher odds of *S. haematobium* infection. Even though this association lacked statistical significance ($P = 0.64$), the observation could be explained by the fact that the Plateau Central holds one of the largest water infrastructures in the country: the Ziga dam (capacity of 200 million m³, watershed of Loumbila provided from Nabaouli and Massili Rivers affluent of the Nakambé River, White Volta) and the smaller Loumbila reservoir (36 million m³ storage) on the Massili River. The Ziga dam primarily supplies drinking water to the city of Ouagadougou (70 % of its needs in 2008) [21, 39]. Effective solutions to control infection with schistosomes include education and behaviour change and access to abundant supplies of clean water [40]. Yet, water resources are scarce in Burkina Faso, with an average annual precipitation of 600 to 800 mm in the Plateau Central, where the main water sources used for providing drinking water are derived from the Ziga and Loumbila dam. These dams which are closely located around the project schools, may provide suitable snail habitat and may lead to increased risks for school-aged children, particularly through increased water exposure due to their accessibility [41].

We found a significantly lower prevalence of intestinal pathogenic protozoa (in multivariate) and *H. nana* (in univariate) infections in the Plateau Central compared to the Centre-Ouest regions. However, the urbanization rate in the Plateau Central is 7.9 % as compared to 13.2 % in the Centre-Ouest, both of which are lower than the national average (22.7 %). The Centre-Ouest region, with Koudougou as the third largest city in Burkina Faso, plays an economically important role in trade, agriculture and some mining activities [21]. It is therefore interesting to note that the current study found a higher odds of intestinal parasitic infections for children from the economically more developed Centre-Ouest region, as compared to their counterparts living in peri-urban settings in the Plateau Central. Yet, several other factors may explain this observation. First, in the absence of latrines and consistent availability of sanitary infrastructures at schools and households, children from the Centre-Ouest practised open defaecation more frequently than children from the Plateau Central ($P = 0.02$); this can directly lead to faecal contamination (absence of water and cleansing tissues/paper), and thus exposure to intestinal parasitic infections. This has also been described in a previous study conducted among Kenyan schoolchildren, where the presence of tissue/paper or water for anal cleansing emerged as the

most important predictor of any soil-transmitted helminth infection [42]. Secondly, water quality also significantly differed between the two study regions; water samples from children's drinking water cups and households showed significantly higher contamination with *Escherichia coli* in the Centre-Ouest, as compared to the Plateau Central (all $P < 0.05$). Despite the lack of association of faecal contamination of drinking water to children's parasitic infection status in univariate and multivariate analysis, the presence of faecal coliforms, *Escherichia coli* in water indicates recent faecal contamination and the possible presence of disease-causing pathogens, such as bacteria, viruses and parasites [3, 15, 33]. Lastly, there was a significant difference in reported household water treatment across study regions (higher in the Centre-Ouest compared to the Plateau Central). However, the treatments caregivers reported to use were sedimentation and filtration (with fabric tissue), which may reduce the contents of harmful bacteria but are unlikely to completely remove pathogenic contaminants [33].

While our univariate and multivariate test of associations between schoolchildren's parasitic infection status and household drinking water source, sanitation and water storage lacked statistical significance, the regional differences found in terms of children's sanitary practices and safe drinking water are key for explaining the higher prevalence of children's infection status in the Centre-Ouest. These are most crucial for addressing intestinal parasitic infections in children, in particular for preventing faecal-oral disease transmission [15, 42-44].

The findings of the present study showed that over half of the infected children had polyparasitism and that, on average, a study participant harboured 1.7 intestinal parasite species concurrently. Similar findings were reported among schoolchildren in Côte d'Ivoire and in Kenya, where children were typically infected with an average of two or more species concurrently [45, 46]. We conclude that multiple-species intestinal parasite infections are common in schoolchildren in the Plateau Central and Centre-Ouest of Burkina Faso, partly explained by social-ecological contexts that govern the presence and transmission of intestinal parasitic infections (i.e. climate, proximity to freshwater sources, sanitation and hygiene behaviours) [1, 47].

Lastly, the high prevalence of pathogenic intestinal protozoa infections (75.3 %) compared to that of helminth infections (10.7 %) in this study is in agreement with previous findings in Burkina Faso [17, 34, 48]. Possible reasons for the lower prevalence of faecal-oral transmitted helminths and *Schistosoma* infections among schoolchildren who all had low infection intensity include regular deworming, which reduces both the morbidity caused by these infections and the occurrence of severe complications [49]. The most recent deworming campaign before our

survey in 2014 and the implementation of national deworming campaigns since 2004 must be taken into consideration when interpreting our data. They could explain the low intensity of helminth infections found. However, our findings indicate that despite continuous efforts through regular deworming, transmission in the target area is not interrupted [50-52].

The results presented here are of relevance for the control of intestinal parasitic infection in Burkina Faso, justified on the following grounds. First, school-aged children in this part of Burkina Faso are at considerable risk of infection with helminths and particularly intestinal protozoa, including *E. histolytica/E. dispar* and *G. intestinalis*. Hence, measures to prevent children from infection with pathogenic intestinal protozoa, such as hygiene education, improved access to clean water and sanitation at school, should be promoted, as school-aged children represent the main reservoirs for *E. histolytica* and partly *G. intestinalis* transmission [53]. A challenge for controlling intestinal protozoa is the current lack of rapid diagnostic tests to identify pathogenic species and/or pathogenic strains. Harmless commensal intestinal protozoa species are ubiquitous and often morphologically indistinguishable to pathogens; an accurate diagnosis is therefore central to guide treatment and control of intestinal protozoan infections [54]. Secondly, the burden of disease due to intestinal protozoa infections can be reduced substantially through the improvement of sanitary conditions, adequate excreta disposal, health education and improved hygiene practices [2]. It is, however, unlikely that *E. histolytica/E. dispar* and *G. intestinalis* are eliminated from the environment (cysts are able to survive outside the host for long periods). Thirdly, for this reason, we recommend an integrated control approach to promote water treatment and safe storage. The diversity and integration of different WASH interventions is critical to reduce parasitic intensity, to manage potential risks from pathogenic intestinal protozoa and helminth infections and thus to reduce morbidity in school-aged children [15, 53, 55]. Fourthly, we believe that schools are an ideal entry and outreach point for children and their caregivers to provide deworming treatments and individual treatments for children infected with helminths and intestinal protozoa, respectively. Most importantly, for a long-term success, we believe that treatment strategies targeting intestinal protozoa infections need to be integrated with the current national deworming programme and complemented with a diversity of WASH interventions to gain and sustain benefits by reducing reinfection and transmission of intestinal parasitic infections. Finally, cross-sectoral interventions hold promise to make a lasting impact on intestinal parasitic infections by combining school- and community-based initiatives that go beyond WASH and include education and nutrition interventions. An inter-sectoral approach to prevent and control parasitic infections may also benefit schoolchildren's physical development and educational achievement [56]. The VgtS project provides an opportunity to link

the school garden programme to WASH interventions primarily at schools but also at children's households. A follow-up study conducted after a 12-month intervention period will contribute to understanding the possible effects of these interventions on schoolchildren's health [1]. Lastly, improvements of WASH infrastructure and appropriate health-seeking behaviour are key to achieve sustained control and elimination of NTDs [57, 58]. Our recommendation of improving WASH infrastructure and appropriate health-seeking behaviour as part of the VgtS project in Burkina Faso would also contribute to ways of moving forward with implementing the Sustainable Development Goals (SDGs) agenda, specifically goal number 6 on "ensuring availability and sustainable management of water and sanitation for all" [59].

There are four main study limitations. First, we pursued a cross-sectional survey in February 2015, and hence, our results only reflect one point in time, i.e. the dry season (November to April). We speculate that the prevalence of parasitic infections might be higher in the rainy season (May to September), when children spend more time outside, work in the fields and might eat more frequently unwashed vegetables and fruits from the garden. Seasonal patterns of intestinal parasitic infections may therefore be underestimated [60, 61]. Secondly, as we only examined a single Kato-Katz thick smear and FECT from two stool samples of two consecutive days from each child, we underestimated the true prevalence of parasitic infections, due to the low sensitivity of the Kato-Katz technique and urine concentration method [62, 63]. Thirdly, children's self-reported hygiene behaviours may have resulted in over- or under-reporting of proper hygiene practices [64]. Fourthly, the findings presented here are representative for the selected schools in two regions, but cannot be generalised for all of Burkina Faso.

6.6 Conclusions

This study provides new insight into schoolchildren's parasitic infection status and its associations to household- and school-level WASH conditions among the Plateau Central and Centre-Ouest regions of Burkina Faso. Our findings call for increased public health measures for schoolchildren and rural communities in Burkina Faso. As part of the VgtS project, WASH, and health education interventions should be implemented to reduce transmission and reinfection among schoolchildren. Our data will serve as a benchmark for subsequent post-intervention surveys and analysis.

Abbreviations

aOR: adjusted odds ratio; CI: confidence interval; DHS: Demographic and Health Survey; EKNZ: Ethikkommission Nordwest-und Zentralschweiz; FECT: formalin-ether concentration technique; IRSS: Institute for Health Sciences Research; KAP: knowledge, attitudes and practices; LMICs: low- and middle-income countries; NTD: neglected tropical disease; ONEA: Office National de l'Eau et de l'Assainissement; PAF: population attributable fraction; SD: standard deviation; SDG: Sustainable Development Goal; Swiss TPH: Swiss Tropical and Public Health Institute; VgtS: Vegetables go to School: improving nutrition through agricultural diversification; WASH: water, sanitation and hygiene; WHO: World Health Organization

Declarations

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Ethics approval and consent to participate

Ethical approval for the study protocol was obtained by the “Ethikkommission Nordwest- und Zentralschweiz” in Switzerland (EKNZ, reference no. 2014-161) and by the “Comité d’Ethique pour la Recherche en Santé, Ministère de la Recherche Scientifique et de l’Innovation, et Ministère de la Santé” (reference no. 2015-02-026). The study is registered with the clinical trial registry ISRCTN (identifier: ISRCTN17968589).

Children and their parents/guardians were informed about the purpose and procedures of the study. Written informed consent was obtained from the child’s parents or guardians. For illiterate parents/guardians, a fingerprint was obtained in the presence of a literate witness from the school (principal or teacher), whilst children assented orally. It was emphasised that participation was voluntary and that children could withdraw anytime without further obligation. Those with informed consent were assigned a unique identifier.

Results were communicated to participants and children found infected with any kind of intestinal protozoa or helminths were treated according to national guidelines (i.e. a 15-50 mg/kg single dose of metronidazole for 5 consecutive against intestinal protozoa infection, a triple dose of 400 mg albendazole against soil-transmitted helminth infections, a 40 mg/kg single dose praziquantel against schistosomiasis, and 4 tablets of niclosamide of 500 mg in two doses for 6 consecutive days to treat *H. nana*). All treatments were provided free of charge. Parasitic drugs were administered by trained teachers, in collaboration with our research team, local health personnel and with close involvement of the parents/guardians of infected children, to ensure proper drug intake and observe adverse events.

Consent for publication

Not applicable

Availability of data and material

The dataset supporting the conclusions are not publicly available due to the reason of being PhD study of the first author but are available from the corresponding author on reasonable request. The questionnaires (in French) are available upon request to the corresponding author.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

SE, SD, PO, AMK, CS, JU and GC designed the study; SE, SD, AMK, TG and AK implemented the study; SE managed and analysed the data and wrote the first draft of the paper; SD, PO, AMK and CS contributed to data analysis and helped interpret the results; JG, CS, JU and GC revised the manuscript and provided important intellectual content. All authors read and approved the final version of the manuscript before submission.

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7 School children's intestinal parasite and nutritional status 1 year after complementary school garden, nutrition, water, sanitation, and hygiene interventions in Burkina Faso

Running head: Children's health and nutrition, Burkina Faso

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7.1 Abstract

The potential health benefits of combined agricultural, nutrition, water, sanitation, and hygiene (WASH) interventions are poorly understood. We aimed to determine whether complementary school garden, nutrition, and WASH interventions reduce intestinal parasites and improve school children's nutritional status in two regions of Burkina Faso. A cluster-randomized controlled trial was conducted in the Plateau Central and Centre-Ouest regions of Burkina Faso. A total of 360 randomly selected children, aged 8-15 years, had complete baseline and end-line survey data. Mixed regression models were utilized to assess the impact of the interventions, controlling for baseline characteristics. The prevalence of intestinal parasitic infections decreased both in intervention and control schools, but the decrease was significantly higher in the intervention schools related to the control schools (odds ratio (OR) of the intervention effect = 0.2, 95% confidence interval (CI) 0.1-0.5). Indices of undernutrition did not decrease at end-line in intervention schools. Safe handwashing practices before eating and the use of latrines at schools were significantly higher in the intervention schools than in the control schools at end-line (OR = 6.9, 95% CI 1.4-34.4, and OR = 14.9, 95% CI 1.4-153.9, respectively). Parameters of water quality remained unchanged. A combination of agricultural, nutritional, and WASH-related interventions embedded in the social-ecological systems and delivered through the school platform improved several child health outcomes, including intestinal parasitic infections and some WASH-related behaviors. Sustained interventions with stronger household and community-based components are, however, needed to improve school children's health in the long-term.

7.2 Introduction

Undernutrition and intestinal parasitic infections remain considerable health issues among school-aged children in Burkina Faso.¹⁻⁴ There are far-reaching negative consequences of undernutrition and ill-health among children, affecting their physical well-being and educational potentials, which undermine social, political, and economic benefits for communities as a whole.⁵

School children's nutritional status and prevalence of intestinal parasitic infections are governed by social-ecological systems, since these health conditions are influenced by human behavior (e.g., dietary practices, open defecation, unsafe hygienic practices, and patterns of unprotected surface water contacts) and ecological characteristics (e.g., agricultural systems and access to clean water).^{6, 7} Undernutrition and intestinal parasitic infections are closely interlinked and share several common risk factors, including a lack of access to clean water, improved sanitation, and adequate hygiene (WASH).⁸⁻¹⁰ Chronic exposure to a contaminated environment due to unsafe WASH conditions (e.g., to feces contaminated with protozoan cysts or helminth eggs) can cause diarrhea or asymptomatic infection;¹¹ which in turn can lead to loss of nutrients, malabsorption, impaired digestion, and ultimately decline childhood growth.¹²⁻¹⁴ It follows that multi-sectoral programs are crucial to address child undernutrition and disease-related causes and consequences.¹⁵ Schools are an ideal entry point for multi-sectoral agriculture, nutrition, and WASH programs.¹⁶ Besides being an obvious place to educate children on healthy diets, schools can promote practical and positive changes in personal hygiene, nutrition, and health by: (1) increasing food availability and diversity with school gardens¹⁷; (2) offering well-balanced and nutritious meals through a school feeding program (in which parts of the garden produce could be used);¹⁸ and (3) promoting handwashing with soap and safe sanitary behaviors.^{16, 19} Yet, there is scarce evidence of the effects of school-based programs on school children's intestinal parasitic infection and nutritional status.^{20, 21} There is also insufficient evidence of combined approaches across the nutrition, health, agriculture, education, and WASH sectors addressing proximate and underlying determinants of undernutrition in children.^{16, 22-26}

To address this issue, a multi-sectoral project entitled "Vegetables go to School: improving nutrition through agricultural diversification" (VgtS) was developed in five countries to determine school children's health in face of implementing school vegetable gardens and other school-based health, nutritional, and environmental interventions.²⁷ As part of the VgtS project, a cluster-randomized controlled trial was implemented in Burkina Faso. Here, we report findings on the impacts of complementary school garden, nutrition, and WASH interventions on school children's intestinal parasitic infections and nutritional status, including WASH-related behaviors to discuss the findings along the hypothesis of the program impacts.

7.3 Material and methods

Ethical considerations

Data reported here stem from a cluster-randomized controlled trial that has been registered with the clinical trial registry ISRCTN (identifier: 17968589). The study protocol was approved by the “Ethikkommission Nordwest- und Zentralschweiz” (EKNZ) in Switzerland (reference no. 2014-161) and by the “Comité d’Ethique pour la Recherche en Santé, Ministère de la Recherche Scientifique et de l’Innovation, et Ministère de la Santé” in Burkina Faso (reference no. 2015-02-026).

Parents or guardians of children were asked for written informed consent (fingerprint for illiterate parents/guardians), while children assented orally. Study participation was voluntary, and hence, children could withdraw anytime without further obligation. Results were communicated to all participants. Specific treatments against parasitic infections were provided free of charge. Mildly and moderately anemic children (hemoglobin (Hb) < 11.5 g/dL for children aged 8-11 years and Hb < 12 g/dL for children aged 12-14 years, including girls aged 15 years, and Hb < 13 g/dL for boys aged 15 years) were referred to a local health center and treated with iron supplements for 40 days. Children found with severe anemia (Hb < 8 g/dL) and severely malnourished children were referred to a local health center for further investigation, following national guidelines.^{28, 29} The Consolidated Standards of Reporting Trials (CONSORT) guidelines were applied to report the results of this study.^{30, 31} The CONSORT checklist is provided as supplemental information (see Supplemental Table 1 in Appendix **Error! Reference source not found..1**).

Complementary school garden, nutrition and WASH interventions

The interventions consisted of four main components. The first component included the provision of seeds and small gardening tools and agricultural trainings given to 12 teachers and four school directors for the school garden activities, which commenced in early 2015. The second component consisted of WASH interventions at schools with several sub-components: (i) installation of latrines; (ii) rehabilitation of water pumps; (iii) installation of handwashing stations and toolkits to make soap; and (iv) installation of safe drinking water stations in classrooms. The third component entailed the educational behavior change strategy provided to (i) teachers and school directors with materials developed for teaching in classroom 1-2 times a week starting in October 2015, and to (ii) community representatives (in total 16) with monthly trainings at schools on hygiene and nutrition launched in November 2015. The fourth component consisted of providing treatments to children found anemic or infected with intestinal parasites (i.e., 15-50 mg/kg single dose of metronidazole for five consecutive days against intestinal protozoa infection, a triple dose of 400 mg albendazole against soil-transmitted

helminth infections, a 40 mg/kg single dose of praziquantel against schistosomiasis, and 4 tablets of niclosamide of 500 mg in two doses for 6 consecutive days to treat *Hymenolepis nana*) in both intervention and control schools, following national guidelines.^{28, 29} All program components were fully implemented within seven months of the end of the baseline survey.

Sample size, sampling method, and study design

The study was originally designed as cross-sectional baseline survey with 85% power to detect a difference in the prevalence of intestinal parasitic infection rates (with $P < 0.05$) as primary outcome measure in the comparison between high- and low-risk children at eight schools for a true odds ratio (*OR*) of at least 2 with a coefficient of variation of 10% in $\ln(\text{OR})$ across schools. A Monte Carlo simulation (5000 iterations) led to a minimal sample size of 400 children aged 8–14 years, assuming a prevalence of intestinal parasitic infections of 40%, a coefficient of variation of 10% across schools and a proportion of high-risk children of 25%. The eight schools to participate in this study were randomly selected from the 30 VgtS project schools in Burkina Faso. At baseline, 55–60 children (boys and girls in ratio 1:1) were randomly selected in each of the sampled schools assuming a 15% drop-out rate. The eligibility criteria for children to participate at baseline were: (i) school children aged between 8 and 14 years; (ii) parents/caregivers of the children providing written informed consent; and (iii) children providing oral assent.

This study reports a secondary analysis of a sample of children followed over one year to assess and compare individual and cluster effects of a package of health interventions. There were eight schools included in a baseline cross-sectional survey. The schools were randomly and evenly allocated by the study investigators to two study arms (“intervention” and “control” group). Four schools were part of the intervention group: two schools in the Plateau Central region, and two schools in the Centre-Ouest region (Figure 7.1). Four schools served as controls; with two schools in each of the respective regions. In order to control for the effect of seasonal fluctuations on specific health conditions, the two surveys were spaced by approximately 1 year (February 2015 and March 2016).

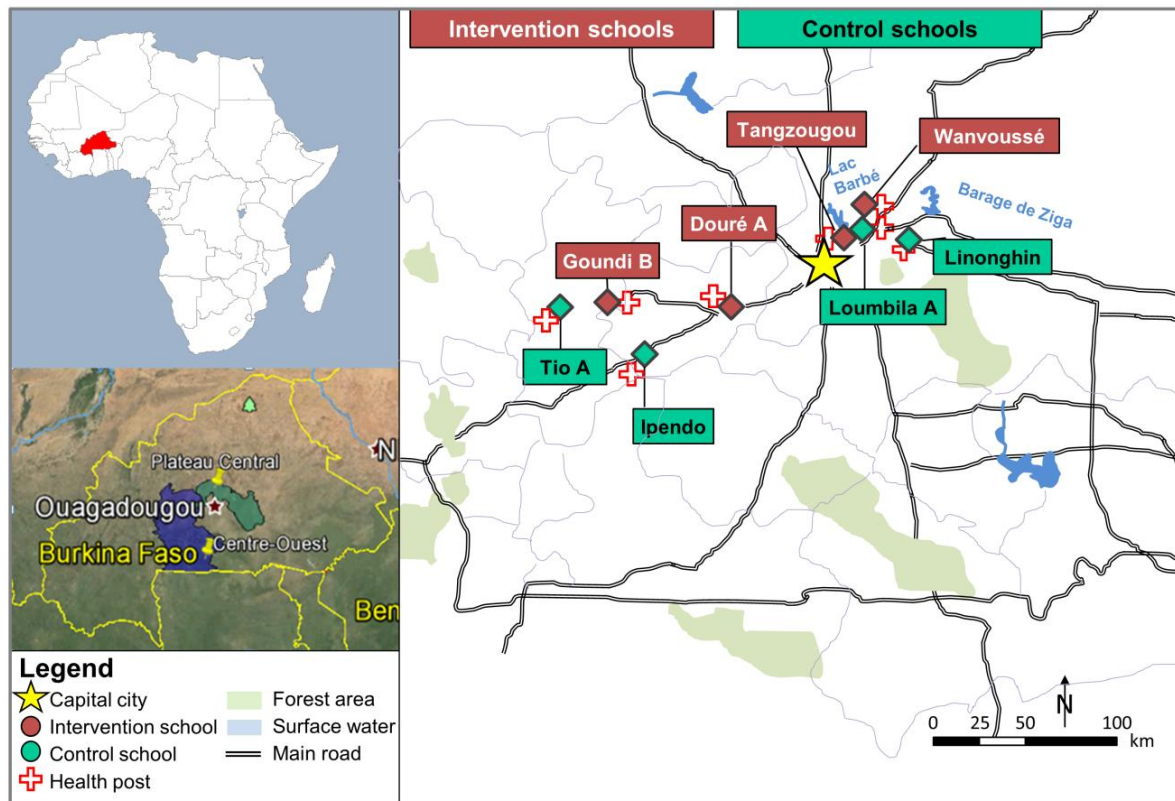


Figure 7.1: Study sites of the cluster-randomized controlled trial in a cohort of children in two regions of Burkina Faso, February/March 2015 and one year later

Outcome definition and measurement

The study measured outcomes using a combination of child anthropometry, specimen (stool and blood) testing, sampling and testing of drinking water, and structured questionnaires. Training and all field activities were overseen by the study investigators (AMK, SD, SE). The baseline survey was conducted between February 2 and 19, 2015, and the end-line survey was conducted between February 15 and March 2, 2016. The same field and laboratory procedures were employed in the baseline and end-line surveys, which have been described in detail elsewhere.^{3, 27}

Main outcomes were defined and measured as follows. In a first step, children's weight and height were measured following standard procedures.³² Second, Hb concentration was determined using a HemoCue® 201+ testing device (HemoCue Hb 201 System; HemoCue AB, Ängelholm, Sweden).³³ Third, a single stool sample was collected from each child on two consecutive days, subjected to the Kato-Katz technique (duplicate thick smears, using standard 41.7 mg templates) and a formalin-ether concentration technique (FECT) for the diagnosis of helminths and intestinal protozoa.^{34, 35} Urine samples were examined for microhematuria using reagent strips (Hemastix, Siemens Healthcare Diagnostics GmbH; Eschborn, Germany). A urine

filtration method was employed to examine urine samples under a microscope for the presence and number of *Schistosoma haematobium* eggs.³⁶ Helminth infection intensity was according to World Health Organization (WHO) criteria.³⁷ Fourth, drinking water samples from the same cohort of children and households as at baseline were analyzed for the presence of bacterial indicators of fecal contamination, using the membrane filtration technique.³⁸ Fifth, a questionnaire was administered at schools and households using tablets (Samsung Galaxy note 10.1 N8010) to investigate children's health knowledge, attitudes, and practices (KAP) and household socioeconomic characteristics,^{2, 27, 39} using open data kit (ODK) software.⁴⁰

Statistical analysis

Data were double-entered into Excel, version 2010 (Microsoft Corp.; Redmond, WA). Anthropometric indices (i.e., stunting (low height-for-age), thinness (low body mass index [BMI]-for-age) and underweight (low weight-for-age)) were calculated with the WHO reference for children aged 5-19 years, using AnthroPlus, version 1.0.4 (WHO; Geneva, Switzerland). Undernutrition was defined as a summary measure including any of these three nutritional indices defined as z-score < -2. Children were classified as overweight if BMI-for-age z-score was > 1.⁴¹

Three types of questions were addressed with different statistical models. Step 1: cross-sectional estimates of prevalences and their differences between study groups were assessed with logistic regression analyses. Baseline and end-line prevalences were computed, including 95% confidence intervals (CIs) for each of the study groups, without adjusting for potential confounders. Robust variance estimates were used to take into account clustering within schools. In parallel, a mixed logistic regression model with random intercepts for schools was used to compare baseline and end-line prevalences between intervention and control schools. As our study groups significantly differed in terms of children's age, caregiver's educational attainment and economic characteristics, we also conducted a factor analysis to characterize household socioeconomic status (SES) from a list of recorded household assets, housing materials, main energy sources used, and caregiver's educational achievement.⁴² Two factors reflecting household SES were retained. Each factor score was then categorized into tertile classes. The following analyses were run with additional adjustments for the two categorical SES variables and age of participating children.

Step 2: changes in prevalence from baseline to end-line were estimated and the differences between study groups assessed using mixed logistic regression models with binary baseline and end-line outcomes as repeated observations. These models included random intercepts for schools and children, the aforementioned variables, the fixed factors period and study group, as well as the interaction between the two fixed factors. Intervention effects on prevalence changes were measured by the odds ratio (OR) of this interaction. Mixed linear regression models with

random intercepts for schools, the factor study group and adjustments for SES and children's age were applied to assess intervention effects on the changes in continuous variables (e.g., weight and height).

Step 3: incidence and persistence of adverse health outcomes (i.e., indices of undernutrition or intestinal parasite infections) among children with or without the respective outcome at baseline were assessed using mixed logistic regression models. These models included the factor study group, the two SES variables and children's age, with random intercepts at the level of schools (see Supplemental Tables 2 and 3 in Appendix **Error! Reference source not found.** and **Error! Reference source not found.**). Statistical significance was defined at a level of 5%. Statistical analyses were conducted with STATA, version 13.0 (Stata Corporation; College Station, TX).

7.4 Results

Compliance and characteristics of study population

Complete datasets were available for 385 children and the equivalent of parents/caregivers at baseline. Overall, 25 children were lost to follow-up. The final analysis included 360 children and caregivers; 176 in intervention and 184 in control schools with complete datasets (Figure 7.2).

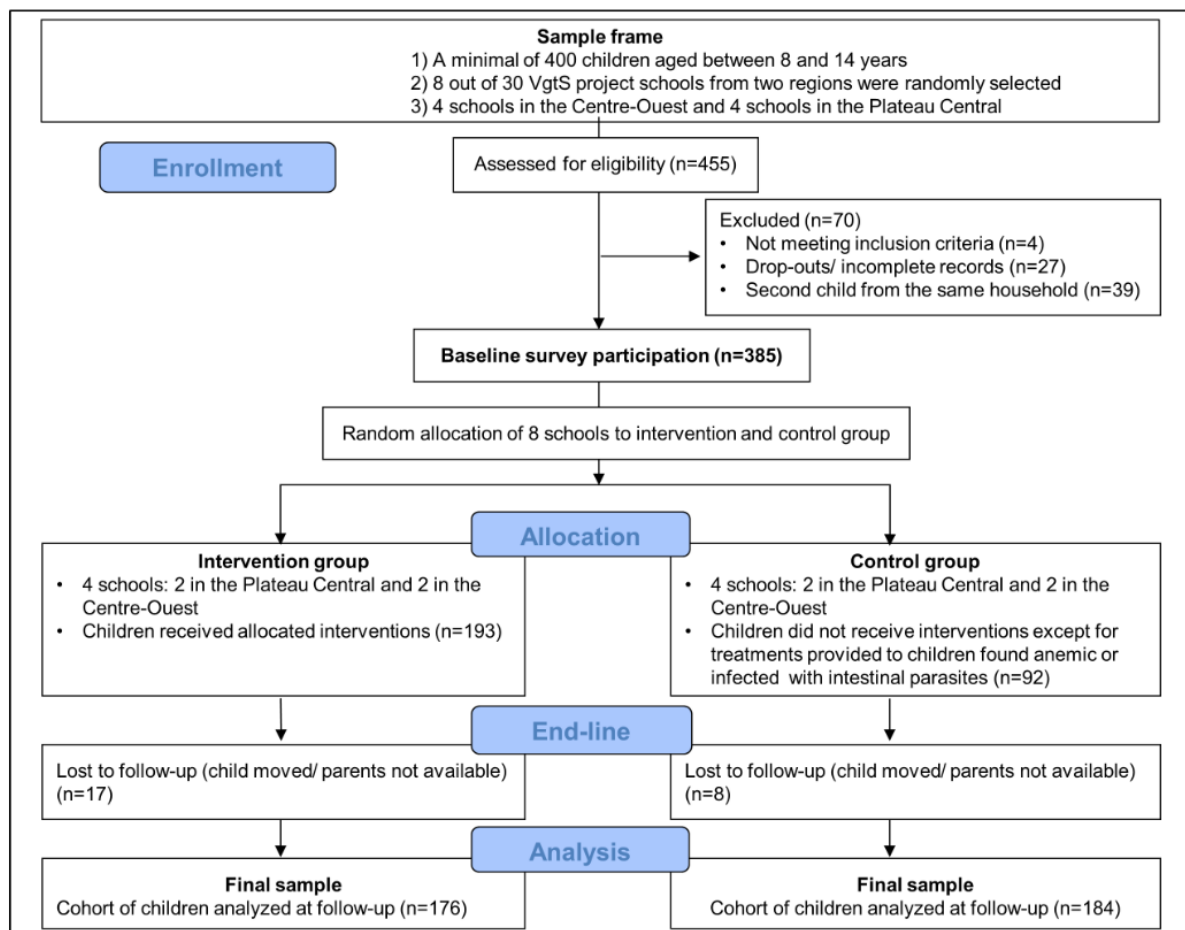


Figure 7.2: CONSORT flowchart: enrollment, intervention allocation, and end-line analysis

Key characteristics of children and their households included in the study sample are shown in Table 1, stratified by study arm. Sociodemographic and economic characteristics between the two groups were similar, with the exception of parents/caregivers' educational attainment and their housing characteristics (roof material), which were significantly lower in the intervention group ($P = 0.001$ and $P = 0.043$, respectively) (Table 7.1).

Table 7.1: Characteristics of the study cohort in the two regions of Burkina Faso at the baseline survey in February/March 2015

	Total	Intervention schools	Control schools	P-value	
Children's demographic characteristics*	n [%]	n [%]	n [%]		
Age of children†				0.052	
Girls	178 (49.4)	85 (48.3)	93 (50.5)	0.670	
Boys	182 (50.6)	91 (51.7)	91 (49.5)		
Age group 1 (8-11 years)	233 (64.7)	95 (54.0)	138 (75.1)	0.095	
Age group 2 (12-14 years)	127 (35.3)	81 (46.0)	46 (25.0)		
Caregivers' demographic and education characteristics					
Caregiver's age‡				0.111	
No formal schooling	268 (74.4)	145 (82.4)	123 (66.9)	0.001	
Primary education	57 (15.8)	21 (11.9)	36 (19.6)		
Secondary or higher education	35 (9.7)	10 (5.7)	25 (13.6)		
Main occupation of head of household					
Agriculture	324 (90.0)	157 (89.2)	167 (90.8)	0.673	
Merchant	6 (1.7)	1 (0.6)	5 (2.7)		
Civil service	9 (2.5)	6 (3.4)	3 (1.6)		
Others (housework, retirement or no employment)	21 (5.8)	12 (6.8)	9 (4.9)		
Socioeconomic domains					
Roof material	Simple (natural and baked clay)	328 (89.7)	168 (95.4)	160 (87.0)	0.043
	Metal cover	32 (10.3)	8 (4.6)	24 (13.0)	
Wall material	Simple (natural clay)	337 (93.6)	166 (94.3)	171 (92.9)	0.923
	Baked or cemented clay	23 (6.4)	10 (5.7)	13 (7.1)	
Floor material	Simple (clay, sand, mud, straw)	241 (66.9)	114 (64.8)	127 (69.0)	0.506
	Baked or cemented clay	119 (33.1)	62 (35.2)	57 (31.0)	
Energy used	Simple (charcoal, firewood)	352 (97.8)	176 (98.9)	178 (96.7)	0.417
	Electricity and gas	8 (2.2)	2 (1.1)	6 (3.3)	

*Mixed linear models were used to compare age and mixed logistic and mixed ordinal regression models with random intercepts at the level of schools to compare binary and ordinal variables, respectively. Statistical significance was defined at a level of 5% (bold values where $P < 0.05$).

†Mean age of 11.0 (± 1.4) years; 11.4 (± 1.3) years in the intervention schools and 10.6 (± 1.4) years in the control schools.

‡Mean age of 44.9 (± 14.0) years; 46.4 (± 14.3) years in the intervention schools and 43.5 (± 13.7) years in the control schools.

Changes of intestinal parasitic infections in children

At baseline, children in intervention schools showed a higher prevalence of total parasite, total intestinal protozoa, and total helminth infections than children in control schools ($P = 0.031$, $P = 0.050$, and $P = 0.807$, respectively). We observed declines of intestinal protozoa infections from 88.6% to 57.4% in the intervention schools and from 79.9% to 70.1% in control schools with an intervention effect (OR = 0.2, 95% CI 0.1-0.5, $P < 0.001$). Total helminth infections decreased in intervention schools (from 11.4% in 2015 to 8.0% in 2016), while it was stable in control schools (9.8% in 2015, 10.3% in 2016). These changes were not significantly different (OR = 0.5, 95% CI 0.1-1.7, $P = 0.265$) (Table 7.2). Of note, when stratifying the analyses of the change in

intestinal parasitic infections by risk factors at baseline (i.e., stunting, thinness, and anemia), the intervention effects were slightly higher among children not being stunted or anemic; however, the differences lacked statistical significance ($P > 0.2$).

Table 7.2: Changes of intestinal parasitic infections in a cohort of school children in two regions of Burkina Faso, in February/March 2015 and one year later

		Intervention schools		Control schools		Intervention effect§
		Baseline (2015)	End-line (2016)	Baseline (2015)	End-line (2016)	
Sample size (n)		176	176	184	184	
Total intestinal parasites‡‡	Prevalence†	90.3 (80.0, 95.6)	61.9 (51.6, 71.3)	81.5 (70.0, 89.3)	72.3 (59.8, 82.1)	0.2 (0.1, 0.5)**
	OR‡		0.1 (0.1, 0.3)***		0.6 (0.3, 1.0)	
Total intestinal protozoa	Prevalence†	88.6 (80.8, 93.5)	57.4 (43.2, 70.5)	79.9 (66.7, 88.7)	70.1 (56.1, 81.2)	0.2 (0.1, 0.5)***
	OR‡		0.1 (0.1, 0.3)***		0.6 (0.3, 1.0)	
<i>Entamoeba histolytica/E. dispar</i>	Prevalence†	69.9 (58.6, 79.2)	36.9 (29.8, 44.6)	62.5 (42.1, 79.3)	47.8 (33.2, 62.8)	0.5 (0.2, 0.9)*
	OR‡		0.2 (0.1, 0.4)***		0.5 (0.3, 0.8)**	
<i>Giardia intestinalis</i>	Prevalence†	30.1 (22.1, 39.5)	25.0 (15.9, 37.0)	26.6 (23.2, 30.4)	25.0 (15.7, 37.3)	0.8 (0.4, 1.7)
	OR‡		0.8 (0.4, 1.3)		1.0 (0.6, 1.7)	
<i>Trichomonas intestinalis</i>	Prevalence†	27.8 (22.4, 34.1)	11.9 (5.6, 26.7)	19.6 (15.5, 24.3)	13.0 (9.4, 17.8)	0.5 (0.2, 1.2)
	OR‡		0.3 (0.2, 0.6)***		0.6 (0.3, 1.0)	
<i>Entamoeba coli</i>	Prevalence†	34.1 (28.3, 40.4)	15.9 (9.0, 26.5)	40.8 (35.8, 45.9)	26.1 (19.7, 33.7)	0.7 (0.3, 1.4)
	OR‡		0.3 (0.2, 0.6)***		0.5 (0.3, 0.8)**	
Total helminths	Prevalence†	11.4 (5.9, 20.7)	8.0 (3.5, 17.2)	9.8 (3.6, 23.8)	10.3 (5.7, 18.1)	0.5 (0.1, 1.7)
	OR††		0.5 (0.2, 1.3)		1.1 (0.5, 2.6)	
<i>Hymenolepis nana</i> ¶	Prevalence†	7.4 (4.1, 12.9)	5.1 (2.1, 12.0)	6.0 (1.4, 21.9)	5.4 (2.3, 12.5)	0.7 (0.2, 2.7)
	OR††		0.7 (0.3, 1.6)		0.9 (0.4, 2.2)	
<i>Schistosoma haematobium</i>	Prevalence†	4.0 (1.3, 11.9)	2.8 (0.7, 10.7)	2.7 (0.4, 14.8)	4.9 (1.8, 12.5)	0.3 (0.1, 1.9)
	OR††		0.7 (0.2, 2.3)		2.0 (0.6, 6.9)	

CI = confidence interval; EPG = eggs per gram of stool; OR = odds ratio; SES = socioeconomic status.

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

† Data are % (95% CI). The CIs are adjusted for clustering within schools by using robust standard errors.

‡ ORs refer to the period effects. Mixed logistic regression models were adjusted for the two categorical SES variables and children's age.

§ ORs refer to the intervention effect defined as ratio between the period effects in intervention and in control schools including random intercepts for schools and children and with adjustment for SES and children's age.

¶ The mixed logistic regression model did not include random intercepts for children due to the low number of children with the respective outcome.

|| Triple interactions involving the factors period and survey arm along with one of the additional variables sex, age group, and prevalence of adverse health outcomes at baseline (i.e., being stunted, thin, and anemic) were also tested in mixed logistic regression models with random intercepts for schools and children, adjusted for the two categorical SES variables. The only significant triple interaction was found for age group, where the intervention effect was significantly greater in children aged 9–12 years (OR = 0.1, 95% CI 0.0–0.9, $P = 0.041$).

†† The mixed logistic regression model was not adjusted for SES variables or children's age, as no convergence in the regression models were achieved.

‡‡ Three children were infected with hookworm, one with *Balantidium coli*, and one with *Schistosoma mansoni* at baseline. None of these parasite species were found one year later. One child was found infected with *Entamoeba hartmanni* in the end-line survey. All helminth infection prevalences were of low intensity at baseline (*S. mansoni* 1–99 EPG; *Hymenolepis nana* and hookworm 1–1,999 EPG, *Schistosoma haematobium* < 50 eggs/10 mL of urine). At end-line, one case of moderate and one heavy *H. nana* infection (2,000–9,999 EPG, and $\geq 10,000$ EPG, respectively) were found; as well as two cases of heavy *S. haematobium* infection (≥ 50 eggs/10 mL of urine, 0.6%).

Changes of anthropometric indices and anemia in children

The rates of undernutrition, stunting, and thinness were slightly higher in intervention schools compared to control schools at baseline, but the difference showed no statistical significance (all $P > 0.05$). At the end-line survey, stunting and thinness were both higher in the intervention schools (38.1% in 2015, 42.0% in 2016 for stunting; 12.5% in 2015, 14.8% in 2016 for thinness) and in the control schools (23.4% in 2015, 26.1% in 2016 for stunting; 10.9% in 2015, 12.5% in 2016 for thinness) compared to the baseline prevalences. Overweight decreased in intervention schools from 1.1% in 2015 to 0.6% in 2016 and increased in control schools from 3.3% in 2015 to 5.4% in 2016. However, no statistically significant intervention effect on any of the nutritional indices, including children's weight or height gain, was found. Anemia increased in both intervention (from 30.7% in 2015 to 35.8% in 2016) and control schools (from 26.6% in 2015 to 37.0% in 2016) over the course of the study, but the changes were not significantly different (Table 7.3).

Table 7.3: Changes of nutritional indicators in the study cohort in two regions of Burkina Faso, in February/March 2015 and one year later

		Intervention schools		Control schools		Intervention effect¶
		Baseline (2015)	End-line (2016)	Baseline (2015)	End-line (2016)	
Sample size (n)		176	176	184	184	
Logistic models (binary outcomes)						OR (95% CI)
Total undernutrition†	Prevalence‡	43.2 (34.8, 52.0)	46.0 (37.9, 54.4)	30.4 (17.4, 47.5)	33.7 (18.8, 52.8)	0.9 (0.3, 3.9)
	OR‡‡		1.5 (0.7, 3.5)		1.7 (0.7, 3.9)	
Stunting (low height-for-age)	Prevalence‡	38.1 (27.6, 49.8)	42.0 (33.6, 51.0)	23.4 (11.7, 41.3)	26.1 (13.3, 44.7)	1.2 (0.6, 2.3)
	OR§		0.8 (0.5, 1.2)		0.6 (0.4, 1.1)	
Thinness (low BMI-for-age)	Prevalence‡	12.5 (9.7, 16.0)	14.8 (9.4, 22.4)	10.9 (6.2, 18.3)	12.5 (7.7, 19.6)	1.1 (0.4, 2.8)
	OR§		0.8 (0.4, 1.6)		0.8 (0.4, 1.5)	
Underweight (low weight-for-age)	Prevalence‡	0.6 (0.1, 3.3)	0.0 (0)	1.1 (0.4, 3.2)	0.0 (0)	n/a
	OR§		n/a		n/a	
Overweight (high BMI-for-age)	Prevalence‡	1.1 (0.2, 6.6)	0.6 (0.0, 3.3)	3.3 (1.0, 9.5)	5.4 (2.1, 13.6)	0 (0.0, 4.0)
	OR§		0.3 (0.0, 8.0)		5.6 (0.4, 71.0)	
Anemia††	Prevalence‡	30.7 (25.4, 36.5)	35.8 (26.2, 46.7)	26.6 (21.6, 32.4)	37.0 (25.8, 49.7)	0.7 (0.4, 1.5)
	OR§		1.0 (0.6, 1.7)		1.4 (0.8, 2.4)	
Linear models (continuous outcomes)						Δ-change (95% CI)§§
Change in height-for-age (stunting)						0.00 (-0.07, 0.08)¶¶
Change in BMI-for-age (thinness)						0.05 (-0.08, 0.17)
Height gain (cm)						0.02 (-0.04, 0.09)¶¶
Weight gain (kg)						0.03 (-0.05, 0.11)¶¶
Change in hemoglobin level (g/dL)						-0.17 (-0.36, 0.02)¶¶

BMI = body mass index; CI = confidence interval; Hb = hemoglobin; n/a = not applicable; OR = odds ratio; SES = socioeconomic status.

† The category of total undernutrition includes all children classified as stunted (low height-for-age), thin (low BMI-for-age) or underweight (low weight-for-age) with z-scores < -2.

‡ Data are % (95% CI). The CIs are adjusted for clustering within schools by using robust standard errors.

§ ORs refer to the period effects. Mixed logistic regression models including random intercepts for schools and children were adjusted for the two categorical SES variables and children's age.

¶ ORs refer to the intervention effect defined as ratio between the period effects in intervention and in control schools including random intercepts for schools and children and with adjustment for SES variables and children's age.

|| The mixed logistic regression model did not include random intercepts for children due to the low number of children with the respective outcome.

†† The category of anemia includes all children classified as anemic (mild, moderate, and severe) based on the concentrations of Hb determined in a finger prick blood sample. The cut-offs for anemia are age-specific: Hb < 11.5 g/dL for children aged 8-11 years, and Hb < 12 g/dL for children aged 12-14 years, including for girls aged 15 years, and Hb < 13 g/dL for boys aged 15 years.

‡‡ The mixed logistic regression model was not adjusted for SES variables or children's age, as no convergence in the regression models was achieved.

§§ Mixed linear regression models including random intercepts for schools were adjusted for SES variables and children's age. The Δ-change stands for the estimated effect of the intervention on the mean of the respective change with the 95% CI.

¶¶ The mean changes of weight (0.89; 0.46, 1.33, $P < 0.001$), height (0.65; 0.17, 1.14, $P = 0.008$), height-for-age z-score (0.17; 0.09, 0.26, $P < 0.001$) and Hb (0.31; 0.02, 0.60, $P = 0.034$) were significantly larger in girls than in boys.

Changes of fecal contamination in drinking water

Escherichia coli-positive samples from households significantly decreased, both in intervention sites (OR = 0.3, 95% CI 0.1-1.0, $P = 0.049$) and control sites (OR = 0.2, 95% CI 0.1-0.7, $P = 0.015$). There was a significant decrease in household drinking water samples contaminated with fecal streptococci in intervention sites (OR = 0.1, 95% CI 0.0-0.6 $P = 0.011$), while the decline was less pronounced in control sites (OR = 0.2, 95% CI 0.0-1.1, $P = 0.068$). Samples contaminated with fecal streptococci from children's drinking water cups also significantly decreased in intervention sites (OR = 0.2, 95% CI 0.1-0.7, $P = 0.007$), while the change in control sites lacked statistical significance (OR = 0.3, 95% CI 0.1-1.4, $P = 0.136$). No statistically significant differences were observed between intervention and control sites for any of the water quality parameters (Table 7.4).

Table 7.4: Changes of drinking water contamination in a subsample of households and children's drinking water samples in two regions of Burkina Faso, in February/March 2015 and one year later

		Intervention sites		Control sites		Intervention effect ^c
		2015	2016	2015	2016	
Sample size (n)	Households	46	46	45	45	
	Children's cups	54	54	53	53	
	Study sites	4	4	4	4	
Water contamination households						
Coliform bacteria	Prevalence ^a	93.5 (80.6, 98.0)	95.7 (88.9, 98.4)	95.6 (87.4, 98.5)	95.6 (77.0, 99.3)	1.5 (0.1, 23.3)
	OR ^d	1.0	1.5 (0.2, 9.6)	1.0	1.0 (0.1, 7.4)	
<i>Escherichia coli</i>	Prevalence ^a	56.5 (37.0, 74.2)	34.8 (24.1, 47.3)	71.1 (53.4, 84.1)	46.7 (28.5, 65.7)	1.6 (0.3, 7.3)
	OR ^b	1.0	0.3 (0.1, 1.0)*	1.0	0.2 (0.1, 0.7)*	
Fecal streptococci	Prevalence ^a	95.7 (88.9, 98.4)	76.1 (50.5, 90.8)	91.1 (69.2, 97.9)	77.8 (62.7, 87.9)	0.4 (0.1, 4.0)
	OR ^b	1.0	0.1 (0.0, 0.6)*	1.0	0.2 (0.0, 1.1)	
Water contamination children's drinking water cups						
Coliform bacteria	Prevalence ^a	90.7 (84.0, 94.8)	81.5 (66.6, 90.7)	88.7 (68.9, 96.5)	90.6 (83.6, 94.8)	0.3 (0.1, 2.0)
	OR ^d	1.0	0.4 (0.1, 1.4)	1.0	1.2 (0.3, 4.5)	
<i>Escherichia coli</i>	Prevalence ^a	42.6 (19.8, 69.0)	25.9 (12.4, 46.4)	52.8 (37.4, 67.7)	30.2 (15.4, 50.7)	1.1 (0.3, 3.8)
	OR ^b	1.0	0.4 (0.2, 1.1)	1.0	0.4 (0.2, 1.0)	
Fecal streptococci	Prevalence ^a	87.0 (78.7, 92.4)	61.1 (49.8, 71.4)	90.6 (74.5, 96.9)	77.4 (51.3, 91.7)	0.6 (0.1, 3.2)
	OR ^b	1.0	0.2 (0.1, 0.7)**	1.0	0.3 (0.1, 1.4)	

CI = confidence interval; OR = odds ratio; SES = socioeconomic status.

* $P < 0.05$, ** $P < 0.01$.

† Data are % (95% CI). The CIs are adjusted for clustering within schools by using robust standard errors.

‡ ORs refer to the period effects. Mixed logistic regression models were adjusted for the two categorical SES variables and children's age.

§ ORs refer to the intervention effect defined as ratio between the period effects in intervention and in control schools, with adjustment for SES variables and children's age.

¶ The mixed logistic regression model was not adjusted for SES variables or children's age, as no convergence in the regression models was achieved.

Changes of health KAP

Handwashing after playing and after defecation significantly increased in intervention schools (OR = 5.7, 95% CI 2.6-12.2, $P < 0.001$ for handwashing after playing, OR = 7.4, 95% CI 3.9-14.1, $P < 0.001$ for handwashing after defecation) and in control schools (OR = 3.1, 95% CI 1.4-6.8, $P = 0.004$ for handwashing after playing, OR = 3.6, 95% CI 2.0-6.5, $P < 0.001$ for handwashing after defecation). A significant beneficial intervention effect was found for handwashing before eating (OR = 6.9, 95% CI 1.4-34.4, $P = 0.018$) and the use of latrines at schools (OR = 14.9, 95% CI 1.4-153.9, $P = 0.024$) (Table 7.5).

Table 7.5: Changes in key indicators from the health questionnaire in a cohort of children in two regions of Burkina Faso, in February/March 2015 and one year later

		Intervention schools		Control schools		Intervention effect¶
		Baseline (2015)	End-line (2016)	Baseline (2015)	End-line (2016)	
Sample size (n)		176	176	184	184	
Selected KAP† indicators						
Before eating	Prevalence‡	82.4 (59.6, 93.7)	97.7 (95.7, 98.8)	93.5 (89.7, 95.9)	96.2 (92.1, 98.2)	6.9 (1.4, 34.4)*
	OR§		13.2 (3.5, 49.6)***		1.9 (0.7, 5.4)	
After playing	Prevalence‡	6.3 (3.0, 12.7)	25.0 (21.5, 28.9)	8.2 (5.3, 12.3)	15.8 (10.6, 22.7)	1.8 (0.6, 5.1)
	OR§		5.7 (2.6, 12.2)***		3.1 (1.4, 6.8)**	
After eating	Prevalence‡	11.9 (8.8, 16.0)	17.0 (9.0, 29.8)	16.8 (9.8, 27.5)	12.5 (7.8, 19.4)	1.8 (0.7, 4.9)
	OR§		1.2 (0.6, 2.5)		0.7 (0.3, 1.4)	
After defecation	Prevalence‡	20.5 (14.1, 28.6)	52.8 (44.5, 61.0)	22.8 (18.8, 27.4)	45.1 (37.1, 53.4)	2.1 (1.0, 4.5)
	OR§		7.4 (3.9, 14.1)***		3.6 (2.0, 6.5)***	
Do not wash hands	Prevalence‡	8.0 (2.0, 27.2)	0.6 (0.1, 3.3)	0 (0.0)	0 (0.0)	n/a
	OR§		0.1 (0.0, 0.8)**		n/a	
With water only	Prevalence‡	92.0 (89.2, 94.2)	90.9 (86.5, 94.0)	87.0 (76.1, 93.3)	88.0 (72.9, 95.3)	0.8 (0.3, 2.3)
	OR§		1.1 (0.5, 2.6)		1.4 (0.7, 3.1)	
With water and soap	Prevalence‡	73.9 (53.7, 87.3)	78.4 (63.7, 88.3)	85.3 (75.1, 91.8)	83.7 (73.7, 90.4)	1.4 (0.6, 3.2)
	OR§		1.2 (0.7, 2.1)		0.8 (0.4, 1.5)	
With ash	Prevalence‡	4.0 (0.7, 19.9)	3.4 (0.9, 12.5)	2.7 (0.4, 16.6)	3.3 (1.4, 7.4)	0.7 (0.1, 5.6)
	OR§		1.0 (0.2, 4.3)		1.5 (0.3, 7.8)	
With mud	Prevalence‡	8.0 (3.3, 18.1)	0 (0.0)	8.7 (6.0, 12.4)	0 (0.0)	n/a
	OR§		n/a		n/a	
Use of latrines at school	Prevalence‡	91.5 (83.4, 95.8)	99.4 (96.7, 99.9)	69.0 (29.5, 92.2)	72.8 (25.9, 95.3)	14.9 (1.4, 153.9)*¶
	OR§		18.4 (2.0, 169.8)*		1.2 (0.5, 3.1)	
Open defecation	Prevalence‡	6.3 (3.0, 12.6)	0 (0.0)	22.3 (5.5, 58.6)	20.1 (2.5, 71.0)	n/a
	OR§		n/a		1.2 (0.5, 2.8)	

CI = confidence interval; Hb = hemoglobin; n/a = not applicable; OR = odds ratio; SES = socioeconomic status.

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

† Knowledge, attitudes and practices

‡ Data are % (95% confidence interval, CI). The CIs are adjusted for clustering within schools by using robust standard errors.

§ Odds ratios (ORs) refer to the period effects. Mixed logistic regression models were adjusted for the two categorical SES variables and children's age.

¶ ORs refer to the intervention effect defined as ratio between the period effects in intervention and in control schools, with adjustment for SES variables and children's age.

|| This result is principally due to one school in the control group, where latrine use was denied by 93.5% of the children at end, line, while the prevalence of reported latrine use was otherwise 85.8% across all intervention and control schools at end-line.

7.5 Discussion

There is a lack of evidence on the potential benefits of combined school garden, nutrition, and WASH interventions on school children's intestinal parasitic infections and nutritional status.^{16, 22-}

²⁶ Results presented here from the parasitologic assessments among school children in two regions of Burkina Faso suggest that VgtS project-related interventions reduced intestinal protozoa, but only marginally improved helminth infections. No measurable improvements in nutritional indices among school children were observed. Environmental assessments showed no improvements in water quality parameters.

There are two main categories of interventions to address undernutrition in children: nutrition-specific interventions and nutrition-sensitive interventions.^{20, 23} While nutrition-specific interventions aim to address the immediate causes of undernutrition (inadequate dietary intake and disease), the objective of nutrition-sensitive interventions is to target the underlying determinants of undernutrition. The current evidence-base for interventions to improve children's nutritional status is primarily part of nutrition-specific interventions; showing beneficial effects on children's anthropometric indices.²⁰ For example, in a study conducted in India with 7- to 9-year-old children receiving fortified foods rich in seven micronutrients, a beneficial effect on linear growth at 12 months follow-up was found.⁴³ Another study conducted with children aged 6-11 years in Tanzania who received a fortified beverage with 10 micronutrients found that children's weight and height significantly improved in the intervention group at a 6-month follow-up.⁴⁴ Even though nutrition-specific interventions in school children have shown to be effective in reversing or improving negative health consequences,⁴³⁻⁴⁵ there is little evidence of multi-sectoral and nutrition-sensitive approaches (e.g., improving access to safe and hygienic environments and to diverse diets),²⁶ such as the VgtS project.²⁷ More recently, Prentice and others (2013)⁴⁶ argued that adolescence represents an additional window of opportunity during which growth-promoting interventions might have beneficial life course and intergenerational effects. However, these arguments have been opposed by Leroy et al. (2013)⁴⁷, for inadequately using changes in z-scores over time to define catch-up growth, highlighting that current evidence is still controversial on whether interventions in older children can induce catch-up growth.^{48, 49}

The significant decrease in total intestinal parasitic infections, particularly total intestinal protozoa infections, in both intervention and control schools is partially explained by anti-parasitic drugs provided to infected children after the baseline survey. However, the stronger decrease in the intervention schools related to the control schools may be indicative for the positive effects of the implemented WASH interventions.^{13, 50} Our study thus confirms the effectiveness of school-based programs to reduce intestinal parasitic infections among school

children. Other school-based health programs, for example the “Fit for School” approach implemented since 2008 in the Philippines, showed similar beneficial effects in terms of reducing the prevalence of intestinal parasitic infections.²¹ This school-based program included a package of several health interventions (e.g., handwashing with soap, improving water supplies and sanitary services, and bi-annual deworming), which has shown lasting effects on soil-transmitted helminth infections among school children.²¹

Schools are considered a convenient platform for concerted multi-sectoral public health action.^{15, 21} Combined school garden, nutrition, and WASH programs, facilitated through the education sector and supported by the health, sanitation, and agriculture sectors, have potential benefits across and beyond these sectors.^{51, 52} However, the overall modest effects found on school children’s intestinal parasite and nutritional status in our study requires a reconsideration of the program design. First, since school-going children spend time not only at school and their home, but also in potentially risky environments (e.g., rivers, lakes, and contaminated fields for open defecation), multiple interventions at various entry points are needed.^{26, 53-55} While children can be effective promoters of health messages received at school to their family members,^{53, 56} our findings are in line with previous studies showing that uptake and translation of health messages to effective behavior changes at their homes may be difficult to achieve (as changing practices takes time; for example to safely store and treat drinking water, but also due to key constraints such as water scarcity).⁵³⁻⁵⁵ A closer involvement of communities and households in school-based programs with a stronger household and community component might be necessary to achieve sustained and meaningful long-term effects for children’s health and well-being.^{7, 13}

Additionally, more comprehensive nutritional and agricultural interventions may be needed given the high rates of undernutrition found in our study regions. As the school feeding program (it is a governmental social protection program providing primarily staple foods to schools)⁵⁷ was not operational during the VgtS program implementation phase in our study sites, harvested vegetables were rarely prepared for consumption at schools. Hence, by widening the intervention approach from schools to the larger community and linking the school garden to home- and community-gardens,²⁶ vegetable production could be increased and used for consumption at children’s homes. This approach was pursued in a 2-year integrated agriculture and nutrition program in Burkina Faso (2010-2012).^{26, 58} The program design included homestead food production (micronutrient rich fruits and vegetables), coupled with a behavior change communication component. The key results from the program evaluation (2016) showed a significant reduction of underweight in mothers and wasting in children aged 3-12 months.^{26, 58} Hence, multi-sectoral nutrition-sensitive interventions offer a unique opportunity; however, more sustained programs linking school-, home-, and community-based interventions tailored to the social-ecological contexts in Burkina Faso are needed to improve school children’s health status

on a long-term basis.²³ Taken together, the baseline and end-line data collected provided a benchmark for assessing changes in school children's health status over a 1-year period. By conducting repeated cross-sectional surveys in a cohort of children, this study has provided setting-specific data on school children's intestinal parasite infections and nutritional status, and calls for longer-term studies addressing school children's health through multi-sectoral and multi-stakeholder school- and community-based programs. The described study methodology presents a suitable approach for evaluating school-based health programs in settings where there is a paucity of health data among school-aged children.²⁷ The present study is among a few evaluations in sub-Saharan Africa that provides new evidence that school-based interventions can improve children's health.^{59, 60}

There are several limitations to our study. First, considering the positive short-term impacts on children's parasitic infection status and the potential for longer-term benefits for children's nutritional outcomes, integrated agriculture, nutrition, and WASH programs should be implemented over longer periods. The 5-6 months allocated here (due to delayed project implementation and end of the project phase in 2016) limit to unveil a potentially larger benefit in improving children's health.^{26, 61} Second, hygiene and sanitary practices of children were self-reported and behavior change was not directly observed. Children may have over- or under-reported proper hygiene practices at baseline or end-line.⁶² Third, the power calculation of this study was conducted to address the initial cross-sectional hypothesis with the aim of comparing the prevalence of intestinal parasitic infection between children considered at high or at low risk of infection. The study therefore had limited power to test effects of the subsequent interventions, which is also reflected in the relatively wide confidence intervals of our results. Fourth, we did not collect data on malaria, which might have provided a deeper understanding for the results pertaining to anemia. Fifth, the diagnosis of helminths using the Kato-Katz technique with only one thick smear per specimen at baseline had a lower sensitivity than the duplicate thick smears employed at end-line survey one year later. The reported values at baseline might therefore be biased downward.⁶³ Finally, the findings may be specific for the selected schools with similar characteristics and may not be representative for a wider area and other regions in Burkina Faso.

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Disclosures

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8 Discussion

The overarching goal of this PhD thesis was to investigate undernutrition and intestinal parasitic infections among schoolchildren in two regions of Burkina Faso and to evaluate the effects of complementary school garden, nutrition and WASH interventions on mitigating ill-health and improving children's nutritional status. The work for this PhD thesis was embedded in the operational research project, "Vegetables go to School: Improving Nutrition through Agricultural Diversification" (VgtS). The thesis research pursued interdisciplinary approaches, applied descriptive and analytical epidemiology and linked field and laboratory work. At the core of this research was a cluster-randomised controlled trial (RCT), including a baseline and an end-line survey (Chapter 4). This PhD research entailed three specific objectives. The first objective was to investigate schoolchildren's nutritional status and associated risk factors (Chapter 5). The second objective was to determine the prevalence of intestinal parasitic infections among schoolchildren and its association with household- and school-level WASH conditions (Chapter 6). The third objective was to generate evidence on the effects of complementary school garden, nutrition and WASH interventions on selected key indicators for schoolchildren's health and nutritional status one year after a baseline cross-sectional survey (Chapter 7).

The following chapter addresses the three PhD study objectives, highlighting the key findings and lessons learned. In the first section (8.1), the findings from the baseline survey are discussed in the broader context of undernutrition, intestinal parasitic infections and WASH in Burkina Faso. The second section (8.2) highlights the results from the cluster-RCT and its contribution to the current scientific discussions on the benefits of linking agriculture, nutrition and WASH interventions to improve children's nutritional and health outcomes. The third section (8.3) describes methodological limitations of our study. The fourth section (8.4) considers the implications for public health by placing our findings in the context of school-based health programmes and the global development and health agenda, showing the opportunities that a focus on schoolchildren in public health strategies can provide. The fifth section (8.5) explains how this PhD work contributes to the three main pillars of Swiss TPH in the field of public health, namely innovation, validation and application (Table 8.1). Finally, the last two sections provide a set of conclusions (8.6), research needs and recommendations (8.7) for further improving schoolchildren's nutrition and health in Burkina Faso and elsewhere.

8.1 Epidemiology of undernutrition and intestinal parasitic infections among schoolchildren in two regions of Burkina Faso

Our findings from the anthropometric survey showed that undernutrition was very common at baseline among the 385 schoolchildren surveyed in the Plateau Central and Centre-Ouest regions. Indeed, more than 35% of children were classified as undernourished (stunted, thin and/or underweight); whereby children aged 12-14 years were at significantly higher odds of undernutrition compared to their younger peers (8-11 years). Comparison to previous studies is limited, as schoolchildren are not included in Demographic and Health Surveys (DHS) and national nutrition surveillance systems in Burkina Faso [1-3]. There is only one recent study, conducted between 2008 and 2009 among schoolchildren in urban and peri-urban schools of Burkina Faso as part of the WHO's "Nutrition Friendly School Initiative" (NFSI) [4]. This NFSI study found a 8.8% prevalence of stunting, which is much lower compared to our findings (29.4% at baseline) [4]. The lower prevalence of stunting found in the NFSI study might be partially explained by the different study settings. Our study was located in rural areas of the Centre-Ouest and Plateau-Central region, whilst the NFSI study was conducted in urban and peri-urban Ouagadougou. In rural areas, inadequate access to food and health services is often a result of poverty and broader social determinants of health that differ from those in urban peri-urban settings [5-7]. Thus, it is not surprising to find a higher percentage of undernutrition in children in rural areas, compared to urban and peri-urban settings [5, 8].

Moreover, we found that a small number of children were overweight (2% at baseline). While in high-income countries (HICs) [9, 10], high rates of overweight and obesity among school-aged children are well documented, it is a recent but rapidly increasing phenomenon in sub-Saharan Africa (SSA), particularly in urban settings where changes in living standards, urbanisation, lifestyles and diets are most remarkable [11-15]. Even though our study was conducted in a rural setting, our findings suggest that overweight is an important parameter to consider and to be monitored in future studies on schoolchildren's health as the transition between under-to-overweight can occur within the same individual and generation [16-18].

Our results stemming from parasitological examinations showed that helminths and particularly intestinal protozoa are still widespread among schoolchildren in Burkina Faso [19]. However, the prevalence of soil-transmitted helminth (STH) and schistosomiasis infections found in our study were lower than those found in retrospective reviews and in studies conducted over the past decade [20-23], but in line with findings from recent cross-sectional surveys (between 1.3% and 1.7%) [24, 25]. In fact, the only STH species found in our study was hookworm (0.8%), while 4.2% of the children were infected with *Schistosoma* spp. (primarily *S. haematobium*), all of which were of low intensity [26]. We found a higher prevalence of children infected with *H. nana* (6.5%) compared to results from previous surveys conducted in Burkina Faso (from

0.3% to 4.0%) [22, 24]. Yet, those studies included individuals of all ages, though schoolchildren are often at highest risk of acquiring STH and other intestinal helminth infections [19, 27, 28].

The relatively low prevalence and intensity of STH and *Schistosoma* infections found among schoolchildren in our study region might be attributed to intensified control measures implemented in the early 2000s [25, 29, 30]. Of note, even though hymenolepiasis is absent from the WHO guidelines for helminth control programmes [31], it is also being addressed by praziquantel as part of the mass drug administration (MDA), or with niclosamide [32]. Sanitary, ecological or seasonal parameters (baseline collection took place during the dry season) may also have had a positive effect on STH infection prevalence and intensity [33, 34]. Of the households participating in our study, 55.3% reported practicing open defaecation and only 23.1% had access to improved sanitary facilities [26]. This finding indicates that sanitary conditions among our study population were considerably inadequate. However, data from the World Health Organization (WHO)/United Nations Children's Fund (UNICEF) 'Joint Monitoring Programme for Water Supply and Sanitation' (JMP) assessment (2015) shows that 75% of the rural population in Burkina Faso practised open defaecation [35], which may suggest recent improvements in our study regions. In any event, the findings from our study and recent assessments by the national disease control programme showing that helminth infection prevalence and intensity are at low levels indicate progress towards achieving elimination of STH transmission [25, 36].

However, intestinal protozoa infections were endemic throughout the study area: indeed, more than three-quarters of the schoolchildren harboured either *Entamoeba histolytica*/*E. dispar* or *G. intestinalis* infection (mean prevalence of 75.3%; 66.5% for *E. histolytica*/*E. dispar* and 28.1% for *G. intestinalis*). Our data also showed higher prevalences compared to results from other studies in Burkina Faso conducted between 1990 and 2012, which reported infection rates between 23% and 39% for *E. histolytica*/*E. dispar* and between 5% and 46% for *G. intestinalis* [21, 22, 24]. These studies were hospital-based and investigated intestinal parasites among individuals of all age groups with gastrointestinal problems [21, 22, 24]. While evidence suggests that *E. histolytica* and *G. intestinalis* are pathogenic protozoa causing diarrhoeal diseases in patients, infections with these intestinal protozoa are frequently asymptomatic [37-39], which may explain the higher prevalences found in our study.

Furthermore, we revealed important sociodemographic factors that contributed to children's intestinal parasitic infection status. *G. intestinalis* in children was associated with "freely roaming" domestic animals, particularly dogs [26], which indicates a potential zoonotic transmission of *Giardia* [40]. While there is considerable genetic diversity within *G. intestinalis*, the major genotypes that are infective to humans are assemblages A and B. Both assemblages have been isolated from animal hosts [41], particularly assemblage A, while B is predominately associated with human isolates [42]. It is important to note that the morbidity associated with

G. intestinalis seems to vary according to the genotypes found [43-45]. For example, a study in Bangladesh (2009) found that *Giardia* infection was associated with protection against diarrhoea, while only assemblage A was associated with acute diarrhoea [44, 46]. Other studies, however, found that assemblage B infection was significantly associated with clinical symptoms of giardiasis [47, 48]. These different findings indicate the inconclusive association between *G. intestinalis* assemblages and clinical symptoms, which might be associated with other factors, such as age, nutritional and immunological status of the host [49]. Future research on the species-specific significance for children's morbidity patterns is needed for a deeper understanding on intestinal protozoa infections and its implications for treatment and prevention strategies [50].

Our results also confirm that children infected with intestinal parasites are at higher odds of being undernourished (Chapter 5) and highlight the frequent coexistence of these two conditions in children [19, 51]. Previous studies have shown that helminth and intestinal protozoa infections can lead to undernutrition in children by causing loss of appetite and nutrients and decreasing nutrient absorption due to, for example, mucosal damage (see Chapter 2.4.3) [19, 52, 53]. In turn, this can result in inadequate dietary intake and further deficiencies of essential nutrients, such as protein, iron, iodine, folate, zinc and vitamin A – all of which are essential for growth and maintaining immune functions (see Chapter 2.3.1) [19, 54, 55]. Nevertheless, there are still many unknowns regarding the pathologic pathways by which intestinal parasitic infections may lead to further health consequences (e.g. changes in gut microbiota and environmental enteric dysfunction, EED) and associated morbidity. Hence, to adequately and efficiently address ill-health among schoolchildren, further investigation on the implications of undernutrition and intestinal parasitic infections for children's health is warranted [56].

In an attempt to answer the question of the burden of undernutrition and intestinal parasitic infections among schoolchildren in our study regions, we present the following issues for consideration, bearing in mind that: children primarily suffered from chronic undernutrition (29.4%); and fewer children were infected with faecal-oral transmitted helminths (7.0%) and *Schistosoma* spp. (4.2 %) than with intestinal protozoa (84.7%). Whilst intestinal protozoa infections need individual treatments [57], current control efforts for STH in Burkina Faso focus on drug interventions. Yet, since 2012, the largest deworming programme has gradually stopped [25, 58]. Our findings call for urgent actions to implement an integrated control approach to address undernutrition and intestinal parasitic infections among schoolchildren and to sustain the gains from previous large-scale disease control programmes. Such actions should make use of multidisciplinary strategies and programmes to address the multitude of proximal and more distal determinants influencing schoolchildren's nutritional and health status. The nutritional, parasitological and questionnaire data reported here served as a benchmark from

which to assess the effects of the complementary school garden, nutrition and WASH interventions with the goal to improve schoolchildren's nutritional and health status.

8.2 Effects of complementary school garden, nutrition and WASH interventions on schoolchildren's health and nutritional status

The rationale for assessing the effects of complementary school garden, nutrition and WASH interventions is based on the assumption that these interventions address proximal and underlying factors of children's nutritional status by (i) improving dietary intake; (ii) reducing and preventing new intestinal parasitic infections (proximal factors); (iii) improving children's health practices; and (iv) improving WASH conditions at schools (underlying factors). As a result of these benefits, children would likely face fewer infections with intestinal parasites and have a better nutritional status. This is expressed in our conceptual framework, which provides the basis for discussion of our findings in this section.

Results from our longitudinal analysis showed that children from the intervention group did not gain more height or weight compared to children from the control group. On the contrary, z-scores for stunting and thinness further decreased in both intervention and control groups (Chapter 7). The lack of improvement of children's nutritional status might be explained by the limited possibilities for addressing proximal factors of undernutrition, particularly inadequate dietary intake, since the national school feeding programme was not operational in our study sites during the VgtS project period. Therefore, a significant increase in children's weight or height gain was not expected.

Nevertheless, we found a significant reduction of multiple intestinal parasitic infections among schoolchildren. Unexpected were, however, the lack of interaction found between the reduction of intestinal parasitic infections and nutritional improvements in children in our study (Chapter 7). There are several possible explanations for this finding. First, children found infected with intestinal parasites from the intervention and the control group received treatments after the baseline survey in March 2015. The effects of treatment on children's nutritional outcomes are therefore difficult to assess, as having an untreated control group would be unethical [59]. Second, the potential benefit of anthelmintic treatments on children's nutritional outcomes has recently been the subject of considerable controversy, particularly the nutritional benefits of single and multiple doses of anthelmintic treatments provided to children in endemic areas [60-62]. However, conclusions across the three reviews were largely consistent with regards to the nutritional benefits of treating children infected with STH (detected by screening), noting that children have a larger average weight gain after anthelmintic treatments. Still, the estimated effect reported in these reviews is fairly modest (between 0.3 kg and 0.8 kg) [61, 62], which might further explain the lack of effect of treatment on average weight gain found.

Chronically undernourished children require sufficient time and specific improvements to their diets (e.g. additional energy and nutrients) in order to restore their health [59, 63, 64] (Chapter 7). It follows that solely reducing the burden of intestinal helminth and protozoa infections by providing anti-parasitic drugs to infected children is unlikely to remedy any underlying nutritional deficits that were caused by these infections [60]. Considering the role of more distal factors, the VgtS project implemented many promising complementary approaches targeting improved sanitation and handwashing with soap, access to clean drinking water at schools and awareness raising (school- and community-based health education). These interventions have proven to contribute to declines in intestinal parasitic infections among schoolchildren of the intervention group (Chapter 7), which is in line with findings from previous studies investigating WASH and intestinal parasitic infections [33, 65, 66], but were unlikely to show their potential for improving children's nutritional status after our five-month intervention period. Furthermore, as intestinal parasites can cause symptomatic infections associated with diarrhoea, as well as asymptomatic infection, such as EED (Chapter 2.5.3), future studies should investigate other pathways and underlying biological mechanisms to enhance understanding of the linkages between WASH and undernutrition.

Findings from previous studies on school feeding programmes, agricultural and WASH interventions among children in low- and middle-income countries (LMICs) showed little effect on nutritional status. Our results show that VgtS project-related interventions hold promise for improving schoolchildren's health and potentially their nutritional status. In order to achieve a more substantial change, several factors must be carefully considered for designing future interventions and for choosing methods to assess the effects of such interventions; these will be highlighted in the next section.

8.3 Methodological limitations and prospects for future studies

Estimating the precise prevalence of stunting, underweight and thinness through anthropometry turned out to be a challenging task. We found several methodological limitations in applying it to our study. First, the use of anthropometric indicators for schoolchildren, including body mass index (BMI), height-for-age (HAZ) and weight-for-age (WAZ) z-scores, are age dependant [67]. At the baseline survey, we noted that many children had their birthdays on 31 December or on 1 January of the indicated year (see Chapter 5). To overcome this limitation, we took a mid-year point as the date of birth. Doing so may have introduced a random bias, resulting in lower or higher anthropometric prevalence estimates [68]. For future research, it may be necessary to construct a local calendar to determine exact ages (day and month) [69], as children in rural parts of Africa are much less likely to be registered in civil registration systems; a fact that holds true for Burkina Faso and our study area, in particular [70, 71].

Second, the classification and the application of different cut-offs may have had an effect on our reported rates of undernutrition [72, 73]. For example, not all schoolchildren who fall below a certain cut-off point are at risk of a nutrition-related morbidity [72, 74]. The cleaning criteria used before data analysis may also have different effects on prevalence estimates [73]. In our study, we used the new cut-offs as recommended by the WHO for data exclusion. Thus, data were excluded if a child's HAZ was below -6 or above +6, if WAZ was below -6 and +5, or if BMIZ was below -5 or above +5 standard deviation (SD) [75]. We included plausible values of nutritional status (i.e. z-score below -3) to indicate severe forms of undernutrition, but which may, in few cases, have yielded random measurement errors [68, 75]. Hence, cut-offs serve as a screening device to identify children, who are more likely to be undernourished [72]. In future studies, it may be useful to combine anthropometry with other indicators indicating nutritional deficiencies, such as muscle strength (e.g. grip strength) or biochemical markers (e.g. for assessing micronutrient status) to investigate morbidity related to undernutrition in school-aged children [74, 76].

In our study, we were particularly interested in assessing how intestinal parasitic infections and inadequate WASH conditions are associated with children's nutritional status. Even though data collected in this study provide an important evidence base and allowed us to describe the extent and implications of intestinal parasitic infections among schoolchildren, the parasitological survey suffers from several diagnostic limitations. Morbidity related to intestinal parasitic infections is likely to be more pronounced (and measurable) in high-intensity helminth infections and depends on the pathogenicity of the intestinal protozoa species found [77-80]. Whilst the Kato-Katz technique is recommended for quantitative diagnosis in epidemiological surveys [81], it lacks sensitivity for diagnosing low-intensity infections [82, 83]. Analysing multiple stool samples from several consecutive days or preparing multiple thick smears from a single sample is known to increase sensitivity for helminth detection [79, 84, 85]. For light-intensity infections, alternative and more sensitive diagnostic methods have been developed, such as the FLOTAC technique (a flotation technique for faecal egg count) [86-89]. Interestingly, the FLOTAC technique is also suitable for diagnosing intestinal protozoa (sensitivity depends on the species investigated) and complements the formalin-ether concentration (FEC) technique [90], which has shown some shortcomings in accurately diagnosing intestinal protozoa [91]. However, the FLOTAC technique requires better equipped laboratories and incurs higher costs [83]. The Mini-FLOTAC – a simplified adaptation of the original FLOTAC technique – may represent a suitable alternative to overcome some of these weaknesses; still, in low-intensity settings, it was shown to have similar or lower sensitivities than the Kato-Katz technique [89, 92]. Future studies could benefit from further investigating the use of the (Mini-)FLOTAC techniques, as helminths and intestinal protozoa species can be detected concurrently. As helminths and intestinal protozoa,

in particular, are common in Burkina Faso, this would present a major step forward in the epidemiological surveillance of polyparasitism [93, 94].

Moreover, considering the differential diagnosis of intestinal protozoa (e.g. pathogenic or non-pathogenic *Entamoeba* species), more sensitive techniques are based on antigen detection or polymerase chain reaction (PCR) assays [38]. Current opinion suggests that molecular techniques are the most promising methods for detecting multiple protozoa species with high sensitivity and accuracy, compared to microscopy, antigen detection or staining methods [38, 80, 95]. Since molecular methods were not applied in our study to differentiate between the pathogenic (*E. histolytica*) and non-pathogenic (*E. dispar*) species or to identify *G. intestinalis* genotypes among infected schoolchildren, nor in the previously cited studies investigating intestinal protozoa infections in Burkina Faso [21, 22, 24], the feasibility of employing these methods routinely in low-resource countries (requiring specified equipment) may need further consideration [96, 97]. Nevertheless, future research including species-specific differentiation using PCR or other more sensitive diagnostic methods is crucial to advance understanding of morbidity related to different *Entamoeba* spp. and *Giardia* genotypes and for reducing transmission in high-endemicity settings, such as Burkina Faso.

Finally, in view of the multifactorial nature of undernutrition, the primary limitations of our longitudinal analysis relate to measuring appropriate outcomes within the interlinked and lengthy impact pathways and a short intervention period. Well-designed RCTs are traditionally considered to be the 'gold' standard in health research [98-101]. Yet, there are some challenges inherent to using RCTs in nutrition-sensitive projects. The impact-pathways are often very long and interlinked and thus nutritional indicators, particularly height- compared to weight-related indices, may not be sensitive enough for detecting the effects of changes in disease or diet in the short-term [68, 100]. Even though RCTs provide the strongest evidence of a causal relationship between intervention and outcome, the combination of RCTs with observational approaches (e.g. structured observation) might be promising for future field studies to provide additional insights into and evidence of certain outcomes of interest, such as dietary or hygiene behaviours, although they are often subject to confounding [101, 102]. Furthermore, a modified cluster-RCT study design with two or more follow-up studies could be employed in future studies over a time period of several years instead of several months, which was often cited as a limiting factor in previous intervention studies [103, 104]. This would grant more time for the implementation and uptake of different project components and would allow researchers to assess longer-term changes in nutritional indices and parasitic reinfection rates.

8.4 Public health implications

The findings from our study showed that intestinal parasites, particularly infections with intestinal protozoa and chronic undernutrition, were common among schoolchildren in two regions of Burkina Faso. A large-scale disease control and a national school feeding programme targeting school-aged children's health in Burkina Faso are underway. The school feeding programme is a promising entry point for nutritional improvement [105, 106], while school-based deworming strategies have shown to be an effective means of reducing STH and *Schistosoma* infections in the country [25]. However, there is a lack of large-scale multi-sectoral nutrition-sensitive programmes addressing underlying determinants of undernutrition and ill-health among schoolchildren. VgtS project-related interventions (i.e. school-based vegetable garden, nutrition and WASH interventions) hold promise for improving schoolchildren's health. However, considering that nutritional indicators at end-line were lower than at baseline and taking into account the high rates of drinking water contamination found, these findings underscore how vulnerable the area is to food insecurity and to challenging socio-ecological and poverty-related conditions [107]. To substantially improve proximal (i.e. dietary intake and intestinal parasitic infections) and more distal factors (e.g. inadequate WASH and food insecurity) of undernutrition in children, there is a need for interventions at both school- and household-level with a strong community involvement. Furthermore, to implement at regional and national scale, long-term collaboration between Ministries of "National Education and Literacy", "Health", "Agriculture, Water, Sanitation, and Food Security", disease control programmes, and other stakeholders are essential (Chapter 7).

As many of the health problems present in school-aged children and adolescents develop during early infancy [108, 109], broadening the current intervention approach to include school-aged children in a maternal and child health life-course perspective could provide a unique opportunity to increase attention to school-aged children's health. It posits that health is indivisible throughout an individual's life and that particularly pregnancy and the early stages of childhood development have an important impact on child and adolescent health later in the cycle [110]. The life-course approach to maternal and child health has recently entered into science and onto the global development agenda. For example, a recent *Lancet* series on "Advancing early childhood development: From science to scale" (2016), focuses on the intergenerational approach of maternal and child health and highlights the importance of growth and development during infancy for the acquisition of skills and learning in middle childhood, throughout adolescence, and into adulthood [109, 111, 112]. Therefore, interventions and strategies promoted as part of the life-course approach to health mainly focus on children under the age of 5 years, women, and adolescence in a reproductive health perspective [109, 111, 112]. This new impetus is also reflected in the launch of the WHO led UN "Global strategy for women's, children's and adolescents' health (2016–2030) [113], which is aligned with several

targets of the Sustainable Development Goals (SDGs)², particularly with SDG 3 on health and SDG 2 on hunger and food security [114, 115].

Even though aspects of school-aged children's and adolescent's health are known to be associated with earlier determinants [6, 116], more emphasis needs to be placed on the transitional period between childhood and adulthood, which is marked by increased dietary requirements, particularly during the adolescence growth spurt [117-119]. From a global health perspective, a truly multi-sectoral approach to school-aged children's health in a life-course approach could be achieved by integrating schoolchildren into the nutrition-related targets (reducing stunting and wasting and preventing obesity) of SDG 2. These nutrition-related targets could be closely linked to the prevention of neglected tropical diseases (NTDs) of SDG 3, where school-aged children are a target group for preventive chemotherapy, often provided through the education sector. Furthermore, as adequate water and sanitation are essential to reduce transmission of intestinal parasites, SDG 3 needs to be implemented in conjunction with SDG 6, where population- and school-based improvements of drinking water and sanitation are essential targets. Finally, primary and secondary schools are a convenient setting in which to promote healthy eating and hygiene practices, thereby complementing the educational objectives of SDG 4 (Figure 8.1) [114]. The SDGs provide a timely opportunity to increase attention to schoolchildren's health through multi-sectoral approaches, concurrently addressing nutrition, disease prevention (clean water, adequate sanitation and hygiene), and educational needs while building a strong foundation for future health and well-being [114, 118].

² SDG 2: "End hunger, achieve food security and improved nutrition and promote sustainable agriculture"
SDG 3: "Good health and well-being";
SDG 4: "Ensure inclusive and equitable quality education and promote lifelong learning opportunities for all"
SDG 6: "Clean water and sanitation"

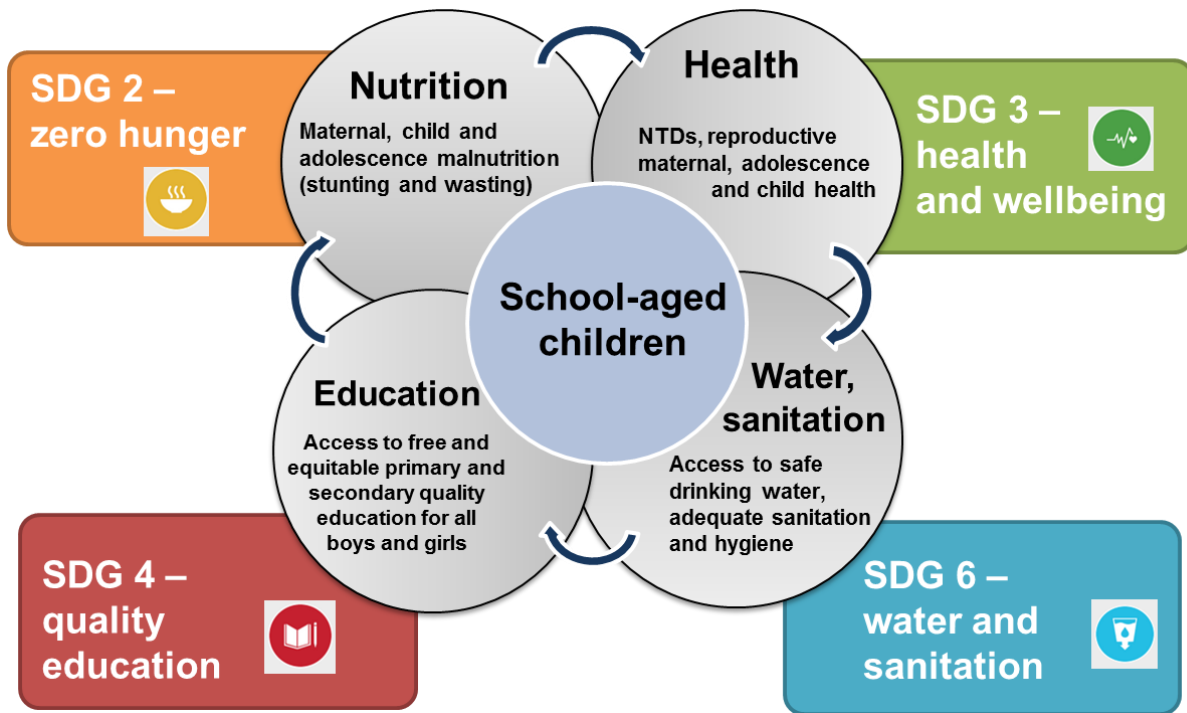


Figure 8.1: Integration of school-aged children in the global development and health agenda as part of the Sustainable Development Goals (SDGs).

8.5 Thesis contribution to innovation, validation and application

The work presented in this PhD thesis was pursued at Swiss TPH and contributes to the institute's three main pillars of activity along the entire value chain from innovation to application. In brief, it provides new evidence and insights that are of significance from a public health point of view for Burkina Faso and elsewhere. Table 8.1 summarises the main contribution of the present PhD thesis research.

Table 8.1: Summary of manuscripts and their contributions to the Swiss TPH nexus of innovation, validation and application

Chapter	Innovation	Validation	Application
4	The study design and evaluation frameworks for the VgtS cluster-RCTs were developed by Erismann and Shrestha et al. (2014) for multi-sectoral school-based projects in Burkina Faso and Nepal.		
5	High prevalence of undernutrition among schoolchildren, particularly among the older age group (12-14 years), and significant associations between undernutrition, intestinal parasitic infections and anaemia.	The baseline and end-line cross-sectional surveys conducted in the two VgtS project regions in Burkina Faso were a validation of the study protocol and the proposed methodology with the following objectives: (i) to estimate the prevalence of undernutrition;	The findings from the baseline survey of the VgtS project helped to design and implement complementary interventions.
6	Particularly high prevalence of intestinal protozoa infections, and role of household environments, particularly domestic animals, as important risk factors.	(ii) to investigate intestinal parasitic infections among schoolchildren; and (ii) to evaluate the effects of the complementary intervention package on schoolchildren's nutritional and health status.	
7	Significant reduction in intestinal parasitic infections among schoolchildren, while indices of undernutrition, anaemia and water quality remained unchanged after the implementation of complementary school garden, nutrition and WASH interventions.		Promising new multi-sectoral approaches to address proximal and underlying determinants of undernutrition in children, i.e. intestinal parasitic infections and inadequate WASH conditions.
8	Nutrition-sensitive programmes in LMICs were rarely assessed for mitigating ill-health and improving nutrition in school-aged children.	The present study is one of just a few operational research studies assessing the effects of complementary school garden, nutrition and WASH interventions on schoolchildren's nutritional and health status in West Africa.	VgtS project-related interventions were promising in improving schoolchildren's health. Future multi-sectoral school-, household-, and community-based programmes are required to further improve nutrition and health in schoolchildren.

8.6 Conclusions

The overarching goal of this PhD thesis was to deepen the understanding of schoolchildren's nutritional and health status in rural schools of the Plateau Central and Centre-Ouest regions of Burkina Faso. Furthermore, this work aimed to provide evidence of the effects of a multi-sectoral programme linking school gardens, nutrition and WASH interventions. The present study is one of just a few operational research studies assessing these complementary interventions on schoolchildren's nutritional and health status in a rural setting of West Africa.

The VgtS project pursued an innovative approach towards linking agriculture, nutrition and WASH interventions with the goal of improving schoolchildren's nutritional and health status. We assumed that an integrated intervention model had the potential to generate powerful synergies in addressing proximal and underlying determinants of undernutrition in schoolchildren, including intestinal parasitic infections and WASH conditions. This PhD research provided detailed characterisation of schoolchildren's nutritional and health status. Specifically, this work showed that chronic undernutrition and intestinal parasitic infections, in particular intestinal protozoa infections, were common among schoolchildren. While intestinal parasitic infections were significantly lower after the implementation of the complementary intervention package, the prevalence of stunting, thinness and overweight slightly increased among schoolchildren over the one-year period. Even though it may be difficult to improve children's nutritional status at school-age originating from earlier in life, this age group nonetheless presents an invaluable opportunity to shape and consolidate safe hygiene and healthy eating practices.

The baseline data collected as part of the study served to inform the design of the school-based interventions. Broad stakeholder engagement was considered essential for the development of the multi-sectoral intervention programme in the frame of the VgtS project. The design and planning process of the intervention activities and the implementation presented unique challenges. For example, different priorities between different sectoral disciplines (e.g. education, agriculture, nutrition and WASH) had to be accounted for, as well as circumstances where resources were limited. Nevertheless, our results showed that the multi-sectoral school-based intervention approach of the VgtS project holds promise for improving schoolchildren's health and potentially their nutritional status. Undernutrition is inseparable from agriculture, WASH, education and larger poverty reduction and economic growth measures. Therefore, long-term investments and strong multi-sectoral collaboration, including the close involvement of households and communities, will be essential in future nutrition-sensitive programmes aiming to improve nutrition and health in schoolchildren.

8.7 Research needs and recommendations

In view of the current evidence-base on nutrition-sensitive programmes, and considering the findings from the present PhD thesis, the following research needs and recommendations arise for future studies on schoolchildren's nutrition and health in LMICs.

- (i) Undernutrition among schoolchildren needs further scientific inquiry. There is a need to adequately distinguish children who have low anthropometry but are healthy from those who are undernourished due to a lack of proper nutrition or disease. Moreover, locally relevant indicators as proxy for schoolchildren's nutritional status and dietary intake should be identified in future studies; these could be used as a complement to anthropometric indicators in resource-constrained settings.
- (ii) A deeper understanding on intestinal parasite species-specific implications for children's morbidity and for treatment and prevention strategies is required, using more sensitive diagnostic methods. It would be particularly interesting to further investigate the role of domestic animals, especially dogs, in *G. intestinalis* transmission and the effects of reducing animal faecal contamination in domestic spaces for children's infection status.
- (iii) Other underlying biological and non-biological pathways leading to undernutrition, such as environmental enteropathy, or the role of household food expenditures (for water, foods) require further investigation. This will be essential to adequately address ill-health among schoolchildren. This will enhance our understanding on the complex interactions of undernutrition, intestinal parasites and WASH, as well as other possible pathways leading to undernutrition and ill-health among children.
- (iv) Furthermore, stronger evidence of the benefits of multi-sectoral programmes will be necessary, particularly for the purposes of advocacy and for the inclusion of schoolchildren as target group in health policies in Burkina Faso. Such multi-sectoral programmes could involve integrated school-, household- and community-based interventions tailored to the social-ecological contexts in Burkina Faso. Health education, information and communication (EIC), access to safe water and community-led total sanitation may be implemented, as well as linking school gardens to home, and community gardens.
- (v) Even though aspects of school-age and adolescent health are known to be associated with early-life exposures, more emphasis needs to be placed on schoolchildren's health. As a public health recommendation drawn from our study, schoolchildren should be included in data collection activities. Data on schoolchildren may be included in existing

sub-national or national (e.g. DHS) health monitoring systems; these monitoring systems have thus far have been measuring health outcomes for children aged under the age of 5 years. This will enhance awareness on schoolchildren's nutritional needs and provide guidance for public health strategies addressing undernutrition among schoolchildren in Burkina Faso. It would particularly also enhance awareness and timely actions to further prevent the nutritional transition, i.e. coexistence of undernutrition and overweight, in schoolchildren and adolescents in Burkina Faso.

- (vi) Finally, school-aged children should receive greater attention in global health and development strategies and policies, for example by integrating schoolchildren into the nutrition- and health-related targets of SDGs 2 and 3, and in respective international monitoring frameworks provided by the WHO, UNICEF and others.

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9 Appendices

9.1 Prevalence of intestinal parasitic infections and associated risk factors among schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso

9.1.1 Additional file 1. Table S1. Results from univariate and multivariate logistic regression analysis for *Giardia intestinalis* and *Entamoeba histolytica*/*E. dispar*

Risk factor	<i>Giardia intestinalis</i> (n = 108)							<i>Entamoeba histolytica</i> / <i>Entamoeba dispar</i> (n = 256)						
	Univariate log. regression*				Multivariate log. regression**			Univariate log. regression*				Multivariate log. regression**		
	N [†]	OR	95% CI	P	aOR	95% CI	P	N [†]	OR	95% CI	P	aOR	95% CI	P
Sex														
Male (197)	64	1.00						125	1.00					
Female (188)	44	0.63	0.40–1.00	0.05	0.63	0.40–1.00	0.05	131	1.38	0.88–2.16	0.16	1.37	0.87–2.15	0.17
Age group														
8–11 yrs (251)	69	1.00						163	1.00					
12–14 yrs (143)	39	1.08	0.68–1.72	0.74	*			93	1.12	0.68–1.85	0.67	*		
Region														
Centre-Ouest (187)	59	1.00						146	1.00					
Plateau Central (198)	49	0.71	0.46–1.11	0.14	0.68	0.42–1.11	0.12	110	0.35	0.18–0.69	0.001	0.35	0.18–0.66	0.001
Hygiene^a														
Middle third (2) (227)	62	1.00						33	1.00					
Lower third (1) (56)	14	0.89	0.73–2.03	0.73	*			154	0.63	0.32–1.22	0.17	0.63	0.32–1.22	0.17
Higher third (3) (102)	32	1.22	0.73–2.03	0.45	*			69	1.01	0.60–1.72	0.97	*		
Drinking water for consumption^b														
From home (239)	70	1.18	0.74–1.87	0.49	*			158	1.10	0.67–1.79	0.71	*		
From school (322)	88	0.81	0.45–1.45	0.48	*			217	1.01	0.49–2.11	0.97	*		
Water risk behaviours														
No water contact (93)	27	1.00						64	1.00					
Playing (5)	1	0.61	0.07–5.72	0.67	*			3	0.45	0.07–3.07	0.42	*		
Fishing (25)	8	1.15	0.44–2.98	0.77	*			16	0.76	0.29–2.03	0.59	*		
Making laundry (56)	9	0.47	0.20–1.09	0.08	1.00	0.88–1.15	0.95	37	0.83	0.39–1.77	0.63	*		
Domestic chores (206)	63	1.08	0.63–1.84	0.79	*			136	0.78	0.44–1.38	0.39	*		
Any water contact ^b (292)	81	0.94	0.56–1.57	0.81	*			192	0.78	0.45–1.34	0.37	*		
Sanitary practices children														
Using latrines at school (307)	86	1.00						204	1.00					
Using latrines at home/teacher's (7)	2	1.03	0.20–5.40	0.97	*			4	1.16	0.22–6.09	0.86	*		
Open defaecation at school ^c (71)	20	1.00	0.57–1.79	0.98	*			48	0.74	0.36–1.53	0.42	*		
Caregiver's education														
Never went to school (288)	83	1.00						189	1.00					
Primary education (59)	16	0.92	0.49–1.72	0.79	*			44	1.61	0.82–3.14	0.17	1.43	0.97–2.10	0.07
Secondary education (38)	9	0.77	0.35–1.69	0.51	*			23	1.17	0.55–2.52	0.68	*		
Caregiver's occupation														
Agriculture (344)	97	1.00						232	1.00					

Civil service (8)	1	0.36	0.04–3.00	0.35	*			4	0.79	0.18–3.59	0.76	*		
Merchant (9)	3	1.27	0.31–5.19	0.74	*			6	0.94	0.21–4.28	0.94	*		
Others ^d (24)	7	1.05	0.42–2.61	0.92	*			14	0.69	0.28–1.75	0.44	*		
Animals^b														
Possession of domestic animals ^g (371)	106	2.4	0.53–10.91	0.26	*			248	1.06	0.33–3.37	0.93	*		
Animals held in the house (246)	79	1.79	1.10–2.93	0.02	2.00	1.20–3.34	0.01	165	1.02	0.64–1.62	0.94	*		
Household sanitary conditions														
Traditional latrine (213)	25	1.00						52	1.00					
No latrines/ open defaecation (83)	64	1.58	0.88–2.84	0.12	1.50	0.82–2.77	0.19	144	0.84	0.47–1.51	0.57	*		
Improved latrine (89)	19	1.59	0.80–3.17	0.19	1.92	0.93–3.93	0.08	60	0.93	0.48–1.83	0.84	*		
Soap for handwashing available ^b (118)	33	0.99	0.61–1.61	0.98	*			79	1.27	0.77–2.08	0.35			
Household drinking water rainy season														
Tap source (37)	13	1.00						19	1.00					
Borehole water (249)	68	0.69	0.33–1.44	0.33	*			163	1.28	0.57–2.88	0.54	*		
Well (87)	25	0.74	0.33–1.69	0.48	*			64	1.42	0.56–3.63	0.46	*		
Rain water, surface water (12)	2	0.37	0.07–1.94	0.24	*			10	1.55	0.44–14.73	0.30	*		
Dry season														
Tap source (34)	13	1.00						17	1.00					
Borehole water (261)	68	0.57	0.27–1.20	0.14	0.83	0.55–1.26	0.39	175	1.27	0.53–3.03	0.59	*		
Well (81)	24	0.68	0.29–1.58	0.37	*			58	1.04	0.37–2.94	0.94	*		
Surface water (9)	3	0.81	0.17–3.80	0.79	*			6	0.73	0.13–4.11	0.72	*		
Household drinking water storage														
Open ^b (278)	77	0.94	0.57–1.54	0.80	*			191	1.53	0.93–2.51	0.10	1.58	0.95–2.60	0.08
Pot or canary (290)	80	1.00						191	1.00					
Basin or bowl (16)	5	1.19	0.40–3.54	0.75	*			13	1.63	0.44–6.12	0.47	*		
Canister (plastic jerrican) (59)	19	1.25	0.68–2.28	0.47	*			42	1.21	0.63–2.31	0.57	*		
Household drinking water treatment^b														
Prior to consumption ^e (69)	22	1.25	0.71–2.20	0.44	*			47	0.97	0.53–1.77	0.91	*		
Water contamination households^b														
Coliform bacteria (89)	29	2.42	0.27–21.64	0.43	*			58	1.45	0.24–8.92	0.69	*		
<i>Escherichia coli</i> (61)	22	1.83	0.71–4.74	0.21	*			41	1.08	0.40–2.89	0.89	*		
Faecal streptococci (88)	29	2.95	0.34–25.65	0.33	*			58	1.94	0.33–11.39	0.46	*		
Safe to drink (34)	0	na			*			0	na			*		
Water contamination children's drinking cups^b														
Coliform bacteria (101)	25	0.67	0.18–2.56	0.56	*			71	2.15	0.58–8.00	0.25	*		
<i>Escherichia coli</i> (55)	12	0.68	0.28–1.61	0.38	*			41	1.60	0.66–3.88	0.30	*		
Faecal streptococci (101)	29	na			*			68	0.58	0.13–2.51	0.47	*		
Safe to drink (61)	0	na			*			2	1.45	0.11–18.62	0.78	*		
Water contamination community sources^b														
Coliform bacteria (13)	8	1.89	0.48–7.49	0.36	*			12	7.20	0.80–65.05	0.08	5.10	0.40–64.25	0.21
<i>Escherichia coli</i> (9)	6	2.31	0.48–11.12	0.30	*			9	na			*		
Faecal streptococci (10)	7	2.92	0.62–13.76	0.18	3.85	0.47–31.34	0.21	9	4.50	0.49–41.25	0.18	2.31	0.14–39.26	0.56
Safe to drink (15)	9	0.35	0.09–1.36	0.13	0.30	0.06–1.58	0.16	14	0.27	0.05–1.51	0.14	0.28	0.03–2.80	0.28

^aA new variable for hygiene behaviour was created using factor analysis with the mode and frequency of handwashing. Children were classified into three categories with poor, middle and good hygiene behaviours.

^bThe odds ratio (OR) refers to the comparison “yes” vs “no”

^cOpen defaecation includes the category of defaecating in the bush and behind the latrines

^dOthers' includes homemakers, retirees and unemployed people

^eHouseholds reported to treat their drinking water through filtration and sedimentation

^fN = positive cases

^gAmong domestic animals held by children's caregivers (cats, cattle, dogs, goats, poultry, sheep and swine), we found a significant association between *Giardia intestinalis* infection in children and the possession of dogs (OR = 2.3, 95% CI 1.26–4.22, $\chi^2 = 7.26$, $df = 1$, $P = 0.007$; aOR = 2.1, 95 % CI 1.15–4.00, $\chi^2 = 14.42$, $df = 7$, $P = 0.016$).

* P -values are based on likelihood ratio test

** P -values are based on likelihood ratio tests between the multivariate regression models with and without the respective variable. The multivariate core model included a random intercept at the unit of the school and the categorical exposure variables sex, age group (8–11 years and 12–14 years), socioeconomic status, and project region, which were set a priori as potential confounders. All the other variables were assessed one by one and retained for the maximal model if their P -value was < 0.2 . The final model was then obtained using backward selection with the same level of 0.2.

9.1.2 Additional file 2. Table S2. Results from univariate and multivariate logistic regression analysis for parasitic infection

Risk factor	<i>Hymenolepis nana</i> (n = 25)							<i>Schistosoma haematobium</i> (n = 15)						Intestinal pathogenic protozoa (n = 290)							
	Univariate log. regression*				Multivariate log. regression**			Univariate log. regression*				Multivariate log. regression**		Univariate log. regression*				Multivariate log. regression**			
	N ^f	OR	95% CI	P	aOR	95% CI	P	N ^f	OR	95% CI	P	aOR	95% CI	P	N ^f	OR	95% CI	P	aOR	95% CI	P
Sex																					
Male (197)	14	1.00					8	1.00						146	1.00						
Female (188)	11	0.81	0.35–1.88	0.62	*		7	0.82	0.28–2.43	0.73	*			144	1.17	0.72–1.90	0.52	*			
Age group																					
8–11 yrs (251)	13	1.00					8	1.00						182	1.00						
12–14 yrs (143)	12	1.32	0.53–3.28	0.55	*		7	0.01	0.33–3.81	0.84	*			108	1.45	0.84–2.52	0.19	1.30	0.75–2.24	0.35	
Region																					
Centre-Ouest (187)	20	1.00					7	1.00						160	1.00						
Plateau Central (198)	5	0.20	0.05–0.80	0.02	0.32	0.09–1.15	0.08	8	2.65	0.04–160.76	0.64	*		130	0.32	0.18–0.57	< 0.001	0.33	0.18–0.58	< 0.001	
Hygiene behaviour^a																					
Middle third (2) (227)	14	1.00					12	1.00						174	1.00						
Lower third (1) (56)	1	0.33	0.04–2.66	0.30	*		0	na			*			41	0.79	0.38–1.64	0.53	*			
Higher third (3) (102)	10	1.63	0.68–3.92	0.28	*		3	0.45	0.12–1.72	0.24	*			75	0.84	0.48–1.47	0.54	*			
Drinking water for consumption^b																					
From home (239)	17	0.99	0.37–2.68	0.99	*		10	1.41	0.42–4.73	0.57	*			179	1.05	0.62–1.78	0.87	*			
From school (322)	15	0.25	0.07–0.85	0.03	0.37	0.15–0.91	0.02	11	0.33	0.07–1.62	0.17	0.31	0.06–1.61	0.16	246	1.17	0.55–2.46	0.68	*		
Water risk behaviours																					
No water contact (93)	7	1.00					1	1.00						70	1.00						
Playing (5)	0	na			*		0	na			*			3	0.32	0.05–2.21	0.25	*			
Fishing (25)	0	na			*		0	na			*			18	0.83	0.29–2.32	0.72	*			
Making laundry (56)	3	0.60	0.14–2.58	0.49	*		1	1.68	0.10–29.40	0.72	*			40	0.81	0.37–1.78	0.60	*			
Domestic chores (206)	15	0.69	0.24–1.94	0.48	*		13	7.38	0.89–61.15	0.06	2.04	1.05–3.97	0.04	159	1.07	0.58–1.97	0.84	*			
Any water contact ^c (292)	18	0.59	0.22–1.60	0.30	*		14	5.11	0.63–41.76	0.13	5.21	0.62–44.11	0.13	220	0.96	0.54–1.71	0.88	*			
Sanitary practices children																					
Using latrines at school (307)	15	1.00					12	1.00						231	1.00						
Using latrines at home/teacher's (7)	1	3.73	0.35–39.37	0.27	*		0	na			*			5	1.31	0.23–7.60	0.61	*			
Open defaecation at school ^e (71)	9	1.49	0.42–5.30	0.54	*		3	2.21	0.50–9.76	0.29	*			54	0.83	0.39–1.77	0.62	*			
Caregiver's education																					
Never went to school (288)	18	1.00					8	1.00						220	1.00						
Primary education (59)	4	0.97	0.30–3.12	0.96	*		6	4.48	1.34–14.95	0.02	1.32	0.60–2.91	0.49	47	1.24	0.81–2.54	0.55	*			
Secondary education (38)	3	1.58	0.41–6.17	0.51	*		1	0.72	0.08–6.49	0.77	*			23	0.62	0.29–1.32	0.22	*			
Caregiver's occupation^b																					
Agriculture (344)	23	1.00					15	1.00						264	1.00						
Civil service (8)	0	na			*		0	na			*			4	0.45	0.10–1.98	0.29	*			
Merchant (9)	1	1.63	0.18–15.11	0.67	*		0	na			*			8	2.34	0.27–20.41	0.44	*			
Others ^d (24)	1	0.43	0.05–3.53	0.43	*		0	na			*			14	0.39	0.15–0.97	0.004	0.74	0.55–0.99	0.05	
Animals^b																					
Possession of domestic animals (371)	25	na			*		15	na			*			281	1.28	0.40–4.17	0.68	*			
Animals held in the house (246)	19	1.76	0.67–4.62	0.25	*		9	0.75	0.24–2.29	0.61	*			186	1.00	0.61–1.64	0.99	*			
Household sanitary conditions																					
Traditional latrine (213)	3	1.00					5	1.00						61	1.00						

No latrines/ open defaecation (83)	19	1.84	0.47–7.25	0.38	*							9	2.61	0.29–23.73	0.39	*					164	1.08	0.58–2.00	0.81	*		
Improved latrine (89)	3	0.96	0.18–5.16	0.97	*							1	3.03	0.33–28.13	0.33	*					65	1.21	0.59–2.48	0.60	*		
Soap for handwashing available (118)	4	0.50	0.16–1.55	0.23	*							2	0.24	0.05–1.14	0.07	0.21	0.04–1.05	0.06			87	1.05	0.62–1.77	0.87	*		
Household drinking water																											
rainy season																											
Tap source (37)	3	1.00										3	1.00								23	1.00					
Borehole water (249)	7	0.31	0.07–1.36	0.12	1.81	0.96–3.40	0.07					6	0.51	0.10–2.70	0.43	*					188	1.55	0.68–3.57	0.30	*		
Well (87)	12	1.45	0.31–6.72	0.64	*							4	0.75	0.11–5.07	0.76	*					67	1.21	0.46–3.19	0.70	*		
Rain water, surface water (12)	3	2.85	0.38–21.33	0.31	*							2	8.62	0.56–132.44	0.12	1.75	0.68–4.51	0.25			12	na			*		
Dry season																											
Tap source (34)	2	1.00										3	1.00								22	1.00					
Borehole water (261)	9	0.53	0.10–2.79	0.45	*							7	0.57	0.11–2.98	0.50	*					198	1.21	0.49–2.98	0.68	*		
Well (81)	12	2.10	0.33–13.30	0.43	*							4	0.76	0.11–5.20	0.78	*					61	0.77	0.26–2.31	0.68	*		
Surface water (9)	2	3.33	0.01–0.30	0.33	*							1	1.95	0.11–36.16	0.65	*					9	na			*		
Household drinking water storage																											
Open ^b (278)	21	2.04	0.65–6.36	0.22	*							15	na			*					213	1.34	0.79–2.29	0.28	*		
Pot or canary (290)	18	1.00										13	1.00			*					218	1.00					
Basin or bowl (16)	2	1.31	0.25–6.87	0.75	*							0	na			*					14	1.75	0.37–8.23	0.48	*		
Canister (plastic jerrican) (59)	4	1.49	0.44–5.00	0.52	*							0	na			*					47	1.27	0.62–2.60	0.52	*		
Household drinking water treatment^a																											
Prior to consumption ^c (69)	7	1.18	0.44–3.20	0.74	*							4	1.13	0.32–4.05	0.85	*					51	0.79	0.41–1.50	0.46	*		
Water contamination households^b																											
Coliform bacteria (89)	2	na			*							3	0.38	0.02–6.62	0.51	*					69	3.30	0.56–19.56	0.19	0.96	0.12–7.63	0.97
<i>Escherichia coli</i> (61)	2	na			*							1	0.25	0.02–3.11	0.28	*					50	2.25	0.81–6.25	0.12	1.11	0.32–3.87	0.87
Faecal streptococci (88)	2	na			*							2	0.09	0.01–1.32	0.08	0.11	0.01–2.73	0.18			69	5.30	0.92–30.35	0.06	2.41	0.32–18.06	0.39
Safe to drink (34)	0	na			*							0	na			*					0	na					
Water contamination children's drinking cups^b																											
Coliform bacteria (101)	6	0.18	0.01–4.61	0.30	*							4	na			*					78	1.13	0.28–4.52	0.86	*		
<i>Escherichia coli</i> (55)	4	0.70	0.11–4.46	0.71	*							3	3.29	0.33–32.61	0.31	*					42	0.93	0.39–2.24	0.88	*		
Faecal streptococci (101)	7	na			*							3	0.31	0.02–3.94	0.37	*					78	1.13	0.28–4.52	0.86	*		
Safe to drink (61)	0	na			*							0	na			*					2	0.59	0.05–6.76	0.67	*		
Water contamination community sources^b																											
Coliform bacteria (13)	1	1.92	0.11–33.41	0.66	*							0	na			*					12	1.09	0.09–13.31	0.95	*		
<i>Escherichia coli</i> (9)	1	3.38	0.19–60.24	0.41	*							0	na			*					9	na			*		
Faecal streptococci (10)	1	2.89	0.16–51.13	0.47	*							0	na			*					10	na			*		
Safe to drink (15)	1	0.67	0.04–11.56	0.78	*							0	na			*					21	0.71	0.06–8.66	0.79	*		

^aA new variable for hygiene behaviour was created using factor analysis with the mode and frequency of handwashing. Children were classified into three categories with poor, middle and good hygiene behaviours.

^bThe odds ratio (OR) refers to the comparison “yes” vs “no”

^cOpen defaecation includes the category of defaecating in the bush and behind the latrines

^d‘Others’ includes homemakers, retirees and unemployed people

^eHouseholds reported to treat their drinking water through filtration and sedimentation

^fN = positive cases

*P-values are based on likelihood ratio tests

**P-values are based on likelihood ratio tests between the multivariate regression models with and without the respective variable. The multivariate core model included a random intercept at the unit of the school and the categorical exposure variables sex, age group (8–11 years and 12–14 years), socioeconomic status, and project region, which were set a priori as potential confounders. All the other variables were assessed one by one and retained for the maximal model if their P-value was < 0.2. The final model was then obtained using backward selection with the same level of 0.2.

9.2 School children’s intestinal parasite and nutritional status one year after complementary school garden, nutrition, water, sanitation, and hygiene interventions in Burkina Faso

9.2.1 Supplemental Table 1: CONSORT 2010 checklist of information to include when reporting a randomized trial

Section/topic	Item no.	Checklist item	Reported in section
Title and abstract			
	1a	Identification as a randomized trial in the title	Title
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts)	Abstract
Introduction			
Background and objectives	2a	Scientific background and explanation of rationale	Introduction
	2b	Specific objectives or hypotheses	Introduction
Methods			
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	Materials and Methods (Sample size, sampling method, and study design)
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons	Materials and Methods (Sample size, sampling method, and study design)
Participants	4a	Eligibility criteria for participants	Materials and Methods (Sample size, sampling method, and study design)
	4b	Settings and locations where the data were collected	Materials and Methods (Sample size, sampling method, and study design)
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	Materials and Methods (Complementary school garden, nutrition, and WASH interventions)
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed	Materials and Methods (Outcome definition and measurement)
	6b	Any changes to trial outcomes after the trial commenced, with reasons	NA
Sample size	7a	How sample size was determined	Materials and Methods (Sample size, sampling method, and study design)
Randomisation:	7b	When applicable, explanation of any interim analyses and stopping guidelines	NA
	8a	Method used to generate the random allocation sequence	Materials and Methods (Sample size, sampling method, and study design)
Sequence generation	8b	Type of randomisation; details of any restriction (such as blocking and block size)	Materials and Methods (Sample size, sampling method, and study design)
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned	NA

Implementation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	Materials and Methods (Sample size, sampling method, and study design)
Blinding	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those assessing outcomes) and how	NA
	11b	If relevant, description of the similarity of interventions	NA
	12a	Statistical methods used to compare groups for primary and secondary outcomes	Materials and Methods (Statistical analysis)
Statistical methods	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses	Materials and Methods (Statistical analysis)
	Results		
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	Figure 2
	13b	For each group, losses and exclusions after randomisation, together with reasons	Figure 2
	14a	Dates defining the periods of recruitment and follow-up	Materials and Methods (Outcome definition and measurement)
Recruitment	14b	Why the trial ended or was stopped	Materials and Methods (Sample size, sampling method, and study design)
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	Table 1
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	Figure 1, Results (Compliance and characteristics of study population)
	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	Tables 2-5, Results
Outcomes and estimation	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended	Tables 2-5, Results
	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory	Supplemental Tables 2 and 3
Ancillary analyses	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	NA
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	NA
Discussion			
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses	Discussion: Limitations
Generalisability	21	Generalisability (external validity, applicability) of the trial findings	Discussion: Limitations
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence	Discussion
Other information			
Registration	23	Registration number and name of trial registry	Materials and Methods (Ethical considerations)
Protocol	24	Where the full trial protocol can be accessed, if available	Reference to study protocol (No 27)
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders	Financial support

9.2.2 Supplemental Table 2: Number and percentage of school children with newly occurring (incidence) and persistent adverse health outcomes, in February/March 2015 and one year later

	Incidence* control	Incidence* intervention	Persistence† control	Persistence† intervention	Total control 2015	Total intervention 2015	Total control 2016	Total intervention 2016
Nutritional indicators (n [%])								
Total undernutrition	15 (11.7)	12 (12.0)	47 (83.9)	69 (90.8)	56 (30.4)	76 (43.2)	62 (33.7)	81 (46.0)
Stunting (low height-for-age)	11 (7.8)	13 (11.9)	37 (86.1)	61 (91.0)	43 (23.4)	67 (38.1)	48 (26.1)	74 (42.1)
Thinness (low BMI-for-age)	9 (5.5)	10 (6.5)	14 (70.0)	16 (72.7)	22 (12.5)	20 (10.9)	23 (12.5)	26 (14.8)
Overweight (high BMI -for-age)	5 (2.8)	1 (0.6)	5 (83.3)	2 (100.0)	6 (3.3)	2 (1.4)	10 (5.4)	1 (0.6)
Anemia	40 (29.6)	39 (32.0)	28 (57.1)	24 (44.4)	49 (26.6)	54 (30.7)	68 (37.0)	63 (35.8)
Intestinal parasitic infections (n [%])								
Total intestinal parasites	14 (41.2)	7 (41.2)	54 (36.0)	55 (34.6)	150 (81.5)	159 (90.3)	133 (72.3)	109 (61.9)
Total intestinal protozoa	23 (62.2)	7 (35.0)	106 (72.1)	94 (60.3)	147 (79.9)	156 (88.6)	129 (70.1)	101 (57.4)
<i>Entamoeba histolytica/E. dispar</i>	35 (50.7)	16 (30.2)	53 (46.1)	49 (39.8)	115 (62.5)	123 (69.9)	88 (47.8)	65 (36.9)
<i>Giardia intestinalis</i>	27 (20.0)	26 (21.1)	19 (38.8)	18 (34.0)	49 (26.6)	53 (30.1)	46 (25.0)	44 (25.0)
<i>Trichomonas intestinalis</i>	19 (12.8)	14 (11.0)	5 (13.9)	7 (14.3)	36 (19.6)	49 (27.8)	24 (13.0)	21 (11.9)
<i>Entamoeba coli</i>	25 (22.9)	21 (18.1)	23 (30.7)	7 (11.7)	75 (40.8)	60 (34.1)	48 (26.1)	28 (15.9)
Total helminths	10 (6.0)	8 (5.3)	9 (50.0)	6 (30.0)	18 (9.8)	20 (11.4)	19 (10.3)	14 (8.0)
<i>Hymenolepis nana</i>	3 (1.7)	4 (2.5)	7 (63.6)	5 (38.5)	11 (6.0)	13 (7.4)	10 (5.4)	9 (5.1)
<i>Schistosoma haematobium</i>	7 (3.9)	5 (3.0)	2 (40.0)	0 (0.0)	5 (2.7)	7 (4.0)	9 (4.9)	5 (2.8)

BMI = body mass index

* Subsample of children without respective adverse health outcome in 2015.

† Subsample of children with respective adverse health outcome in 2015.

9.2.3 Supplemental Table 3: Intervention effects* on newly emerging (incidence) and persistent adverse health outcomes in the cohort of school children in two regions of Burkina Faso, in February/March 2015 and one year later

	OR incidence†	OR persistence‡	P-value difference§
Nutritional indicators			
Total undernutrition	0.9 (0.4, 2.0)	1.5 (0.5, 4.6)	0.421
Stunting (low height-for-age)¶	1.6 (0.7, 3.7)	1.6 (0.5, 5.5)	0.968
Thinness (low BMI-for-age)	1.1 (0.4, 2.8)	1.3 (0.3, 5.6)	0.810
Overweight (high BMI-for-age)¶	0.2 (0.0, 2.0)	n/a	0.993
Anemia	1.0 (0.4, 2.6)	0.5 (0.2, 1.5)	0.162
Intestinal parasitic infections			
Total intestinal parasites	0.3 (0.1, 1.2)	0.6 (0.3, 1.5)	0.230
Total intestinal protozoa	0.3 (0.1, 1.6)	0.5 (0.2, 1.5)	0.532
<i>Entamoeba histolytica</i> / <i>E. dispar</i>	0.4 (0.1, 1.2)	0.7 (0.3, 1.5)	0.359
<i>Giardia intestinalis</i>	1.0 (0.4, 2.8)	0.7 (0.2, 2.4)	0.606
<i>Trichomonas intestinalis</i>	0.8 (0.3, 2.1)	1.1 (0.3, 4.3)	0.767
<i>Entamoeba coli</i>	0.7 (0.3, 1.3)	0.3 (0.1, 0.7)	0.151
Total helminths	0.8 (0.3, 2.2)	0.3 (0.1, 1.5)	0.343
<i>Hymenolepis nana</i> ¶	1.4 (0.3, 6.5)	0.4 (0.1, 1.9)	0.227
<i>Schistosoma haematobium</i> ¶	0.7 (0.2, 3.2)	0.0 (0)	0.991

CI = confidence interval; n/a = not applicable; OR = odds ratio; SES = socioeconomic status.

* The intervention effect is given by the OR (with 95% CI) associated with the factor intervention. Analyses involved mixed logistic regression models with random intercepts for schools, adjusted for the two categorical SES variables and children's age.

† Subsample of children without respective adverse health outcome in 2015.

‡ Subsample of children with respective adverse health outcome in 2015.

§ The P-value reflects the difference in outcomes between the two intervention effect ORs.

¶ The mixed logistic regression model was not adjusted for SES variables or children's age, as no convergence in the regression models were achieved.

Curriculum vitae

PERSONAL INFORMATION

Full name	Séverine Erismann
Email	severine.erismann@swisstph.ch, severine.erismann@gmail.com
Date of birth	14 December 1985
Place of birth	Zürich (ZH)
Place of origin	Schlossrued (AG)
Nationality	Swiss
Languages	German (mother tongue), French (mother tongue), English (fluent), Spanish (good), Italian (good), Portuguese (basics)

EDUCATION

01/2014 – 12/2016	<p>PhD Candidate in Epidemiology and Public Health, Department of Epidemiology and Public Health, Ecosystem Health Science Unit, Swiss Tropical and Public Health Institute, Switzerland</p> <p><i>Research interests:</i> School-aged children’s nutrition and health, neglected tropical diseases, nutrition- and health- sensitive agriculture, water, sanitation and hygiene (WASH)</p> <p><i>Courses:</i> Epidemiology, public health, human nutrition, water and sanitation, neglected tropical diseases, quantitative and qualitative methods</p>
2014	<p>Graduate Certificate in Delivery Sciences for International Nutrition, TUFTS University, Friedman School of Nutrition Science and Policy</p> <p><i>Courses:</i> Nutrition programme development and delivery, theories of behavior change and positive deviance, monitoring and evaluation of nutrition programmes</p>
2009 – 2011	<p>Master in Development Studies at the Graduate Institute for International and Development Studies (IHEID), Switzerland (Geneva)</p> <p>Specialization: Global ecology and sustainable development</p> <p>Thesis: ‘Gaps in agri-biotech companies’ biodiversity conservation strategies?’</p>
2005 – 2008	<p>Bachelor of Science, University of Fribourg, Switzerland</p> <p>Major in geography; minors in political sciences and media and communications</p>
03/2007 – 07/2007	<p>International Student Exchange Programme, Pontificia Universidad Católica de Valparaíso, Chile</p> <p>Studies in geography, journalism and Spanish</p>
1999 – 2005	<p>High School Alpenquai Lucerne, Switzerland</p> <p>Major in biology and chemistry</p>

EMPLOYMENT HISTORY

01/2014 – present	<p>Research Fellow for the “Vegetables go to School: Improving Nutrition through Agricultural Diversification” project by the Swiss Development Cooperation with the Swiss Tropical and Public Health Institute, Switzerland (Basel)</p> <ul style="list-style-type: none"> • Contribution to project-related research activities and development of integrated research tools in nutrition and health • Support in planning, development and implementation of nutrition and WASH interventions
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- Reporting and monitoring of project activities
 - Support in budgetary, strategic and operational planning
 - Development of several studies (clinical studies, questionnaire surveys, environmental assessments)
- 05/2013 – 11/2013 **Junior Consultant / Interim Focal Point Agriculture and Food Security (A+FS) Network, Global Programme Food Security, Swiss Development Cooperation, Switzerland (Bern)**
- Animation of the A+FS Network and provision of thematic and practical support
 - Coordination of the postharvest management programme in sub-Saharan Africa, the Young Professionals' Platform for Agricultural Research and the Global Donor Platform for Rural Development
- 03/2012 – 04/2013 **Assistant Project Manager, Permanent Representation of Switzerland to FAO, IFAD and WFP, Italy (Rome)**
- *Support to Policy Dialogue*: Preparation and participation in WFP's and IFAD's Executive Boards, seminars and operational briefings and contribution to the formulation of Swiss positions through relevant background information
 - *Analytical research and conceptual work*: Preparation of background documents on the Rome-based agencies' and SDC's engagement in "Resilience" and "Gender"
- 09/2011 – 02/2012 **Project Assistant, Sustainable Business Associates, Switzerland (Lausanne)**
- Developing a waste management project in Tunisia, including a two week mission to Tunisia for the elaboration of the project proposal
 - Coordinating projects in sustainable tourism in Morocco, ISO 14001 certification in Morocco, and environmental economic assessments in Mozambique
- 02/2009 – 08/2009 **Internship, Office of the High Commissioner for Human Rights (OHCHR), Switzerland (Geneva)**
- Providing support to the Indigenous Peoples and Minorities Section
 - Thematic research on Indigenous Peoples rights in international law, especially on international jurisprudence on land and resource rights
- 05/2008 – 12/2008 **Internship, Verkehrsclub der Schweiz (VCS), Switzerland (Bern)**
- Planning and implementation of the winter campaign against air pollution (pm10)
- 08/2006 – 09/2006 **Course Assistant, Swiss Red Cross, Switzerland (Bern)**
- Organization of the Emergency Response Unit (ERU) course at the center of emergency response, first aid and rescue operations
 - Active participation and training in logistics during the ERU course

VOLUNTARY WORK

-
- 01/2010 – 09/2010 Project Coordinator intercultural study trip to Brazil, Initiative for Intercultural Learning (IFIL), Switzerland (Geneva) and Brazil (São Paulo)
- 08/2005 – 09/2005 English teacher in a primary school in Puerto Ayora, Galapagos Islands, Ecuador

PROFESSIONAL TRAINING

-
- 08/2015 **Water and Sanitation Engineering: from Emergency towards Development**
A 10-day training on WASH in emergency and development contexts by the University of Neuchâtel and the International Committee of the Red Cross, Switzerland (Neuchâtel)
- 06/2013 **Management of Networks**: A two-day training and learning event on the organization and the management of networks organized by the SDC, Switzerland (Fribourg)

01/2013	Conducting and Monitoring Climate Change Adaptation Projects: A two-day exchange and learning event organized by the SDC, Switzerland (Bern)
6/2011	Workshop on Embedding Sustainable Agriculture Strategies in Companies A two-day training organized by the International Institute for Management Development (IMD), Center for Corporate Sustainability Management and the Sustainable Agriculture Initiative Platform (SAI), Switzerland (Lausanne)
2002-2003	Youth for Understanding (YFU) exchange year, Pennsylvania, USA High school exchange year in Mechanicsburg

PUBLICATIONS

Erismann S, Diagbouga S, Schindler C, Odermatt P, Knoblauch A, Gerold J, et al. School children's intestinal parasite and nutritional status 1 year after complementary school garden, nutrition, water, sanitation, and hygiene Interventions in Burkina Faso. *American Journal of Tropical Medicine and Hygiene*. 2017; 93: 904-913.

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Shrestha A, Schindler C, Gerold J, Odermatt P, Erismann S, Sharma S, et al (2017). Intestinal parasitic infections and risk factors among school-aged children in Dolakha and Ramechhap districts, Nepal. *Parasites & Vectors* (submitted).

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Shrestha A, Sharma S, Gerold J, Erismann S, Sagar S, Koju R, et al. Water quality, sanitation and hygiene conditions in schools and households in Dolakha and Ramechhap districts, Nepal: results from a cross-sectional survey. *International Journal of Environmental Research and Public Health*. 2017; 14:89.

Diagbouga S, Kientega T, Erismann S, Ouermi D, Saric J, Odermatt P, et al. Evaluation of a Real-Time Polymerase Chain Reaction for the Laboratory Diagnosis of *Giardia intestinalis* in Stool Samples from Schoolchildren from the Centre-Ouest and Plateau Central Regions of Burkina Faso. *Appl Micro Open Access*. 2017; 3:1.

Erismann S, Diagbouga S, Odermatt P, Knoblauch A, Gerold J, Shrestha A, et al. Prevalence of intestinal parasitic infections and associated risk factors among schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso. *Parasites & Vectors*. 2016; 9:554.

Erismann S, Shrestha A, Diagbouga S, Knoblauch A, Gerold J, Herz R et al. Complementary school garden, nutrition, water, sanitation and hygiene interventions to improve children's nutrition and health status in Burkina Faso and Nepal: a study protocol. *BMC Public Health*. 2016;16:244.

CONGRESS PARTICIPATION

10/2015	12th European Nutrition Conference (FENS), Berlin, Germany. <i>Inadequate dietary and hygiene practices and knowledge of school-aged children in Burkina Faso</i> . Poster presentation.
09/2015	European Congress on Tropical Medicine and International Health (ECTMIH), Basel, Switzerland. <i>High intestinal parasitic infections and malnutrition among school-aged children in Burkina Faso</i> . Oral presentation.