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EDUCATION OUTSIDE THE CLASSROOM – IMPLICATIONS FOR STUDENTS' PHYSICAL ACTIVITY, STRESS REGULATION, LEARNING AND SOCIAL DIMENSIONS

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List of Abbreviations

ANOVA	Analysis of variance
AUC	Area under the curve
AUCg	Area under the curve with respect to ground
AUCi	Area under the curve with respect to increase
BHLM	Bayesian hierarchical linear model
CG	Control group
CRI	Credible interval
cfDNA	Circulating cell-free deoxyribonucleic acid
CoDA	Compositional Data Analysis
EOtC	Education Outside the Classroom
GE	Green Exercise
IG	Intervention group
LMM	Linear mixed-effects model
LPA	Light physical activity
MCMC	Markov chain Monte Carlo
MVPA	Moderate-to-vigorous physical activity
NHST	Null-hypothesis-significance testing
OE	Outdoor Education
PA	Physical activity
PE	Physical education
PR	Peak reactivity
SB	Sedentary behaviour
UK	United Kingdom
USA	United States of America
WHO	World Health Organization

1 Summary

Many children and adolescents do not address the recommended levels of physical activity, are obese and suffer from psychological disorders. Possible causes lie in changes in human lifestyles and living environments during the last decades: a rapid urbanisation; a more common use of digital media; a shift towards unhealthy diet; and an achievement-oriented society. These circumstances can have a great impact on one's health status in later life and should urgently be addressed. The literature indicated that green residential areas and the active usage of natural green or blue environments are associated with mental and physical health benefits. Nature therefore provides the potential to promote one's health development. The compulsory educational systems in western countries offers great opportunities to reach every child in terms of educational programs that foster physical activity and mental well-being. At the same time, due to long school days and a shift towards full-day schools, these are very important living environments for children and adolescents. Therefore, great obligations to promote children's PA and mental well-being can be attributed to educational systems and innovative teaching concepts are therefore necessary. Education Outside the Classroom is defined as regular compulsory school and curriculum-based education in a natural or cultural setting outside the school building. Especially in Scandinavian countries, Education Outside the Classroom is widely practiced. This dissertation thesis encompasses three articles that are published in international peer-reviewed journals. In a systematic literature review, 13 peer-reviewed articles were identified and the existing evidence of Education Outside the Classroom was summarized and critically appraised. Education Outside the Classroom seems to promote students in terms of social, academic, physical and psychological dimensions. The methodological quality has to be considered as moderate-to-low and more rigorously designed research studies are necessary warranted. Based on an explorative longitudinal quasi-experimental study, in two articles the associations between students' stress responses, sedentary behaviour and physical activity during Education Outside the Classroom and normal indoor teaching settings was examined. Students' cortisol and circulating cell-free deoxyribonucleic acid as well as their sedentary behaviour and physical activity was assessed on three measuring points during the school year 2014 / 2015. In article 2, fitted linear mixed-effects models revealed that students within the intervention group showed a steady decline of cortisol during the school day, which is an indicator of a healthy diurnal cortisol biorhythm. Furthermore, students

within the intervention group showed higher levels of moderate-to-vigorous physical activity in comparison to students within the control group. In article 3, fitted Bayesian hierarchical linear models revealed that relative high amounts of light physical activity during Education Outside the Classroom were associated with the decrease of cortisol levels from morning to noon. The explorative study indicates that Education Outside the Classroom can promote students physical activity and that it might have a positive impact on their diurnal cortisol rhythm.

Future research studies are encouraged to continue investigations on students' physical and psychological health status, academic achievement, motivational aspects and social relations in comparison of regular compulsory indoor and outdoor teaching situations. Study designs and methodologies of high quality are needed to inform educational policy and public regarding beneficial aspects of Education Outside the Classroom. The dissemination of Education Outside the Classroom in Germany and other German speaking countries has not been systematically assessed, but it is approximately rather scarce in comparison to Scandinavian countries. Therefore, opportunities to conduct robust large-scale studies that account for specific circumstances of national or regional educational systems are limited. The dissemination of Education Outside the Classroom in Germany and other German speaking countries should be assessed and the development of a network between practitioners, educators and researchers following the example of Denmark could be beneficial.

2 Introduction

Nowadays, children and adolescents spend many waking hours per day in schools, many do not achieve to fulfil physical activity guidelines, they less frequently visit natural places, and they more often suffer from psychological as well as learning disorders in comparison to previous generations. Furthermore, they are encouraged to learn more curricular contents in relatively shorter periods to be well prepared with academic knowledge and skills to succeed in diverse future labour markets. Considering these circumstances, questions arise on how the compulsory educational system can tackle these challenges and if there are existing promising approaches that aim to antagonize such detrimental developments.

In chapter 2, the state of research regarding physical activity, mental wellbeing and cognition in relation to education and nature are discussed. Furthermore, different concepts of Outdoor Education are described and empirical results of short- and long-term studies are discussed. The underlying concepts of Education Outside the Classroom are linked to theoretical assumptions regarding health, cognition and motivation. Following the theoretical foundation, the state of research and identified gaps in the existing literature, guiding research aims for this dissertation thesis are formulated. Chapter 3 provides a detailed overview regarding the methods applied in study 1 and study 2, chapter 4 summarizes the three published peer-reviewed articles, and chapter 5 provides a general discussion and conclusion.

2.1 Physical Activity, Mental Wellbeing and Cognition in the Educational Context

A great number of children and adolescents do not address the recommended levels of physical activity (PA), (1-4), are obese (5, 6) and suffer from psychological (7-9) as well as learning (10, 11) disorders. These circumstances can lead to a detrimental health status in ones later life (12-14). Several changes in human lifestyles and living environments during the last decades can partly be responsible for this drawback in societies general health status: a rapid urbanisation (15), a more common use of digital media (16), a shift towards unhealthy diet (17) and an achievement-oriented society, already starting in school (4, 18, 19). Several measures and programmes have been developed and implemented to counteract these developments. The World Health Organization (WHO) has recently proposed a Global Action Plan on Physical Activity (20) and stated that “Quality physical education and supportive school environments can provide physical and health literacy for long-lasting healthy, active lifestyles.” (20 p. 20). Therefore, the compulsory educational systems in western countries offer a great chance to reach every child in early age, because children spend a considerable amount of their waking hours in school (21). Physical education (PE) has an important role for students PA and health literacy, but PE cannot solve the PA and health crisis alone. That becomes more obvious in the worldwide UNESCO PE report (22): primary schoolchildren receive on average only 103 minutes of PE weekly (range: 25–220 minutes), PE as a subject and PE teachers often have a lower status than other subjects or other teachers and PE lessons are more often cancelled in comparison to other subjects. That is because the main subjects are often considered more important for students’ overall academic development in comparison to PE lessons. Instead of promoting and increasing PE and PA in school, as lately demanded by the WHO (20), both aspects still have a rather low status. That is especially important, as the literature suggest that “adding time to ‘academic’ or ‘curricular’ subjects by taking time from physical education programmes does not enhance grades in these subjects and may be detrimental to health” (23 p. 1) and positive associations between PA and cognitive and academic performance have been reported (24, 25). That the school environments—including compulsory lessons, extracurricular activities and the physical environments—support possibilities for being physically active is of special interest from a German perspective. The dissemination of all-day schools consistently increased in Germany for all school forms and class levels: e.g. from 10,3 % in 2002 to 68,2 % in 2017 in primary schools and from 12,2 % in 2002 to 64,2 % 2017 in grammar schools (26, 27). Therefore, more children and adolescents have less leisure time for being physically active. As a conclusion, a great potential and a great obligation to focus on children’s overall health can be attributed to the compulsory school system. Innovative teaching concepts that not only involve PE lessons, but rather focus on interdisciplinary teaching and that incorporate environments outside the school building can be one solution to promote children’s PA, mental well-being and overall health. In such teaching concepts, the teachers often intentionally visit natural green or blue environments as they provide rich learning opportunities via real world encounter (28, 29) (see chapter 2.3 Outdoor Education). Besides these learning opportunities, being in nature and being physically active in nature can have several important health benefits. Therefore, the relationships of PA, health and nature are elaborated in the following chapter.

2.2 Physical Activity, Mental Wellbeing and Cognition in Relation to Nature

It has been assumed that synergistic health benefits exist if one is being physically active while directly being exposed to the natural world (30). Therefore, the concept of Green Exercise (GE) has been developed by the Green Exercise Research Group at the University of Essex. Studies conducted by this group suggest positive effects of being physically active in the presence of real or artificial nature on short and long-term health outcomes in adults: improved memory capacity, enhanced mental well-being, greater feelings of revitalization and increased energy (31-34). In their recent systematic literature review, Mnich et al. (35) especially focused on children and adolescents and concluded that GE might be beneficial from a psychosocial and physiological health perspective. However, due to heterogeneity and overall methodological quality of published research for these age groups, the authors conclude that more rigorous research designs are needed to better understand the health benefits of GE. Additionally, in comparison to the potential acute and direct health effects of the active engagement in nature as described in the GE literature, the characteristics of human living environments seem to have an influential effect regarding one's health and well-being. To grow up in proximity to green environments or to move to green environments seems to be associated with positive effects on stress resilience and it can be seen as a preventive factor for psychological diseases (36-38). In her systematic literature review, McCormick (39) researched the associations between children's access to green space and their mental well-being. Children's and adolescents' access to green spaces can be positively associated with several health-related outcomes: "attention restoration, memory, competence, supportive social groups, self-discipline, moderates stress, improves behaviours and symptoms of ADHD and was even associated with higher standardized test scores" (39 p. 3). In their 12-month longitudinal study, Dadvand et al. (40) applied a comprehensive approach to research associations between primary schoolchildren's living environments and their cognitive development: high-resolution satellite data was used to assess the amount of green spaces at home, at school and during commuting while also controlling for air pollution rates. The results indicate a better cognitive development of primary schoolchildren when they were exposed to green spaces next to their near home, during commuting to school and especially in the school surroundings. The reduction of the air pollution rate partially meditated the positive associations of green space on cognitive developments.

These positive relationships between PA, health, cognition and nature are of great relevance, because students spend many waking hours in school and while commuting to school (see chapter 2.1). If educational concepts consider these potential benefits and intentionally visit natural green or blue environments more often for curricular purposes, it could imply positive impacts on children and adolescents overall development. The historical developments, different aims and meanings, the dissemination and potential benefits of such teaching concepts are further described in the following chapter 2.3 Outdoor Education.

2.3 Outdoor Education

Outdoor Education (OE) involves many of the aforementioned aspects, as it potentially promotes PA, cognition and well-being and often involves natural environments. A general definition of OE is hardly possible because of several different meanings, aims and practises in relation to countries and cultures and several different terms are used for somehow related forms of educational practise (41-43). The local traditions of OE have originated in relation to specific circumstances: e.g. geographical landscapes and the public access to wilderness areas; cultural traditions in relation to encounter with nature; economic aspects such as the financial endowment of the education sector; or political contexts such as the long lasting political will to implement OE in formal curricula. Therefore, OE has its different shapes depending on different countries and regions and to what degree concepts from other regions have been adapted by practitioners (29, 43, 44). A major commonality for all outdoor-related educational practise is using direct experiences from a participant's perspective. The educators (in both public and in private educational settings) take their participants out of their familiar environment to enable these direct experiences by real world encounters. In the Anglo-American contexts and in the United Kingdom (UK), OE is often understood and practised as nature-based and often adventure-related activities that aim to provide nature experiences, personal growth and learning, and skill- and leadership development for different age and target groups (e.g. 45, 46, 47). These OE developments are historically inspired, amongst others, by the Outward Bound programmes initiated by Kurt Hahn (48). His strong emphasis on character development, social service, physical fitness and intellectual studies, and the focus on expeditions served as guiding principles for many OE programmes. From a Scandinavian perspective, OE practise is closely linked to the so-called *friluftsliv* (49), a term that cannot be translated, but has meanings in relation to *being in nature*. For Norwegians, friluftsliv is a strong cultural connection to nature. Motives to spent leisure time in nature involve e.g. recreation, encounter with the nature or the silence of nature. However, a clear definition is not possible, as friluftsliv has very individually meanings and is individually practised. In comparison to other outdoor and nature related activities, in which the time being outdoors in nature is often seen as an escape from the normal everyday live, the leading Norwegian friluftsliv pioneer Niels Faarlund emphasises rather the opposite as he describes that "Nature is the Home of Culture" and therefore "Friluftsliv is a Way Home" (50 p. 395). Many OE concepts and programmes especially involve school-related and curriculum-related contents (41), which may vary in duration, regularity, aims and targeted age groups. For instance, forest schools in the UK especially focus on children between the age 3 to 9 and the approach is inspired by Scandinavian traditions and practise to visit local natural environments for different activities, e.g. outdoor play, explore and describe nature phenomenon's and gardening (51, 52). In the context of compulsory school education, both short-term overnight residential trips and long-term, regular Education Outside the Classroom (EOtC) can be subsumed under the umbrella-term of OE. This dissertation thesis explicitly sets focus on OE in relation to formal education in school. Therefore, in the following subchapters, both forms of short-term overnight residential trips and EOtC are described in terms of their background, aims and research results.

2.3.1 Residential Outdoor Education

Short-term overnight residential outdoor education trips have a long tradition especially in the Anglo-Saxon countries. For instance, Higgins (43) describes the history of OE in Scotland as a combination of rich wilderness landscapes, a great interest in adventures and growth in wealth and leisure-time in end of the 19^{th} and the first half of the 20^{th} century. These circumstances lead to great provision of most often residential-based OE activities. In 1978, all Scottish secondary schools had firmly integrated OE programmes with professional employed staff. Furthermore, in 1982 approximately 163 residential centres offered courses for public and private educational purposes. However, the provision of formalised and non-formalised residential OE declined. Many residential centres were closed and only a few schools offered programmes in 1998. That was due to high financial costs of the residential trips and the maintenance of the often publicly funded residential centres (53). These developments also inspired the implementation of the so-called OE programme *Journeys outside the classroom*, that is located in the nearby school environments (54; see chapter 2.3.2) A similar historical development of OE can be seen for example in New Zealand: with a shift from most often recreational activities over residential-based OE programmes towards more and more formal curricular-embedded and school-related concepts (55).

In general, the offered programmes often focus on social and personal development, for instance leadership and survival skills, communication or teamwork, and environmental contents like nature conservation, but also more curriculum-related contents of natural and social science subjects (55-59). Different research studies have evaluated the potential benefits of curriculum-related residential OE programmes. Christi et al. (59) concluded that the published literature provides only limited research evidence regarding the educational benefits enabled by UK-based residential OE programmes. Their own study comprised more than 800 students and provided rather inconclusive results: potential educational benefits were found based on qualitative methods; no such benefits were found based on quantitative methods. Dettweiler et al. (60, 61) evaluated a residential outdoor education programme conducted in a German alpine region regarding potential educational benefits in learning motivation. The one-week programme seems to promote lower secondary students intrinsic motivation towards science class contents. To perceive oneself as competent during the class seems to be especially important for the development of intrinsic motivation. The authors concluded that those teaching techniques that have successfully been applied in residential OE programmes should be implemented more often on a regular basis within the normal school curriculum. The nearby school environments offer many opportunities to teach relevant curricular contents outside the school building. These assumptions are in concordance with increased provision of the aforementioned *Journeys outside the classroom* (54). In the following chapter, the mentioned regular, curriculum-related OE programmes that take place in the nearby school environment, are described in detail.

2.3.2 Education Outside the Classroom

Regular weekly or bi-weekly compulsory school and curriculum-based education in a natural or cultural setting outside the school building is often described as EOtC (29). The use of the term is based mainly on the descriptions of teaching concepts called *udeskole* or *uteskole* from Denmark and Norway, and has since been used in several scientific articles from outside Scandinavia. As udeskole in Denmark is considered a grassroots movement initiated by enthusiastic teachers and not a governmental programme, it has by nature different shades in its meanings and practise. EOtC should therefore not be understood as an exclusive definition or gold standard to describe certain teaching practises. The proposed definition is intended above all to facilitate comparisons of OE projects and research results from different countries and cultures (29). Besides this discourse in the European scientific community, the term *Education Outside the Classroom* is furthermore used in other settings and countries. For instance, the New Zealand Department of Education used the term already in the 1980s, as described by Boyes (56) and specific "EOTC Guidelines" on how out-of-school environments can be used for curricular purposes are still provided by the New Zealand Ministry of Education (62). Furthermore, the Education and Skills Committee of the UK House of Commons published its report on *Education Outside the Classroom* in 2005 (63). It emphasises the great potential in curriculum-related OE and at the same time the rather patchy provision by schools in UK. Greater efforts were called for at various levels (department, local authorities, and schools) to enable outdoor learning opportunities for all students.

In this dissertation thesis, the term EOtC refers to educational practises outside the school building that are closely related to the formal curriculum and that are conducted on a regular weekly or bi-weekly basis. EOtC involves educational programmes that have country-specific different terms such as learning outside the classroom, udeskole/uteskole, expeditionary learning, Draussenenschule/Draussenunterricht, journeys outside the classroom or forest school. It has to be mentioned that local projects and initiatives which use these terms need to be individually inspected regarding their educational practise and the respective overlapping's with the core scientific EOtC definitions.

From a historical perspective, the aforementioned grassroots movement of udeskole in Denmark was shaped and influenced by the similar uteskole practises from Norway (64, 65), in which the teachers use natural and cultural places to teach curricular contents. Both concepts are inspired by the aforementioned Scandinavian tradition of friluftsliv (49). A small group of Danish teachers began with udeskole in the 1990s and the grassroots movement led to a widespread provision in Denmark. In 2014, 17.9% of all public schools and 19.4% of all independent and private schools practiced udeskole with at least one class (66). That is furthermore a remarkable increase in udeskole dissemination in comparison to 14 % in 2007 (67).

A somehow similar OE concept called *Journeys outside the classroom* evolved in Scotland. It was argued that the common residential OE programmes might be too much pre-defined, too much instructor- or teacher-led and too far away from children's and adolescent's real world environments. Additionally, the provided residential experience comes with rather high financial costs to deliver educational contents. Journeys outside the classroom comprise cross-curricular activities that involve the local school environment and that enable real-world encounters on a regular basis (54, 68). In contrast to the udeskole/uteskole concepts, Simon Beames, the author of Journeys outside the classroom, is an experienced outdoor educator, but also a scientific researcher in field.
A comprehensive overview regarding specific teaching concepts from different countries in relation to EOtC is not available in English. However, Dettweiler & Mygind (69) recently conducted a survey with researchers and practitioners from 19 countries regarding EOtC practise, provision and policy. Taking the concept of udeskole as a starting point, the answers are summarised and discussed in Danish language. The authors concluded that EOtC could, besides other important academic and pedagogic aims, serve to tackle global challenges such as climate change and nature conservation. However, more international collaboration between stakeholders from both research and practise is certainly necessary for this purpose (69).
Several academic studies, especially from Scandinavia and the UK, suggested that EOtC could be beneficial to students PA, social behaviour, academic achievement and mental health (29, 54, 70-74). Research studies conducted in Germany are very limited and especially focus on EOtC in primary school, which is foremost explained by the fact that EOtC is not widely disseminated in Germany. The published articles 2 (75) and 3 (76) embedded in this dissertation thesis (see chapter 4) add further empirical value to the body of literature and article 1 (77) aimed to provide a comprehensive literature overview regarding EOtC.

2.3.3 Relevance of a Systematic Literature Review and Sound Methodological Quality in Outdoor Education Original Research Studies

Several literature reviews have been conducted in the field of OE since the early 1990th (41, 42, 47, 78-81). The authors summarized and discussed the benefits of a great variety of OE programmes. However, a systematic literature review focusing on EOtC has not been conducted until 2016. This circumstance has been acknowledged in this dissertation thesis and article 1 (77) provides a systematic overview regarding the international research on EOtC (see chapters 3.1 and 4.1 for detailed descriptions of the conducted work).

In their review article, Scrutton and Beames (47) analysed quantitative studies in the field of Outdoor Adventure Education concerning personal and social development. They pointed out a strong need for good-quality research. Especially while interpreting the results, one must carefully consider samples sizes, used questionnaires, appropriate study designs and certain different variables. Otherwise, research results from outdoor related fields will remain vulnerable. Additionally, Bentsen and colleagues mentioned limitations regarding existing EOtC studies: mainly case studies were conducted, a limited number of students and teachers were involved and a potential publication bias exists. It was pointed out that there is "need for an increased number of studies to increase understanding of different programmes, people, places and processes and make more secure conclusions about impacts and outcomes" (29 p. 35). Based on these experiences and first results, the Danish researchers took advantage of the wide dissemination of EOtC and conducted the extensive research project TEACHOUT. It involved approximately 800 students in a longitudinal quasi-experimental design and investigated their PA, social relations, academic learning and attitude to school (82; see chapter 5 General Discussion for further details). Furthermore, this circumstance has been acknowledged in this dissertation thesis by the application of sound methodological approaches in article 2 and 3.

2.4 Theoretical Foundation

The underlying concepts of EOtC are related to different theories and models: the Attention Restoration Theory (ART; 83, 84, 85)¸ the Self-Determination Theory (SDT;86, 87, 88), and the Biopsychosocial Model of health (BPS-M; 89, 90, 91).

ART describes the relationship between humans' ability to pay attention to stimuli in different environments and the corresponding consequences. "ART is based on past research showing the separation of attention into two components: involuntary attention, where attention is captured by inherently intriguing or important stimuli, and voluntary or directed attention, where attention is directed by cognitive-control processes." (85 p. 1207). Natural green or blue environments are associated with stimuli that rather employ humans' involuntary attention, whereas urban build environments are associated with stimuli that rather employ humans' directed attention. Directed attention is a resource that can be depleted and according to the ART, being in nature can be beneficial to restore the directed attention capacity and overcome mental fatigue. To be able to pay directed attention is an important factor to successfully solve demanding cognitive tasks. In their recently published systematic review, Stevenson et al. (92) appraised the scientific evidence that is based on ART. The authors concluded that especially participants working memory, cognitive functioning and attentional control were improved after being in nature. However, only a very limited number of studies included children or adolescents.

SDT describes the development of intrinsic motivation as a very important success factor in educational settings. When students are largely motivated to follow the curricular contents because of their inherent interest in learning these contends and because of experienced enjoyment and fun, it is very likely that long lasting knowledge acquisition is established and students become lifelong learners (93, 94). An important role for the development of students' intrinsic motivation can be attributed to the SDT-related satisfaction of the basic psychological needs (BPN), namely autonomy, competence and relatedness (95). If the learning environment created by the teacher supports students' autonomy, if they experience competence during the learning process and if they are involved in positive social relations to both classmates and teachers, it is very likely that students develop intrinsic motivation (96). Intrinsic motivation has been identified as a main predictor of students' academic achievement (97). Furthermore, literature suggest that BPN satisfaction is positively associated with students' health-related quality of life and well-being (98-102), self-esteem (99, 103) and self-control skills (101).

Engel argued that the established biomedical model of medicine (104) is too narrow and that human health cannot be solely explained by and reduced to physiological measurements and parameters (89). Engel described in the BPS-M the interaction between biological, psychological and social determinants that influence human health or illness and proposed the importance of the Body-Soul-Unit. All three parameters are to some degree important and a specific health status or a recovery process of a patient cannot be explained in insolation by just one parameter. For example, the recovery process of a child patient relies on biological factors, such as genes, on psychological factors, such as empathy of the physicians and nurses, and on social factors, such as the family environment. As health promotion is an inherent aim in the educational school system (105, 106), a holistic approach to reach that aim is necessary and it cannot solely be transferred to single subjects as PE with its anticipated beneficial physiological effects of PA.

EOtC teachers' aim to facilitate student-centred, hands-on, and experimental learning situations and to promote students PA. They strongly focus child-led approaches with their teaching concepts: phenomena in natural green and blue environments and cultural places are observed and investigated by the children. In comparison to regular indoor teaching concepts, EOtC enables more opportunities for PA, as students are not forced to sit for long periods and often reach the different out-of-school places by active commuting (29).

Based on the discussed theoretical assumptions, EOtC can be seen as a promising holistic framework to satisfy students BPN, promote their learning motivation and PA, and contribute to students' academic performance and well-being.

2.5 Aims and Outline of the Dissertation

In consideration of the state of research and the theoretical framework, the following research desiderata were identified as relevant and will therefore be addressed by the two studies conducted in this dissertation thesis.

Empirical research, especially from Denmark and UK, indicates that EOtC can be beneficial for students. However, many studies are characterised as case studies and the international literature has not been systematically summarized and critically appraised. It is therefore difficult to provide reliable recommendations to which extent and in which cultural settings EOtC can promote students development.

Study 1 aims to close this research gap by systematically summarising and appraising the international literature and providing answers to the question: Can EOtC be beneficial for participating students aged 5–18 with respect to their social and academic development, their PA and their and mental well-being?

Study 2 is based on the theoretical foundation and the identified and appraised empirical research. The aforementioned theoretical and empirical research indicated that being exposed to the natural world—no matter if the subjects are being rather physically inactive or active—could have several positive effects for human health and cognition. Study 2 therefore aims to provide new knowledge regarding students' PA levels, their stress response and the potential relationship between both aspects during EOtC, which takes place in a natural environment. These are aspects which provide important implications for students overall development, however have been researched only in few studies so far.

3 Study Design and Methods

Two studies have been conducted in this dissertation thesis to extent the research regarding EOtC. In study 1, a systematic literature research has been conducted, as systematic literature reviews and meta-analyses have become very important to summarize and appraise the current state of research in a specific research field. The results of systematic literature review and meta-analysis can serve as justifications and a starting point for planned studies, inform scholars about research gaps and the quality and reliability of existing research (107). However, especially in the field of education the informative value of a systematic literature review should be acknowledged. For instance, high quality research designs like randomized controlled trials, which are often prerequisites in systematic literature reviews, are rather difficult to be implemented in everyday school practise (108). Further details regarding study 1 are described in the following subchapter 3.1. In study 2, a longitudinal quasi-experimental study design was applied to collect empirical data based on a newly implemented EOtC school trial. The chosen study design allowed following the same students over the course of one school year with multiple measurement points. It therefore partly accounts for influential factors such as season and personal adaption. Details regarding the key methods and variables are described in the following subchapters. Further details regarding study 2 are described in the following subchapters 3.2. Finally, further details regarding the published results of study 1 are provided in article 1 (77) in chapter 4.1; further details regarding the published results of study 2 are provided in article 2 (75) in chapter 4.2 and in article 3 (76) in chapter 4.3.

3.1 Study 1: Systematic Literature Review

Study 1 is characterized as a systematic literature review and aimed to identify studies about regular compulsory school- and curriculum-based OE programmes and to summarize and discuss the specific outcomes on student level. According to the preferred reporting items for systematic review and meta-analysis (PRISMA) guidelines (109, 110), authors of systematic literature reviews should strictly apply recommended guidelines and report their strategy transparently. We conducted a systematic literature review according to the PRISMA guidelines

(110) to systematically analyse the current state of research regarding EOtC. The electronic bibliographic databases PubMed, Green File, PsycARTICLES, Education Source, ERIC, SPORTDiscus, SocINDEX and Scopus have been screened for studies reporting on *Outdoor Education* research projects, published in peer-reviewed journals. The review has been pre-registered and the protocol can be obtained under the Prospero Reference Number CRD42016033002 (111). All titles, abstracts and full text of studies identified for eligibility were screened independently by two authors (Becker, Lauterbach) according to the previously defined criteria. Studies not meeting the eligibility criteria were excluded. Any disagreement between reviewers concerning the eligibility of particular studies have been resolved through discussion. For each included study, data from full text articles were extracted. The methodological quality of the included studies was assessed by the use of different quality appraisal assessment tools. The reported outcomes were categorized regarding the categories *learning dimensions*, *social dimensions* and *additional outcomes*. A narrative synthesis has been conducted because of great variety of measured and reported outcomes.

3.2 Study 2: Longitudinal Quasi-Experimental Study Design

Study 2 is part of a research project that aimed to analyse student's stress response and stress resilience, PA, learning motivation and social interaction during EOtC. The focus of study 2 was to evaluate the associations between students stress response – measured by cortisol and circulating cell-free deoxyribonucleic acid (cfDNA) – and their SB, LPA and MVPA – measured by 3-axis accelerometry.

The EOtC teaching concept was implemented by two teachers at a private grammar school in Heidelberg, Germany. Three 5th grade classes were enrolled in the EOtC concept on one compulsory school day per week and served as the intervention group (IG). The EOtC concept involved the subjects Biology, Geography, Nature Phenomenon's and PE. The teaching setting was a nearby forest. The data collection was conducted in the school year 2014-2015. The data collection comprised one school day in three seasons (fall/spring/summer). Students from a regular 5th and a regular 6th grade class served as a control group (CG).

3.2.1 Measurement of Sedentary Behaviour and Physical Activity via Triaxial Accelerometry

Students' SB and PA was determined using triaxial Axivity AX3 acceleration sensors (Axivity Ltd., Newcastle upon Tyne, UK). Accelerometry is a widely applied and accepted method to differentiate between human PA levels. It does not rely on participants self-reports and is therefore classified as an objective measure. Several different sensors and software systems to measure and analyse sleep, SB and PA are commercially available. These different systems do not measure human behaviour in the exact same way and different sensor models from the same manufacturer are not comparable per se. Therefore, one has to carefully consider the most suitable system for the specific research with respect to target group and study aim. Furthermore, the mentioned comparability issues need to be considered while comparing results of different research studies. The applied AX3 sensors have been used in several research studies, including large-scale studies (112) and validation studies (113, 114). One sensor was attached to the students' lower back above the upper point of the posterior iliac crest, with the aid of a medical tape (115). The sensors were initialized at 100 Hertz and ±8G bandwidth. The raw vector magnitude acceleration data was converted into the ActiLife file format by an in-house software developed by the University of Southern Denmark (116). Cut-off points reported by Romanzini et al. (117) were used to distinguish between SB, LPA, and MVPA; children's PA levels were analysed using ActiLife v.6.11.4 (ActiGraph, Pensacola, United States of America, USA).

3.2.2 Concept of Stress and Measurement of Students' Stress Response via Cortisol and CfDNA

An individual can have several positively, as well as negatively, connoted physiological and psychological responses to stimuli. The concept of "stress" and its potential positive and negative association with health has been critically discussed in recent literature (118). The authors concluded that uncontrollability or unpredictability of stimuli are necessary conditions to speak of "stress". Many stimuli do challenge a person, however, with the given abilities and resources he or she can often handle the situation, which leads to an adaptive process and personal (physical and or psychological) growth. Such situations and their underlying physiological and psychological mechanisms are not to be considered as "stress". In comparison, uncontrollable or unpredictable situations can often be observed in school: examinations, testimonials, increased mental loads or prolonged social pressure (4, 18). The analysis of cortisol levels is a well-established and accepted but also critically discussed (119) indicator for stress response. The level of cortisol depends—besides the potential stimulus of interest—on several internal and external circumstances, e.g. functionality in the hypothalamic pituitary adrenocortical axis, time of the day, age, gender, nutrition, PA or medication. Under normal conditions, the physiological cortisol secretion follows a relatively stable diurnal rhythm with high levels in the morning and a steady decline until the evening (120). Prolonged interruptions or deviations from a normal diurnal rhythm are associated with mental and physical health problems (121). Therefore, the analysis of student's diurnal cortisol rhythm can be a promising approach to gain information on their acute stress levels. The cfDNA as a promising diagnostic biomarker has gained much scientific attention, especially in cancer (122) and exercise (123) research. The cfDNA is highly sensitive to physical exercise as a stressor and especially increased in participants at a moderate PA below the level of the aerobic–anaerobic transition (124, 125). Therefore, cfDNA can be seen as promising although not yet fully established biomarker for stress response. Both samples for saliva cortisol and saliva cfDNA were collected using Salivette® /Cortisol-Salivette® collection tubes (Sarstedt, Nümbrecht, Germany). As described before, cortisol levels can be affected by participants' nutrition. Therefore, all students were advised not to eat 15 minutes prior to saliva collection. Salivary samples for cortisol quantification were collected by research assistants from the Department of Clinical Psychology, Central Institute of Mental Health Mannheim, University of Heidelberg. Salivary samples for cfDNA quantification were collected by the author of this thesis. Salivary cortisol quantification was performed at the Biopsychology Laboratory, Technical University Dresden, and further processed at the Central Institute of Mental Health Mannheim. Salivary cfDNA quantification was performed at the Department of Sports Medicine, Johannes Gutenberg University of Mainz.

3.2.3 Calculation of Indices: Peak Reactivity and Area Under the Curve

To account for the time- and metabolism-dependent change in endocrinological parameters, many different summary indices are used in the literature (126, 127). One of the most often applied approach is the computation of the area under the curve (AUC), which e.g. allows estimating the circadian change of measured hor-

mones in repeated measurement designs. However, the exact calculation, validity, reliability and meaning of certain indices remained often unclear in the literature, which complicates the comparability of study designs and results (126). Therefore, Khoury et al. (128) identified and compared 15 different summary indices that were often reported in the literature, to provide information regarding interrelations and meanings. The authors concluded that the practical application of 15 indices is not required and that it furthermore complicates the comparison of research results. The area under the curve with respect to ground (AUCg), the area under the curve with respect to increase (AUCi), both special forms of the AUC, and the peak reactivity (PR) were proposed to be suitable indices to account for multiple measurements per day. The AUCg is recommended if the overall secretion of an endocrinological parameter is relevant, the AUCi and the PR are recommended if the change over time is the main aspect (128). The saliva samples for cortisol and cfDNA measurements were taken at the time points 08:30 AM, 10:30 AM, and 12:30 PM. The primarily research interest was the cortisol/cfDNA change over time from 08:30 AM over 10:30 AM to 12:30 PM. Therefore, the AUCi and PR indices were computed instead of the AUCg, which especially takes the overall secretion of cortisol/cfDNA via the absolute baseline value into account. This is especially relevant, as the individual cortisol baseline values of every participant have a greater impact on the respective cortisol change over time when calculated by the AUCg in comparison to the AUCi.
The PR index was calculated using the difference between cortisol/cfDNA measurements at 10:30 AM compared with 8:30 AM, and 12:30 PM compared with 10:30 AM. The same information was used for calculating SB/LPA/MVPA time-intervals. The AUCi index was calculated based on the formulas published by Pruessner et al. (126).

3.3 Statistical Analysis

To account for both the study design and the complex relationships between the applied variables, different statistical approaches were applied to conduct the data analysis in article 2 and 3. In article 2, linear mixed-effects models (LMM) (see subchapter 3.1) were fitted to evaluate if students' showed different patterns of cortisol secretion depending whether they were enrolled in the IG or the CG and depending on the season. MVPA served as a control variable to account for potential influence of PA on the cortisol levels (129). After the publication of article 2, an updated analysis strategy has been developed to better explain the potential underlying reasons for the found differences in students' cortisol response. Based on further literature research, we concluded that isolating MVPA from SB and LPA in regression analysis can lead to potential misleading results and that SB, LPA and MVPA should be treated as a composition. The Compositional Data Analysis (CoDA) approach, besides different discussed alternatives, has been identified to be a possible solution to solve this methodological problem (see chapter 3.3.2). In comparison to the well-established biological stress marker cortisol, the analysis of cfDNA gained more and more attention as a prospective biological stress marker (see chapter 3.2.2). Additionally, Bayesian inference bears in comparison to the more common frequentist inference several advantages (see chapter 3.3.2). Considering these assumptions, we conducted article 3 to account for and take advantage of these recent developments with the aim to better understand the associations between students stress response and their levels of SB and PA with respect to different teaching settings. In the following subchapters, the statistical approaches applied in article 2 and 3 are described in detail.

3.3.1 Linear Mixed-Effects Models

According to the study design described in this chapter (see chapter 3.2. for the overall design and chapter 3.2.3 for the performed calculations), the data is characterised by repeated measurements for every individual in the IG and the CG. LMMs are an appropriate statistical approach to account for complex repeated measurement designs that include different treatment groups and hierarchical structures of variables (130, 131). In comparison to general linear models, such as analysis of variance (ANOVA) (132), LMMs enables that independent variables serve as a combination of fixed and random effects. Random intercepts account for the variation between group means whereas random slopes account for the variation within group responses. It is recommended to use both random intercepts and random slopes to fit models whenever possible (133, 134). That is because more flexibility in the model is given to fit the data if random slopes are chosen and random slope models have been shown to be superior to reduce the risk of both type I and type II errors in comparison to random intercept only models. The estimation of separate slopes for every treatment group requires a relatively large sample size to enable model convergence (130, 134, 135). In the case of study 2, the data showed great inter-individual differences of the cortisol trajectories, which would furthermore indicate the fitting of a random intercept and random slope model. However, due to the rather small sample size and an unbalanced distribution of students between groups, random intercept only models were fitted and showed good model fit to the data.

3.3.2 Compositional Data Analysis

The relevance to apply a suitable statistical approach and to consider possible spurious correlations when analysing data sets representing components that sum up to 100% has first been described by Pearson (136) in 1896. Aitchison (137) described statistical techniques to handle these issues and CoDA is widely used in scientific fields where compositional data naturally occurs, e.g. compositions of sediments in geological sciences or compositions of genomes in microbiology. Variables of a composition are perfectly co-dependent. Applying standard regression techniques to compositional variables lead to multicollinearity issues. One solution would be to ignore the least relevant and least correlated variable. That is the predominant procedure in various scientific fields that analyse the relationships between time-depended human behaviour (sleep, SB and PA) and health, as e.g. LPA and MVPA are most often analysed independent of SB. However, not to account for one or more parts of a composition leads to loss in information and therefore questionable results. Recent studies discussed the great importance of correct data handling when dealing with variables that have an inherent compositional nature to provide reliable evidence on human health (138-143). During a complete day of 24 hours, human behaviour that has been assed via accelerometry is commonly classified in time spent during e.g. sleep, SB, LPA and MVPA. Each increase or reduction in one behaviour leads necessarily to an increase or reduction in at least one of the other behaviours. School days represent a finite amount of time, as a subcategory of a complete day of 24 hours. For obvious reasons, times of sleep are not included in the analysis of students' behaviour during school days. The amount of students SB, LPA and MVPA can furthermore be regarded as absolute values in hours and minutes or as relative proportions that sum up to 100 % of the certain school day.
Standard methods in CoDA are data transformations, e.g. isometric log-ratio (ilr) or centred log-ration (clr) transformation (137). These transformations aim to account for the relative distribution between the variables, e.g. the relative distribution of time that one has spent in different behaviours. Ilr- and clr transformations are recommended in most recent literature (e.g. 138, 140, 144). However, alternative approaches such as Multivariate Pattern Analysis (143, 145), Isotemporal Substitution Model (139, 146) or Bayesian Dirichlet Distribution (147, 148) are

available. All three approaches account to a certain degree for the compositional nature of accelerometry-derived data on SB and PA. Specifically, Multivariate Pattern Analysis enables a comparably high-resolution analysis by providing more detailed PA intensity profiles in comparison to the common used SB, LPA and MVPA. This is especially important for studies analysing PA in relation to cardio-metabolic health related biomarkers. Isotemporal Substitution Models enable the estimation of the effect of replacing one specific behaviour with another behaviour for the exact same amount of time, which is also relevant in cardio-metabolic health related research. The Bayesian Dirichlet Distribution approach was the first to account for multicollinearity between compositional dependent variables, which is very important from e.g. sport science perspectives. The use of ilr-transformations that are commonly used in CoDA were lately criticised. Mekary & Ding (149) argued that the results gained by ilr-transformed SB and PA can be difficult to interpret in relation to PA guidelines as not the absolute amounts for SB, LPA and MVPA in minutes per day but the relative amounts in percentages are reported. Aadland et al. (150) strongly recommended the use of raw instead of transformed SB and PA data. In their analysis, the ilr-transformation led to a distorted correlational structure within PA variables, which in turn distorted the associations between the PA variables in relation to certain indices of cardio-metabolic health.

In our first attempts to account for the compositional nature, we applied an ilr-transformed composition of students SB, LPA and MVPA to explore the respective associations with their cortisol and cfDNA trajectories. However, accordingly to Aadland et al. (150), we also observed a somehow distorted correlational structure in the posterior distribution provided by Bayesian hierarchical linear models (BHLM) using Markov chain Monte Carlo (MCMC) methods. Therefore, we chose to apply a Bayesian ridge regression technique, which is fit to handle the multicollinearity with raw compositional SB and PA data.

In the following subchapter, the advantages of ridge regression techniques are described in a first step. In a second step, further details in the basic principles of Bayesian inference and the applied methods of BHLM are explained.

3.3.3 Ridge Regression, Bayesian Inference and Bayesian Hierarchical Linear Models

Ridge regressions are an alternative to avoid distorted distributions introduced by transformations of raw compositional data. Hoerl and Kennard (151) have introduced the ridge regression technique and proposed that the ridge regression estimator or shrinkage parameter *k* is more appropriate to handle the problem of multicollinearity in comparison to e.g. ordinary least square methods. Ridge regression has since been applied in many empirical as well as simulation studies in research fields where multicollinearity might occur and therefore be problematic (see 152 for a comprehensive review of methods and models, 153). Congdon (154) described different possibilities to account for multicollinearity in the Bayesian framework: (I) to introduce extra information; (II) to reduce the set of covariates; or (III) to apply ridge regression techniques. Only ridge regression seems feasible with respect to the circumstances of study 2, as strictly all information within students SB and PA are of particular interest. Prior defined restrictions on the parameters (I) or e.g. the use of principal component analysis (II) would probably introduce further unwanted bias (see chapter 3.3.2). Bayesian ridge regression already finds first applications in empirical research to handle the multicollinearity between explanatory variables (155). A Bayesian ridge regression approach for the application in R suggested by Parnell (156) has been applied in article 3 (76).

The Bayesian inference is grounded on the Bayes rule, which has first been communicated by Thomas Bayes in 1763 (157). Bayes rule is a mathematical theorem of the probability theory that describes the calculation of conditional probabilities. In short, Bayes stated that there is a relationship between the conditional probability of two events P (A | B) and the inverse form P (B | A) (158). In Bayesian inference, prior (subjective) beliefs or existing evidence regarding a certain phenomenon can be applied to inform the a priori probability and therefore shapes the posterior beliefs/ the posterior probability. That means, ones degree of believe for a certain probability can change depending on the availability of related new evidence. In contrast, in frequentist inference the probability is defined as the frequency of random events in a long run experiment. It assumes infinite repetitions of the experiment are performed, which is practically far away from any real world scientific experiment or real world data collection. A common practise in frequentist inference is the so-called null-hypothesis-significance testing (NHST): by rejecting the null hypothesis, it is assumed that the alternative hypothesis must be “true”. Bayesian inference allows for a more direct view on the posterior distribution in comparison to NHST, as the probability of the alternative hypothesis is not indirectly gained via rejection of the null hypothesis and the data informs the probability of the hypothesis, not vice versa (159). Bayesian inference with its “subjective probability” is furthermore in contrast to the frequentist inference with its “objective probability”. The postulated “objectivity” in frequentist inference has been critically discussed in the literature (160-162). Especially the opportunity within Bayesian inference to inform an analysis with prior knowledge—based on scientific evidence and/or "beliefs"—is more and more recognised and since MCMC analysis with its possibility to draw many random samples from the posterior distribution became much more practical due to computational power, Bayesian inference gained attention in the scientific community (163-165). Bayesian inference is furthermore not restricted to arbitrarily defined cut-off values such as p-values < 0.05 that indicate statistical significance. The usefulness to rely on and report p-values was critically discussed in recent literature (159, 166, 167). This is especially important in our explorative pilot study 2: with the newly introduced teaching setting, the rather small sample size and the uncertainties within measured indices (see chapter 3.2), we aimed to gather first insights and inform future research studies rather than provide generalizable results.

To analyse the associations between students' cortisol/cfDNA response in relation to SB and PA in different teaching setting, different BHLMs (168) were fitted. BHLMs share many similarities, and therefore the same advantages, with the described LMMs in comparison to e.g. ANOVAs, when applied in complex hierarchical designs. Furthermore, BHLMs make use of the described advantages within the Bayesian framework. That means, that e.g. prior information regarding students' cortisol trajectories were used to inform the analysis. Furthermore, the parameters of the posterior distribution were estimated by MCMC algorithms. Based on the given distribution of the collected data, MCMC performs a simulation to generate many thousand random values, from which random samples are drawn to estimate the posterior distributions. To test whether the simulated data matches the collected data, the MCMCs were checked for convergence. The MCMC approach is an especially powerful approach in small sample size studies (169). A practical guidance to set up well-designed statistical approaches in outdoor studies has recently been published (170). A special focus is set on small samples sizes—which is often unavoidable in OE—, hierarchical models, and advantages of Bayesian interference in comparison to frequentist inference.

4 Publications

This dissertation thesis contains three articles that are published in international peer-reviewed journals. Article 1 is a theoretical work in which the existing literature regarding EOtC is systemically summarised and appraised and belongs to the research project described in chapter 3.1 Study 1: Systematic Literature Review. Article 2 and 3 are empirical works and belong to the research project described in chapter 3.2 Study 2: Longitudinal quasi-experimental study design. The three articles are published under Open Access conditions and are freely available. The author of this dissertation thesis has co-authored additional peer-reviewed articles that are published under Open Access conditions. All published articled are listed in chapter 8 List of Publications.

4.1 Article 1

Title: Effects of Regular Classes in Outdoor Education Settings: A Systematic Review on Students' Learning, Social and Health Dimensions

Authors: Christoph Becker, Gabriele Lauterbach, Sarah Spengler, Ulrich Dettweiler, Filip Mess

Reference: Becker, C., Lauterbach, G., Spengler, S., Dettweiler, U., & Mess, F. (2017). Effects of Regular Classes in Outdoor Education Settings: A Systematic Review on Students' Learning, Social and Health Dimensions. International Journal Environmental Research and Public Health, 14(5). doi:10.3390/ijerph14050485

Contributions: Christoph Becker was the main investigator for this article. Christoph Becker, Sarah Spengler and Filip Mess conceived and designed the study. Christoph Becker and Gabriele Lauterbach performed the systematic literature search and selection process. Christoph Becker, Gabriele Lauterbach, Ulrich Dettweiler and Filip Mess performed the methodological quality assessment. Christoph Becker analysed the data and wrote most of the manuscript. All authors contributed to and approved the final manuscript.

Summary

The aim of this article was to systematically identify and summarize published and peer-reviewed research studies that evaluated EOtC practise. We categorised and evaluated the reported outcomes, assessed the methodological quality, and discussed the proposed benefits for participating students. The online databases PubMed, Scopus, Education Source, ERIC, Green File, PsycARTICLES, SPORTDiscus and SocINDEX were searched and the PRISMA guidelines to conduct systematic literature reviews were applied. The methodological quality of included articles were assessed by different tools. Thirteen studies were included in the analysis. Many studies used a case-study design and the methodological quality was on average moderate to low. Tendencies were found that EOtC could be beneficial for students in terms of social, academic, physical and psychological dimensions. The fewest results were reported concerning students' PA and mental health.

4.2 Article 2

Title: Stress in School. Some Empirical Hints on the Circadian Cortisol Rhythm of Children in Outdoor and Indoor Classes

Authors: Ulrich Dettweiler, Christoph Becker, Bjørn H. Auestad, Peter Kirsch, Perikles Simon

Reference: Dettweiler, U., Becker, C., Auestad, B. H., Simon, P., & Kirsch, P. (2017). Stress in School. Some Empirical Hints on the Circadian Cortisol Rhythm of Children in Outdoor and Indoor Classes. International Journal of Environmental Research and Public Health, 14(5), 475. doi:10.3390/ijerph14050475

Contributions: Ulrich Dettweiler was the main investigator in this article. Peter Kirsch, Perikles Simon and Ulrich Dettweiler received the funding grant and designed the study. Christoph Becker collected the data. Ulrich Dettweiler, Peter Kirsch, Perikles Simon and Bjørn H. Auestad analysed the data. All authors contributed to and approved the final manuscript.

Summary

In an explorative longitudinal quasi-experimental study, students within EOtC classes were compared to students in normal classes in terms of their stress levels measured via cortisol and their MVPA levels measured via accelerometry. Overall, 48 children were enrolled in a private grammar school in Heidelberg, Germany: 37 5th grade students in the IG; 11 5th and 6th grade students in the CG. The EOtC setting contained one full school day per week and the subjects Biology, Geography, Nature Phenomenon's and PE. The EOtC classes took place in a nearby forest. LMM were fitted to analyse group differences in the cortisol level; MVPA and season and were taken into account. The IG showed a greater decline of cortisol compared to CG. MVPA was not statistically significant associated to cortisol; however, MVPA levels were higher in the IG compared to the CG. The IG showed a steady decline of cortisol during EOtC. That is in accordance with a normal healthy diurnal cortisol rhythm. No such effect was found for the CG.

4.3 Article 3

Title: Children's Cortisol and Cell-Free DNA Trajectories in Relation to Sedentary Behavior and Physical Activity in School: A Pilot Study

Authors: Christoph Becker, Sebastian Schmidt, Elmo W. I. Neuberger, Peter Kirsch, Perikles Simon, Ulrich Dettweiler

Reference: Becker, C., Schmidt, S., Neuberger, E. W. I., Kirsch, P., Simon, P., & Dettweiler, U. (2019). Children's Cortisol and Cell-Free DNA Trajectories in Relation to Sedentary Behavior and Physical Activity in School: A Pilot Study. Frontiers in Public Health, 7(26). doi:10.3389/fpubh.2019.00026

Contributions: Christoph Becker was the main investigator in this article. Peter Kirsch, Perikles Simon and Ulrich Dettweiler received the funding grant and designed the study. Christoph Becker and Ulrich Dettweiler analysed the data. Christoph Becker collected the data and wrote most of the manuscript. All authors contributed to and approved the final manuscript.

Summary

In article 2, we were not able to fully explain the significant differences in student's cortisol levels between CG and IG. It seemed to be independent of their MVPA levels. However, objectively measured SB, LPA and MVPA are collinear and therefore share a common relation to the cortisol response. Furthermore, literature showed that cfDNA is a promising stress biomarker, whereas with a potentially different response to SB and PA in comparison to cortisol. Therefore, based on the results from article 2, this article aimed to evaluate the association between students' cortisol, cfDNA levels, and their SB, LPA MVPA. We fitted BHLMs to evaluate the association between cortisol and cfDNA, and the composition of SB, LPA, and MVPA. For the IG, the shown decline of cortisol was associated with relatively high levels of LPA and a high amount of cfDNA was positively associated relatively high levels of SB. The associations for all other variables and for the CG were less strong. LPA seems to promote students' healthy cortisol decrease during EOtC. The specific driving factor of the teaching environment is also important, but the underlying mechanisms are less clear and need further investigations.

5 General Discussion

The overall aim of this dissertation thesis was to close a research gap by systematically summarising and appraising the international literature on EOtC from a students' perspective and to provide answers, if EOtC can be beneficial for students with respect to their social and academic development, their PA and their mental well-being. Furthermore, this dissertation thesis aimed to provide new empirical results to enlarge the anticipated rather small body of literature that focused on students' PA and stress response.

Study 1 can be considered as the first conducted systematic literature review that strictly and transparently assessed, summarised and critically appraised the existing literature in the field of EOtC. Hence, a more comprehensive picture of the published peer-reviewed results was provided. It can be of use for researchers to identify specific gaps in the literature and to promote self-reflection towards applied scientific standards in study design and analysis. It can furthermore guide teachers and related practitioners to identify potential benefits of EOtC on student level and to develop own teaching concepts focusing out-of-classroom environments, even though the body of appraised literature with 13 identified articles is rather small and the methodological shortcomings have to be considered. The conducted systematic literature review revealed specific gaps within the published results on EOtC. From the 13 identified articles, eight reported results regarding students' social dimensions and seven set focus on learning outcomes. Students' PA and health were only evaluated by three articles. Furthermore, the demographic data of the publications showed that only one study was conducted in Germany and the majority have been conducted in Scandinavia and the USA. These circumstances emphasize a great demand for more studies evaluating student's PA and health during EOtC as well as more studies need to be conducted in Germany. During the same school year 2014/2015 in which the data collection of study 2 took place; a research group from Denmark conducted a large-scale quasi-experimental EOtC study called TEACHOUT. Several articles were subsequently published. However, due to the publication dates they were not eligible for inclusion in the systematic literature review. The respective results and implications will be discussed below at the end of this chapter.

Study 2 can be considered as the first empirical research that has been conducted with a focus on EOtC involving 5th and 6th grade students in Germany. It was furthermore conducted in a longitudinal quasi-experimental design and sound methodological approaches have been applied. Hence, the methodological shortcomings in existing literature identified in study 1 have to a certain degree been explicitly accounted for in the articles 2 and 3. Specifically, as article 3 builds upon article 2, the recently made available approaches of CoDA have been applied for a better understanding of the associations between students SB, PA and their stress response. Several authors discussed the necessity to account for the co-dependent structure of objectively measured SB and PA during a finite amount of time, especially from a health perspective (138, 139, 143). Article 2 concluded that the decrease of students' cortisol during EOtC lessons was independent of their respective level of MVPA. The reason for the decrease therefore remained somehow unclear. By taking the compositional nature of SB, LPA and MVPA during a finite amount of time into account, LPA seems to be positively associated with the cortisol decrease. As the amounts of LPA were similar in both groups, but no decrease of cortisol was observed in the CG, the characteristics of the outdoor environment and the teaching concepts seem to be relevant concerning students stress response and need further investigation. As EOtC is often practised in natural green or blue environments, sunlight exposure can be an important factor to be considered in future research. Literature reports that light exposure has an influence on human cortisol levels; however, the direction of association is controversially discussed and strongly depends on the light intensity, duration of exposure and the timing in relation to the diurnal cortisol rhythm (171, 172). In relation to PA in nature, the growing body of literature of GE indicates potentially advanced health benefits of being physically active in the presence of nature in comparison to similar PA indoors (34, 173). However, research results for psychosocial and physiological health outcomes in children and adolescents are scarce and inconclusive. Further research is needed for a better understanding of the potential inherent benefits of being physically active in the presence of nature for that age group (35).

The associations between the measured cfDNA and students SB and PA were less clear. As cfDNA and cortisol show different time- and exercise-related metabolisms, these biomarkers cannot be directly compared. The explorative study 2 has been conducted in a real world situation. Nevertheless, it provides first insights regarding a potential relationship between physiological induced cfDNA levels, SB and PA in children. Further laboratory-based research studies that already exists for adults, are needed to better understand the role of cfDNA as biological stress marker for children (125). Future research studies should closely investigate the underlying mechanisms regarding students stress response during school time, as these have a great relevance from a psychological health perspective (174).

As outlined above, certain articles from the TEACHOUT study from Denmark were published lately. They represent a more robust body of literature regarding EOtC, as data collection was performed nation-wide, EOtC-classes were compared to control groups and strong methodological approaches were used (82). The TEACHOUT studies indicate that EOtC can lead to increased PA among boys (175, 176), improved learning motivation (177), improved social behaviour (178), improved well-being (179) and better reading performance (180); no differences were reported for learning outcomes in mathematics (181). Bølling et al. (178) observed a negative correlation between EOtC and hyperactivity-inattention and peer problems for students with a comparatively low socioeconomic status. Additionally, a recent study based on the '*Development of Udeskole'* project initiated by the Danish Ministry of Education and the Danish Ministry of Environment suggests that breaks and rest periods during EOtC-lessons in close proximity to natural green or blue environments seem to have a positive effect on students stress reduction (182). That is in concordance with the results of two recently published systematic reviews that summarised the research results regarding the impact of children's interaction with nature on their mental, physical and social health (183, 184). Very little is still known about the academic performance of students attending EOtC in comparison to regular class control groups. Therefore, future EOtC-studies are encouraged to set focus on students' academic achievement in robust experimental designs.

These discussed results furthermore strengthen the body of literature on EOtC, especially as in part larger sample sizes were included and more robust methodological approaches were used in comparison to the articles identified in study 1. The results are in concordance with the presented results from study 1 and 2 and demonstrate valuable arguments to promote a further dissemination of EOtC.

5.1 Limitations and Future Research Perspectives

As mentioned before, many promising studies have been published since the publication of article 1. Therefore, an update and extended version of the systematic literature review could be useful within the next years. Most research on EOtC focuses on specific outcomes on student level but only some articles examine the perspectives of involved teachers (e.g. 185, 186, 187). However, very little is known about how the teaching practice in the out-of-classroom environments (185) and if specific curricula for EOtC exist and to what extent they are applied

in different countries. A comprehensive analysis of the EOtC teaching practise and curricula would certainly be helpful for a better understanding why EOtC can be beneficial to promote students' development. It can furthermore help to differentiate between suitable and rather unsuitable contents for EOtC lessons. Lloyd B. Sharp, one of the first OE pioneers already stated in 1943 "that which ought and can best be taught inside the schoolrooms should there be taught, and that which can best be learned through experience dealing directly with native materials and life situations outside the school should there be learned" (188 p. 363).
Although a strictly systematic and transparent approach has been applied in study 1, one cannot claim that an 'objective' truth regarding EOtC research was enabled. A systematic literature review is inherently biased by the subjectivity of the involved researchers and furthermore potentially valuable research articles could have been missed (108).
Study 2 must explicitly be interpreted as a strictly exploratory pilot study, because the rather small sample size and the non-randomization of students to the groups do not allow a generalization. Furthermore, questionnaire-based information regarding students' stress and mental well-being were not assessed and the collected hair cortisol probes were not fit to be analysed. Both measures would have enabled crosschecks on the cortisol data and insights regarding students' long-term cortisol response. We collected saliva probes at 8:30 AM, 10:30 AM and 12:30 PM. A finer graduation of the measuring points, that also involved measuring points immediately after the awakening, could have delivered a clearer picture regarding the individual cortisol trajectories. These showed a great variance, with e.g. some students in the IG having the exact opposite cortisol response compared to the group mean: an increase from morning to noon. Furthermore, the observed seasonal differences remained unclear based on the collected data. These might somehow be related to different levels of UV-light, or just are simple artefacts.
The applied approach and the gained results provide valuable information for future study designs and analyses. Specifically, informed priors in BHLMs could be applied in future research studies with similar designs. A greater awareness of the compositional nature of human behaviours such as sleep, SB and PA is necessary. Future research studies in the field of EOtC must carefully consider applying robust methodologies to provide reliable evidence. Only thereby, the gained research results can contribute to inform educational policy and public.

The dissemination of EOtC has only been systematically assessed in Denmark (66, 67), but not in Germany or other German speaking countries. It can be assumed that several schools, most notably primary schools, practise different forms of EOtC in Germany. To conduct robust large-scale studies that account for specific circumstances of national or regional educational systems, the dissemination of EOtC in Germany and other German speaking countries should be assessed. A first step towards a better understanding about EOtC in German speaking countries could be the development of a network between practitioners, educators and researchers following the example of the Danish udeskole network UdeskoleNET (189).

5.2 Conclusion

This dissertation thesis provides sound scientific research in the still small, but evolving field of OE, especially regarding effects of EOtC. The systematic literature review in study 1 identified potential benefits when students receive education outside the classroom. However, only a very limited number of peer-reviewed articles were identified, and many of them applied rather poor or moderate methodologies. The empirical studies 2 and 3 identified potential benefits of EOtC for students' PA levels and stress response. Furthermore, the relationship between students' SB, PA and stress response has been evaluated with a sound methodological approach. The gained results are useful to inform practitioners and therefore foster the dissemination of EOtC practice as well as researchers to strengthen the methodological approaches. Future research studies in the greater field of OE must certainly apply methodological approaches of high quality to provide valuable and reliable results that can have an impact on educational policy and public. Furthermore, more research is needed that also examines teacher perspectives and specific EOtC curricula to better understand why and how teachers make use of the EOtC approach.

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8 List of Publications

Becker, C., Lauterbach, G., Spengler, S., Dettweiler, U., & Mess, F. (2017). Effects of Regular Classes in Outdoor Education Settings: A Systematic Review on Students' Learning, Social and Health Dimensions. International Journal of Environmental Research and Public Health, 14(5) 485. doi:10.3390/ijerph14050485

Dettweiler, U., Becker, C., Auestad, B. H., Simon, P., & Kirsch, P. (2017). Stress in School. Some Empirical Hints on the Circadian Cortisol Rhythm of Children in Outdoor and Indoor Classes. International Journal of Environmental Research and Public Health, 14(5), 475. doi:10.3390/ijerph14050475

Becker, C., Schmidt, S., Neuberger, E. W. I., Kirsch, P., Simon, P., & Dettweiler, U. (2019). Children's Cortisol and Cell-Free DNA Trajectories in Relation to Sedentary Behavior and Physical Activity in School: A Pilot Study. Frontiers in Public Health, 7(26). doi:10.3389/fpubh.2019.00026

Dettweiler, U., Lauterbach, G., Becker, C., Ünlü, A., & Gschrey, B. (2015). Investigating the motivational behaviour of pupils during outdoor science teaching within self-determination theory. Frontiers in Psychology, 6(125). doi:10.3389/fpsyg.2015.00125

Dettweiler, U., Lauterbach, G., Becker, C., & Simon, P. (2017). A Bayesian Mixed-Methods Analysis of Basic Psychological Needs Satisfaction through Outdoor Learning and Its Influence on Motivational Behavior in Science Class. Frontiers in Psychology, 8, 2235. doi:10.3389/fpsyg.2017.02235

Jaitner, D., Rinas, R., Becker, C., Niermann, C., Breithecker, J., & Mess, F. (2019). Supporting Subject Justification by Educational Psychology: A Systematic Review of Achievement Goal Motivation in School Physical Education. Frontiers in Education, 4(70). doi:10.3389/feduc.2019.00070

9 Appendix

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International Journal of
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MDPI

Review

Effects of Regular Classes in Outdoor Education Settings: A Systematic Review on Students' Learning, Social and Health Dimensions

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Abstract: Background: Participants in Outdoor Education Programmes (OEPs) presumably benefit from these programmes in terms of their social and personal development, academic achievement and physical activity (PA). The aim of this systematic review was to identify studies about regular compulsory school- and curriculum-based OEPs, to categorise and evaluate reported outcomes, to assess the methodological quality, and to discuss possible benefits for students. Methods: We searched online databases to identify English- and German-language peer-reviewed journal articles that reported any outcomes on a student level. Two independent reviewers screened studies identified for eligibility and assessed the methodological quality. Results: Thirteen studies were included for analysis. Most studies used a case-study design, the average number of participants was moderate (mean valued (M) = 62.17; standard deviation (SD) = 64.12), and the methodological quality was moderate on average for qualitative studies (M = 0.52; SD = 0.11), and low on average for quantitative studies (M = 0.18; SD = 0.42). Eight studies described outcomes in terms of social dimensions, seven studies in learning dimensions and four studies were subsumed under additional outcomes, i.e., PA and health. Eleven studies reported positive, one study positive as well as negative, and one study reported negative effects. PA and mental health as outcomes were underrepresented. Conclusion: Tendencies were detected that regular compulsory school- and curriculum-based OEPs can promote students in respect of social, academic, physical and psychological dimensions. Very little is known concerning students' PA or mental health. We recommend conducting more quasi-experimental design and longitudinal studies with a greater number of participants, and a high methodological quality to further investigate these tendencies.

Keywords: outdoor education; school; children; adolescents; curriculum; social; learning; health; review

1. Introduction

Within the past 20 years, Outdoor Education Programmes (OEPs) in general have been reported to show a number of positive effects on personal and social development, physical activity, academic achievement and leadership skills for a wide range of participants and age groups [1–3].

With a more specific focus on education within the school context, regular compulsory school-based and curriculum-based outdoor education programmes seem to have several positive effects on students' physical activity levels [4], mental health status [5], social competences and relations [6,7], and academic achievement [8].

An all-encompassing definition of outdoor education is scarcely possible due to different meanings, understandings and practices within various research areas, countries and cultures [9]. Common terms include: learning outside the classroom, udeskole, friluftsliv, outdoor adventure education and forest school. In general, outdoor education can be described as teaching and/or learning and/or experiencing in an outdoor and/or out-of-school environment. The content of learning and teaching is therefore different and depends on the general aim of the programme, the target group and the outdoor setting, e.g., the gaining of knowledge in natural sciences; increased PA (physical activity), leadership skills, personal and social development; survival skills; and improved skills in relation to nature-oriented sports.

In contrast to these more general outdoor education programmes, we have concentrated on programmes that are embedded within the curriculum and are conducted regularly within the school schedule. These programmes focus on student-centred classes and interdisciplinary subjects, hands-on learning, possibilities to explore and experience oneself and the environment, and the use of natural and cultural places as a "classroom" [10,11]. Regular school-based and curriculum-based outdoor education programmes are still a rare phenomenon—with the exception of the grassroots movement of udeskole/uteskole in Scandinavia [12] which has increased during the last decade. It shows that, for example, 17.9% of all public schools and 19.4% of all private schools in Denmark participate in regular outdoor teaching [13]. However, research results regarding those programmes are often only based on case studies using an arsenal of different methodological approaches.

Nevertheless, recent educational school reforms can be observed in several countries. The Danish reform "Improving the Public School" explicitly aims to increase PA during the school day; a longer school day with a special focus on learning, motivation and well-being; and working more closely with local sports clubs and cultural centres [14]. Recommendations to teach several curriculum content areas outside the classroom can be found in the new regional curriculum in Bavaria, Germany [15]. Furthermore, a shift towards multi-disciplinary, phenomenon- and project-based teaching was projected within the "National Core Curriculum 2016" in Finland [16,17]. Well-structured and curriculum-integrated outdoor education programmes could therefore offer great opportunities in helping to achieve the above-mentioned objectives.

In the last decades, six important reviews and meta-analyses in the field of outdoor education have been published [1–3,18–20]. Rickinson et al. [2], for example, set a wide focus on outdoor learning by evaluating the impact of: (i) fieldwork and visits; (ii) outdoor adventure activities; and (iii) school grounds and community projects. The authors summarised diverse benefits for each category, e.g., an increase in PA and academic achievement, development of social skills and a favourable attitude towards the environment. The recent systematic review of Fiennes et al. [20] partially updated the work of Rickinson et al. [2] by analysing primary research studies on outdoor learning from the UK that have been published since 2003. Similar to the conclusion of Rickinson et al. [2], most of the studies showed positive effects on a wide range of outcomes. The main study topics were still adventurous and residential activities while only a few studies were strongly linked to core curriculum subjects.

Only one review took a close look at the context of regular outdoor education within the school curriculum. Waite, Bølling and Bentsen [1] compared studies on Danish udeskole and English forest schools with a focus on purposes, aims, pedagogy, content, outcomes and barriers. The authors especially highlighted that both concepts seem to support children in their social and academic achievement, as well as their physiological and psychological well-being.

The existing reviews and meta-analysis in the wider field of outdoor education give a valuable overview on outdoor education research and practise. However, the literature shows a wide range in the intervention length, target and age groups, programme approaches, and the methodologies used. Three publications analysed programmes in the context of Outdoor Adventure Education/Outdoor Adventure Programming [3,18,19]. Two reviews set a very wide [2,20], and one review a narrow [1], focus on different OEPs within the school context. In addition, in most of the reviews the included primary studies are limited to selected countries. Only one review [20] used a systematic approach with respect to approved guidelines, i.e., the Reporting of Primary Empirical Research Studies in Education (REPOSE) Guidelines [21], and two reviews were not published in peer-reviewed journals [2,20].

Our purpose was to summarise studies on regular compulsory school- and curriculum-based outdoor education programmes for participants aged 5–18 that had been published in peer-reviewed journals. We aimed at: (i) categorising and evaluating reported outcomes; (ii) assessing the methodological quality of the included studies; and (iii) discussing possible benefits on students' development by such programmes.

2. Methods

To identify and analyse the existing literature on regular compulsory school- and curriculum-based outdoor education programmes, we chose to endorse a systematic review approach. Systematic reviews in the context of education were, however, criticised by several authors [22–24]. It is concluded that one has be aware of the respective possibilities as well as limitations a systematic review can offer. Therefore, we see our work in relation to the model of education research developed by Andrews [22]. According to this model, we tried to summarise what is published and what methodological approaches were used, to identify the gaps and methodological shortcomings in the reviewed studies [22]. We conducted the systematic review in accordance with the preferred reporting items for systematic review and meta-analysis (PRISMA) guidelines [22]. The PRISMA guidelines are a well-accepted tool for systematic reviews and meta-analyses, they provide a valuable overview on how to structure the research process and help authors to account for transparency, validity and reproducibility.

2.1. Search Strategy

On 8 April 2016, we searched through the electronic PubMed, Scopus, Education Source, ERIC, Green File, PsycARTICLES, SPORTDiscus and SocINDEX databases for English and German language peer-reviewed journal articles. The search string included two components: "objective" and "setting". Whereas "objective" represented relevant terms in respect of the synonyms for outdoor education programmes, "setting" described the defined educational environment. We used the following search terms for "objective" and "setting":

Objective: "outdoor education", "outdoor learning", "outdoor teaching", "learning outside the classroom", "out-of-classroom", "experiential learning", "expeditionary learning", "udeskole", "uteskole", "friluftsliv", "forest school", "nature school", "environmental education", "place-based education", "Draußenschule", and "Draussenschule".

Setting: "school" and "curriculum".

We used Boolean search operators, parentheses, search fields and asterisk according to the database specifications. Furthermore, we screened reference lists and citations of included articles to identify additional relevant studies.

For a detailed protocol and search strategy, please refer to our registered and published protocol under the International Prospective Register of Systematic Reviews (PROSPERO) Number: CRD42016033002. These documents are also available under Supplementary Materials.

2.2. Eligibility Criteria

We only included studies meeting the following eligibility criteria:

- All types of study designs (e.g., control group design, quasi-experimental design, and case studies);
- Any type of formal school- and curriculum-based outdoor education programme involving children and adolescents (5–18 years);

- Regular weekly or bi-weekly classes in a natural or cultural environment outside the classroom with at least four hours of compulsory educational activities per week over a period of at least two months; and
- At least one reported outcome on a student level.

No restrictions on publication periods were given.

2.3. Selection Process

Two independent reviewers (CB and GL) gradually screened all the titles and abstracts of studies identified for eligibility according to the criteria. Based on given information within the titles and abstracts, we made decisions about inclusion or exclusion. For studies that looked as if they would fulfil the inclusion criteria, we screened the full texts. If insufficient information was given in the abstract in order to make a clear exclusion decision, the full text was also screened. Any disagreements between reviewers were resolved by discussion. Both reviewers carefully documented their results after each step. We contacted the corresponding authors of 30 studies and requested additional information about the intervention and analyses procedures.

Both reviewers screened the reference lists and citations of included studies listed in Scopus using the same procedure to identify additional relevant studies.

2.4. Data Extraction

For each included study, we extracted data using a piloting form in respect to the required items. When essential information was not available from the full texts, we asked the corresponding authors to provide more information. Extracted data included:

1. Study characteristics: Citation, author, date of publication, journal, study-design, and country;
2. Population: Age, gender, sample size, and type of school;
3. Intervention characteristics: intervention and data acquisition period, and amount of intervention;
4. Methodology and analytic process.
5. Reported outcomes and main results.
6. Barriers and limitations.
7. Information for assessment of the risk of bias; and
8. Source(s) of research/project funding and potential conflicts of interest.

2.5. Analysis and Synthesis

Options for statistical quantitative analyses, including, risk ratios and standardised mean differences, were limited due to the heterogeneity of study designs, the range of measured outcomes and the overall small number of included studies. We therefore firstly provide a flow chart on the search and selection process and three tables presenting the main descriptive characteristics as well as the reported main outcomes of the included studies. Secondly, we qualitatively describe the most important outcomes of the studies in a narrative synthesis. Thirdly, we present results of the methodological quality assessment of included studies both in tables and narrative text.

2.6. Methodological Quality Assessment

Two reviewers (CB and GL) assessed the methodological quality of included studies. Additionally, one more independent reviewer (FM) had to specifically evaluate one article [23] which had been included in the review, due to the authorship of GL and UD who are part of the review team. Any disagreements between the reviewers were resolved through discussion and by referring to a third reviewer (UD). The quality of quantitative studies was appraised using the Child Care and Early Education Research Connections (CCEERC) Quantitative Research Assessment Tool [24]. The quality of qualitative studies was appraised using the Joanna Briggs Institute (JBI) Checklist for Qualitative

Research [25]. Both tools were used for studies using quantitative, as well as qualitative, methods. For each tool, an overall rating was conducted based on the given assessment criteria. Quantitative studies were rated on 12 questions using a scale: 1, 0, −1, and n/a (not applicable); to account for completeness one question on research ethics was adapted by the JBI Checklist for Qualitative Research. Qualitative studies were rated on 9 questions using a scale: y (yes), n (no), u (unclear), and n/a (not applicable). One item was excluded due to inappropriateness within the research field. For further analyses, we adjusted the qualitative scale similar to the quantitative scale to the level of 1 (y), 0 (u), −1 (n), and n/a. For both quantitative and qualitative studies, an overall rating is presented in Appendix A Tables A1 and A2 with mean values and standard deviations. Based on the mean values, we provide an overall rating regarding the categories low, moderate and high methodological quality. The cut-off values are defined as follows: low = $M < 0.30$; moderate = $0.30 \leq M \leq 0.60$; and high = $M > 0.60$. They are based on theoretical assumptions in relation to methodological quality. Our approach, including the cut-off values based on the mean values, should be seen as a relative rating in relation to our data to provide a comparison of methodological quality. To our knowledge, no other rating system is available in relation to the applied tools. No studies were excluded from the review based on their methodological quality assessment results to ensure that all the potential valuable results are presented [26].

3. Results

Figure 1 shows the selection process in general, numbers for each stage of the selection process and reasons for exclusion after screening the full papers. After the exclusion of direct duplicates, the literature search in the various databases yielded 7830 potentially relevant publications. After we screened titles and abstracts, we retrieved 193 studies in full-text. Thirteen studies met all the eligibility criteria. We looked at reference lists and citations of included studies listed in Scopus. Both the reference list search and the cited-by-search yielded no additional studies that met all the eligibility criteria. Finally, we included 13 studies in this systematic review.

3.1. Characteristics of Included Studies

Table 1 shows the main descriptive characteristics of the 13 included studies. Table 2 shows specific information concerning the interventions and data collections. Four studies were conducted in Denmark [4,6,7,27], three in the USA [28–30], and one each in Germany [23], New Zealand [31], Sweden [5], the UK [32], and Norway [33]. One study included data from the UK, India and Kenya [34]. The sample sizes varied considerably across the studies, from five [6] to 230 [5] children/adolescents being involved. Nine studies are defined as case studies [4,6,7,27,29,31–34], three studies used a quasi-experimental design [5,28,30], and one study used a cross-sectional design [23]. Three publications [4,6,7] are based on the same intervention, while all other publications are based on individual interventions. Six studies collected and analysed data on a solely student level [4,23,27,29,34,35], five studies also included data from teachers, staff and parents [6,30–33] and one study [5] only included data from parents. Eight studies used interviews [6,27,29–34], six studies used questionnaires [5,7,28,30,31], three studies used learning assessments [29–31], two studies used observations [32,34] and, in each case, one study used a postal survey [23], written documents [29], drawings and concept maps [34], and accelerometry [4]. The quantity of compulsory educational activities in a natural or cultural environment outside the classroom varied from one school day bi-weekly to a duration of eight weeks [27], and a six-month full week programme [23]. The chosen environments also differ between the studies: gardening projects on school grounds or nearby community properties [27,32,34], classes in a local forest [4,6,7,31,33], prairie [30] and farmland areas [29], the use of nearby school environments [5,28], and an overseas sailing expedition [23].

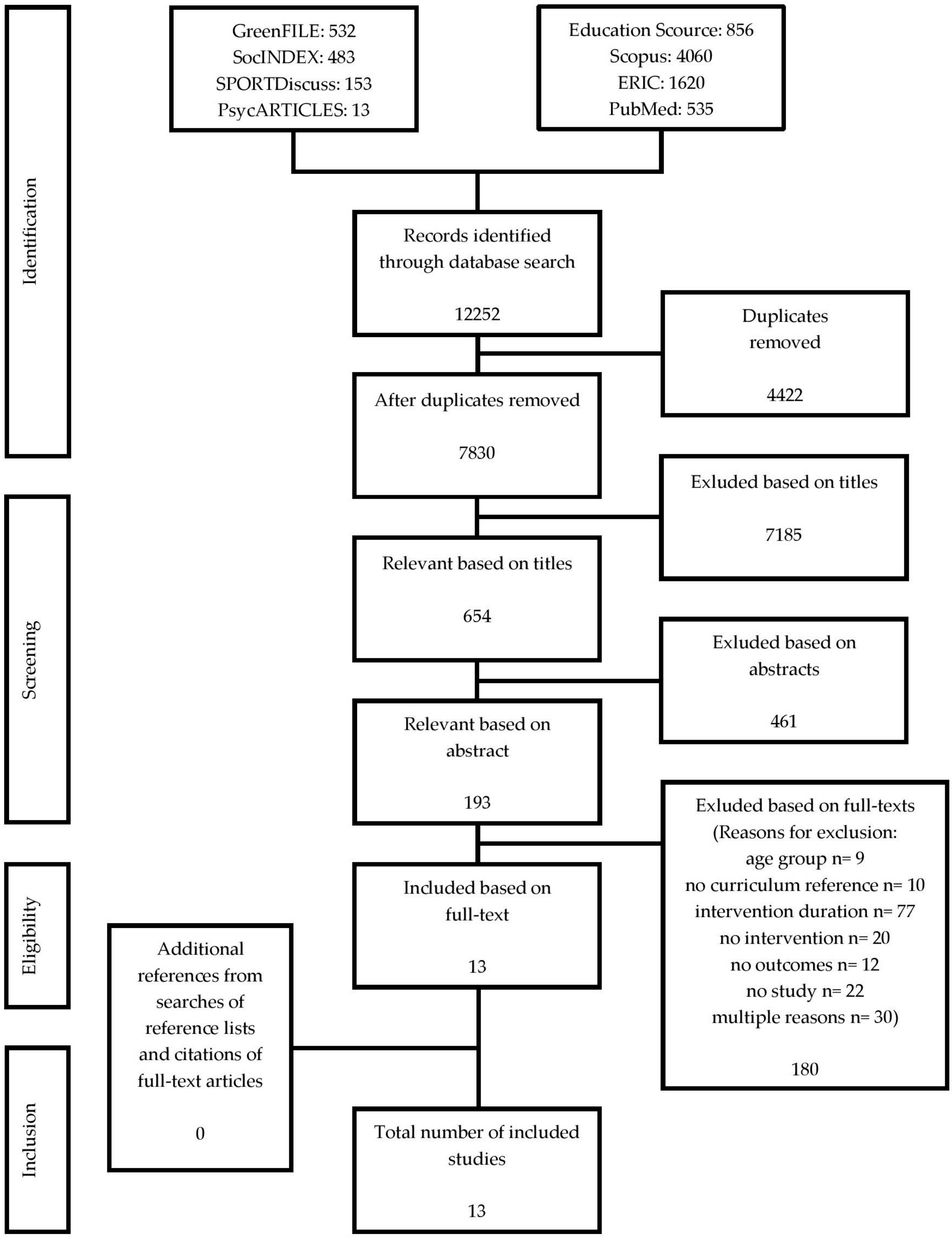

Figure 1. Flow chart of study search and selection process.

Table 1. Descriptive characteristics of studies on regular school- and curriculum-based outdoor education programmes.

Source	N	Age	Distribution of Sex (% Male)	Country	Study Design	Administrator of Data Acquisition	Type of School
Mygind [4]	19	9–10	26.3	Denmark	case-study	chn	primary school
Mygind [7]	19	9–10	26.3	Denmark	case-study	chn	primary school
Dettweiler et al. [23]	56	14–20	n/a	Germany	cross-sectional retrospective	adol	secondary school
Hartmeyer et al. [6]	5 ^{adol}, 2 ^{t}	16	40 ^{adol}	Denmark	case-study retrospective	adol, t	primary school
Martin et al. [28]	45 ^{IG}, 67 ^{CG}	14–15	51.1 ^{IG}, 47.8 ^{CG}	USA	quasi-experimental	adol	secondary school
Santelmann et al. [29]	40	12–15	n/a	USA	case-study	chn, adol	secondary school
Moeed et al. [31]	85 ^{adol}, 1 ^{t}	15-24	61 ^{adol}	New Zealand	case-study	adol, adul, t	secondary school
Gustafsson et al. [5]	121 ^{IG}, 109 ^{CG}	8.6 ± 1.6 ^{IG}, 8.1 ± 1.5 ^{CG}	56.2 ^{IG}, 51.4 ^{CG}	Sweden	quasi-experimental	chn	primary school
Bowker et al. [34]	72	7–14	n/a	UK, India, Kenya	case-study	chn, adol	primary + secondary school
Sharpe [32]	9 ^{chn}, 2 ^{t}, 5 ^{p}, 2 ^{s}	10–11	n/a	UK	case-study	chn, t, p, s	primary school
Fiskum et al. [33]	9	10–11	55.6	Norway	case-study	chn	primary school
Wistoft [27]	98 $^{chn/t}$, 135 ^{p}, 6 ^{s}	-	n/a	Denmark	case-study	chn, p, t, s	primary school
Ernst et al. [30]	90 ^{chn}, n/a $^{p\ s}$	10–11 ^{chn}	n/a	USA	quasi-experimental	chn, p, s	secondary school

Note: adol: adolescents; chn: children; p: parents; t: teacher; s: staff; IG: intervention group; CG: control group; n/a: not available.

Table 2. Characteristics of intervention and data collection of studies on regular school- and curriculum-based outdoor education programmes.

Source	Data Collection	Intervention Period and Data Acquisition	Intervention Length
Mygind [4]	objectively-measured physical activity; accelerometry devise: CSA 7164 activity monitor	IP: school years 2000/2001/2002 DA: school years 2000/2001/2002	three school years; one outdoor school day each week
Mygind [7]	adapted version of "About my self—a questionnaire for children" on self-perceived physical activity level, social relations and learning behaviour	IP: school years 2000/2001/2002/2003 DA: school years 2000/2001/2002/2003	three school years; one outdoor school day each week
Dettweiler et al. [23]	postal survey; hand written letter	IP: 2008/2009/2010/2011 DA: 2012	six months; each expedition
Hartmeyer et al. [6]	semi-structured interviews	IP: school years 2000/2001/2002/2003 DA: 2010	three school years; one outdoor school day each week
Martin et al. [28]	Children's Environmental Virtue Scale (CEVS) Questionnaire, adapted and modified by Children's Environmental Attitude and Knowledge Scale (CHEAKS)	IP: 10/2005-01/2006 DA: 10/2005+01/2006 [IG]; spring semester 2006 [CG]	10 weeks; at least one half day per week
Santelmann et al. [29]	interviews, written documents, learning assessment	IP: school year 2006/2007 DA: 2006/2007	one school year; one outdoor school day in 1/3 of all weeks
Moeed et al. [31]	unspecified self-evaluation questionnaire, interviews, learning assessment	IP: 1997–1998 DA: 1997–1998	two school years; four hours bi-weekly year nine; four hours weekly year 10
Gustafsson et al. [5]	Strengths and Difficulties Questionnaire (SDQ), parent-version	IP: school year 2002/2003 DA: autumn 2002/autumn 2003	one school year; five days per week; at least one hour per day
Bowker et al. [34]	concept maps, semi-structured group interviews, contextual observations, drawings	IP: school year 2004/2005 DA: school year 2004/2005	one school year; four hours on average each week
Sharpe [32]	semi-structured individual interviews, group interview, observations	IP: school year 2012/2013 DA: summer/autumn 2013	one school year; four hours on average each week
Fiskum et al. [33]	group interviews	IP: school years 2004–2008 DA: autumn 2008/spring 2009	five school years; one outdoor day per week, years 1–4, one outdoor school day bi-weekly, year five
Wistoft [27]	group interviews, individual interviews, unspecified questionnaires	IP: school year 2010/2011 DA: school year 2010/2011	eight weeks; one outdoor school day bi-weekly; 7–8 h on average per day
Ernst et al. [30]	Skills Self-Report questionnaire; Affective Self-Report and Parent Survey questionnaire, both developed by the author; standardised assessment test: Minnesota Comprehensive Assessments in Maths and Writing; individual interviews	IP: school year 2003/2004 DA: school year 2003/2004	one school year; five days per week; two hours per day

Note: IG: intervention group; CG: control group; IP: intervention period; DA: data acquisition.

The included studies are very heterogeneous in respect of their study design, used methods and instruments, learning environments and measured outcomes. We categorised measured outcomes and presented the results of each study according to the study design. Seven studies reported outcomes on learning dimensions [7,27,29–32,34] and eight studies on social dimensions [6,7,23,27,28,30,31,34]. Two studies reported on students' physical activity [4,7], one study [5] on students' mental health, one study [33] on students' action regulation behaviour and one study [31] on students' environmental attitude and behaviour.

3.2. Methodological Quality Assessment of Included Studies

The methodological quality for most of the quantitative studies [4,7,28,30,31] can be classified as low (M = −0.14, SD = 0.31) to moderate (M = 0.50; SD = 0), with mean values ranged from −0.45 to 0.45; one study was rated as high, with a mean value of 0.67 (M = 0.67; SD = 0) [5]. Main reasons for the low or moderate ratings result from a poor description of the population of interest, the non-random selection of participants, insufficient presentation of means and standard variations/standard errors for numeric variables, the handling of missing data, the inappropriateness of statistical techniques and handling of alternative explanations, insufficient information according to current ethical criteria, and missing model coefficients and standard errors for main effect variables.

The methodological quality for most of the qualitative studies [6,27,29–33] can be classified as moderate (M = 0.41, SD = 0.12), with mean values ranged from 0.33 to 0.56. Two studies were rated as high (M = 0.78, SD = 0) [23,34], with mean values of 0.78 each. Main reasons for the low ratings result from insufficient information about the influence of the researcher on the observed or interviewed participants, and vice-versa; insufficient information according to current ethical criteria; and an inappropriate connection between the conclusions and the analyses.

A detailed description of the methodological quality assessment is presented in Appendix A Tables A1 and A2.

3.3. Categorised Outcomes

We categorised the reported outcomes of studies on regular school and curriculum OEP. Table 3 shows the main outcomes in order to categorise students' learning dimensions, social dimensions and additional outcomes.

3.3.1. Outcomes on Learning Dimensions

Six case studies [7,27,29,31,32,34] analysed datasets concerning learning dimension. Mygind [4] conducted a study with primary school children attending a three-year outdoor education project. Students were asked about their perceptions on teaching and learning during indoor and outdoor classes by means of a questionnaire. Significant differences were found in three out of 14 statements: students liked the outdoor setting more than the indoor setting ($p < 0.05$), they were more careless about homework in the indoor setting ($p < 0.01$) and more disturbances in group work activities occurred during the indoor setting ($p < 0.05$). No significant differences were found for the other 11 statements.

Santelmann et al. [29] conducted a study with sixth- to eighth-grade students participating in a one-year place-based curriculum. The authors analysed documents written by students, and interviews conducted by students using a semi-quantitative content analysis. It is concluded that, through direct interaction with landowners, students developed a better understanding of decision-making in farm and forest enterprises, and received insights into the global interconnectedness of agricultural markets. The students' learning benefit during outdoor lessons was especially mediated through hands-on learning and active participation. In a self-evaluation learning assessment, 75% of the students reported having gained new knowledge about farms, forests and wildlife refuges, and 25% developed better communication skills towards adults.

Table 3. Reported outcomes of studies on regular school- and curriculum-based outdoor education programmes.

Source	Outcomes on Learning Dimensions	Outcomes on Social Dimensions	Additional Outcomes
Mygind [4]			PA significant higher during outdoor classes compared to indoor classes ($p < 0.001$, 2000/2001); no significant differences in PA between outdoor classes and indoor classes including 2 PE lessons ($p = 0.52$, 2002); significant −level: 0.05
Mygind [7]	higher preferences for learning in the outdoor setting compared to indoor setting; significant differences in three out of 14 statements	significant more positive social relations in the outdoor setting compared to the indoor setting ($p < 0.001$); significance-level: 0.05	significant higher perceived PA in the outdoor setting ($p < 0.01$); significant −level: 0.05
Dettweiler et al. [23]		long-term educational overseas expedition can lead to symptoms of a reverse culture shock; similar readjustment problems and development of coping strategies for all the participants, shown in a U-curve model; the longer the students had time to readjust, the more positive they report on perceived programme effects, shown as a linear function; no differences between cruises and gender	
Hartmeyer et al. [6]		identification of six important conditions for the improvement of social relations: play, interaction, participation and pupil-centred tasks—important for positive social relations during udeskole; co-operation and engagement—consequences of improved social relations in subsequent years	
Martin et al. [28]		IG: significant decrease in 5 CEVS domains: courage ($p < 0.006$); temperance ($p = 0.084$); acceptance ($p = 0.014$); compassion ($p = 0.109$); humility ($p = 0.009$); CG: significant decrease in courage ($p = 0.169$) and increase in temperance ($p = 0.389$); acceptance ($p = 0.553$); compassion ($p = 0.796$); humility ($p = 0.553$); significance-level: 0.1	
Santelmann et al. [29]	improved understanding of decision-making on farm and forest enterprises; insights into the global interconnectedness and ecodynamic drivers of agricultural markets		
Moeed et al. [31]	year 10 students: improved horticulture skills (85% improved grade with 13%); year 9 students: strong level of commitment to develop knowledge and skills		former students: long term effects of the programme concerning positive environmental behaviour: growing own vegetables, participating in community-based planting programmes, taking own students outdoors within environmental projects, cleaning the Himalayas

Table 3. *Cont.*

Source	Outcomes on Learning Dimensions	Outcomes on Social Dimensions	Additional Outcomes
Gustafsson et al. [5]			overall positive, but not significant effect on mental health in the IG ($p > 0.1$); significant decrease in mental health problems for boys in IG compared to CG ($p < 0.001$); no significant differences for girls; significance-level: 0.1
Bowker et al. [34]	gardening experience has a positive impact on curriculum learning: indication of direct association between gardening activities and improved learning	overall sense of pride, excitement and high self-esteem; gardening experience had a positive impact on students' general school experience: indication of direct association between gardening activities and self-esteem	
Sharpe [31]	strong contextualised learning opportunities for children in Maths, English and Science; learning is perceived as fun through imaginative and creative learning opportunities; transfer from the indoor and outdoor classroom to real-life situations	building of trusting relationships and educationally-focused symbiotic relationships; growth in self-confidence; experience to take active responsibility for the environment	
Fiskum et al. [33]			gender differences: boys more often grasped affordances specific to the outdoor environment and used own creativity; girls more often grasped affordances not specific to the outdoor environment and used attached objects especially designed for them; girls more often regulate their action in the outdoor setting
Wistoft [27]	students developed a desire to learn through participation in the programme; they learned through enjoyment and experiences, they perceived learning as fun	students developed social competencies through participation in the programme	
Ernst et al. [30]	significant higher reading + writing scores for IG compared to CG ($p = 0.03$); positive significant increase in science process, problem-solving, technology skills, skills in working and communication for IG compared to CG ($p < 0.01$); students in the IG became more interested in school and learning fostered by outdoor learning	positive significant difference in students' attitudes towards the prairie wetlands environment for IG compared to CG ($p = 0.02$); IG students improved their classroom behaviour and prompted a sense of belonging	

Note: IG: intervention group; CG: control group, PA: physical activity; sig: significant; PE: physical education.

Moeed et al. [31] conducted a study with ninth and tenth-grade students in a school-led and community-supported environmental education project. A subgroup of tenth-grade students was pre- and post-tested on horticulture skills using a learning assessment. Eighty-five per cent of the students improved their grade skills for all four skill sets (preparing seeds for germination, pricking out, transplanting seedlings, planting) by 13%. The students were also able to transfer the gained skills into a different context.

Bowker et al. [34] carried out a study with primary and secondary school children participating in a one-year school gardening project. The authors used a qualitative content analysis to analyse concept maps, drawings, interview transcripts, and contextual observations. Based on these analyses, the authors stated that the gardening experiences can have a positive impact on students' curriculum learning.

Sharpe [32] evaluated how a one-year community gardening programme can be beneficial for fifth-grade students in building confidence and being prepared for academic success. Sharpe used a qualitative content analysis to analyse semi-structured interviews, contextual observations and drawings. It is reported that the students had strong contextualised learning opportunities in mathematics, English and science, which allowed them to apply learned content to real-life situations.

Wistoft [27] carried out a study with primary school children attending a half-year community-led garden project. The author applied a qualitative content analysis to analyse interview transcripts and questionnaires. A summary of students' learning dimensions yielded in three main categories: (i) learning through enjoyment and experiences; (ii) the ability to use knowledge, understanding and the skills acquired; and (iii) learning through the outdoor life. Students' learning opportunities were made possible by the teachers' passion and love for teaching. As a main conclusion, the students developed a desire to learn through participation in the programme, which can be seen as an indicator of positive learning motivation.

Ernst et al. [30] evaluated learning dimensions of a one-year out-of-school science programme for fifth-grade students. The authors used standardised assessment tests to compare students' learning achievements in reading, writing and mathematics, and found significantly higher reading and writing scores for students within the intervention group (IG), compared to students within the control group (CG) ($p = 0.03$). No results were given for scores in mathematics. Based on the self-report questionnaire analysis, a positive and significant increase in the science process, problem-solving, technology skills, skills in working, and communication for students within IG compared to students within CG ($p < 0.01$) was found. Ninety-eight per cent of the parents from students within IG stated in a questionnaire that their children learned science, maths and writing better than they would have done in a normal school setting. Parents mentioned hands-on learning practise, interdisciplinary instructional strategy and real-world applications within outdoor teaching as the main conditions for this positive learning environment. Students within IG stated in interviews that they became more interested in school and learning through the outdoor teaching.

3.3.2. Outcomes on Social Dimensions

Six case studies [6,7,27,30,32,34] analysed datasets concerning social dimensions. Mygind [7] used a questionnaire to ask students about their social relations during teaching and during the breaks, comparing indoor and outdoor classes. Significant differences were found in two out of 10 statements: students liked the outdoor setting more than the indoor setting ($p < 0.05$) and it was noisier during the indoor setting ($p < 0.05$). No significant differences were found for the other eight statements, i.e., "I tease my classmates in the..." or "I try to assist my classmates in the . . . " Hartmeyer et al. [6] conducted a study with students and teachers seven years after the students had participated in a three-year primary school outdoor education project. In a qualitative content analysis, semi-structured interviews with students and teachers were analysed. In conclusion, six conditions influenced students' social relations during their school years. In detail, the students improved their social relations and four conditions seem to have been important for that: "play", "interaction", "participation" and "pupil-centred tasks". Furthermore, this improvement in social relations, enabled through the four conditions, positively influenced the pupils' ability to "co-operate" and to "engage" in subsequent school years. Bowker et al. [34] concluded that students who had taken part in a one-year gardening project developed an overall sense of pride, excitement and high self-esteem. The gardening experience had a positive impact on students' general school experience, which was interpreted as an association between gardening activities and self-esteem. Sharpe [32] reported that students developed trusting relationships and educationally-focused symbiotic relationships during the one-year project. Furthermore, the project fostered students' growth in self-confidence and experiences leading to them taking active responsibility for the environment. Wistoft [27] reported that students developed social competencies through active participation in the gardening project: improved team-working and communication skills, improved social relatedness, and an understanding of the importance of taking responsibility and having respect for others' work and property.

One quasi-experimental study [28] analysed datasets concerning social dimensions.

Martin et al. [28] conducted a study to research the effects of a 10-week expeditionary learning programme on seventh and eighth-grade students' environmental virtue. Students completed questionnaires and the environmental virtue score decreased significantly for students' in intervention group (IG) in four out of five domains of environmental virtue: courage (p = 0.006); temperance (p = 0.084); acceptance (p = 0.014); and humility (p = 0.009). For students in the control group (CG), the changes in environmental virtue score were not significant.

One cross-sectional study [23] analysed datasets on social dimensions. Dettweiler et al. [23] conducted a study with students who had participated in one of overall four six-month overseas learning expeditions. To evaluate the students' social readjustment strategies, they were asked to write letters about their experiences after they returned from the expedition. In a mixed-method approach, the authors analysed students' readjustment strategies. The time intervals between the return and the data collection were different for every expedition. The statements on readjustment strategies from the students being at home for eight months were most negative compared to the students having less or more time to readjust. Therefore, students can experience symptoms of a reverse culture shock after a long-term overseas expedition. However, the longer the students had time to readjust, the more positive they report on perceived programme effects. No gender differences were found.

One quasi-experimental study [30] analysed datasets on social dimensions. Ernst et al. [30] compared students' attitudes towards a specific local natural environment. The attitudes towards the environment of students in the IG changed significantly compared to that of students in the CG (p = 0.02). One hundred per cent of the parents of students in the IG stated in a questionnaire that their children expressed a positive attitude towards outdoor teaching and 98% stated that the outdoor teaching fostered students' excitement about school in general. Students in the IG stated in interviews that their social behaviour had improved and that the outdoor lessons had advanced their social relatedness.

3.3.3. Additional Outcomes

In addition to the aforementioned two categories of Outcomes on Learning and Social Dimensions, we clustered five studies [4,5,7,31,33] with specific outcomes under additional outcomes as they do not fit precisely into any other category.

Two case studies [4,7] analysed datasets on students' physical activity. Mygind [4,7] conducted the studies with students participating in a three-year outdoor education project and measured their PA during outdoor and indoor learning. Objectively-measured PA was significantly higher during one outdoor learning day, compared to one traditional indoor school day, in 2000 and 2001 (both $p < 0.001$), while no significant differences in PA was found for one outdoor learning day, compared to one normal school day including two physical education lessons, in 2002 (p = 0.52) [4]. Students were asked, by means of a questionnaire, about their perceived physical activity. Students reported to have used their body significantly more often during classes in the outdoor teaching setting ($p < 0.01$) compared to the indoor setting and also to have been more active during the breaks in the outdoor setting ($p < 0.01$) compared to the indoor setting [7].

One case study [31] analysed datasets regarding students' environmental attitude and behaviour. Moeed et al. [31] applied a qualitative content analysis to analyse interview transcripts. The authors conducted the interviews six or eight years respectively, after the students participated in an environmental project. The students showed a strong awareness of environmental issues and were actively involved in environmental community projects. The students traced both aspects back to their participation and experiences in outdoor classes.

One case study [33] analysed datasets with respect to students' action-regulation behaviour. Fiskum et al. [33] conducted a study with fifth-grade primary school students who participated in outdoor classes over the period of five years. In a qualitative content analysis group interviews were analysed with a special focus on interaction between affordances, action-regulation, and learning. The authors reported that outdoor learning environments can offer a great variability in children's choices of activity during classes. The main results relate to gender differences. Boys mainly grasped affordances specific to the outdoor environment and used their own creativity, whereas girls mainly grasped affordances not specific to the outdoor environment and used attached objects especially designed for them. Girls more often regulated their action in the outdoor setting compared to boys. Both girls and boys reported on several learning contents related to grasped affordances. It has been concluded that outdoor education compared to teacher-directed learning in the classroom, may provide better opportunities to reach the third level of cognitive process dimension—apply—by enabling conscious relationships concerning content and objects.

One quasi-experimental study [5] analysed datasets on students' mental health status. Gustafsson et al. [5] conducted a study with primary school children who had attended an outdoor education project over a period of one school year. In a questionnaire, the parents stated their observations regarding their children's psychiatric symptoms. When adjusted for demographics, no significant overall effect on mental health was found for students in the IG, compared to students in the CG with respect to total difficulties, as well as all the subscales (all *ps* > 0.1). However, a gender effect of the intervention was found. Mental problems significantly decreased for boys compared to girls, with respect to total difficulties ($p < 0.001$), as well as the subscales of "emotional symptoms" ($p = 0.044$), "conduct problems" ($p < 0.003$), and "hyperactivity" ($p = 0.005$). Effects were not significant for peer problems and pro-social behaviour.

4. Discussion

We aimed at systematically reviewing the current state of research on regular compulsory school- and curriculum-based outdoor education programmes. Specifically, we categorised and evaluated reported outcomes of 13 included studies and rated their methodological quality.

4.1. General Aspects

The current state of research is relatively small with only 13 identified and evaluated studies. This can partly be explained by the fact that outdoor education research is quite a young field of research, although, with a rising number of publications within the last years. The small number of included studies can also be attributed to the fact that efforts to conduct regular curriculum-based outdoor teaching face many barriers. Waite, Bølling and Bentsen [1] summarised the cost of transportation and extra teachers, travel-time, a crowded curriculum and teacher qualifications as main obstacles for more outdoor learning projects in schools in the UK and in Denmark.

We also applied certain inclusion criteria, such as a minimum intervention length of eight weeks. By further opening-up these criteria, more studies could naturally have been evaluated, but this would have simultaneously led to a renunciation of the comparability of the assessed studies and outcomes. Waite, Bølling and Bentsen [1], for example, therefore chose different inclusion criteria—less strict concerning, e.g., age group, intervention duration, publication type—and thus compared

39 similar studies concerning school-based outdoor education programmes. Compared to the related field of Outdoor Adventure Education/Outdoor Adventure Programming, the aforementioned literature reviews and meta-analyses reviewed several studies, e.g., 96 studies regarding the overall effects of adventure programmes [3], and 43 studies concerning outdoor adventure programmes for adolescents [19]. This can also be seen as an indication that more studies on regular compulsory school- and curriculum-based outdoor education programmes are needed, in order to gain a deeper understanding of the possible benefits.

4.2. Methodological Quality Assessment

The methodological quality assessment for most of the studies yielded moderate results.

Particularly, those results of studies with moderate or low methodological quality have to therefore be considered with caution. Apart from that, some important specific circumstances regarding the included studies have to be considered. Due to the nature of educational interventions, not all requirements for preventing possible methodological bias (e.g., randomisation, a high number of participants) can be fulfilled in practice and we applied two relatively strict assessment tools. In contrast to most natural science domains, formal ethical approvals are still not obligatory in some educational and sociological domains. Furthermore, official ethic committees still have to be established to a certain extent. Another explanation could be that researchers are incidentally unaware of the importance of such formal ethical issues. Furthermore, the aim of most (case) studies included in this systematic review was rather to explore the field and to describe specific (rare) cases, instead of giving the opportunity to generalise the results gained to a wider population. As mentioned above, several studies do show a lack of methodological quality. Although the methodological quality of research studies is not the main focus of this review—and one should not overestimate it when considering the possibilities of conducting studies in educational settings—these ratings can be seen as indicators for detecting shortcomings in this particular scientific field, and this is in concordance with results of the review by Scrutton et al. [18]. The authors examined studies in the related field of Outdoor Adventure Education, focusing on personal and social development. They stated that, frequently, the sample sizes used were too small, and went on to discuss the questionable usage and handling of questionnaires, as well as the statistical management of variables. Scrutton and colleagues [18] requested that future research should be carefully designed with regard to methodological rigour if the researchers' aim is to actually inform and change educational policy.

Certain results must therefore be interpreted with respect to the study design used and its corresponding possibilities and weaknesses as regarding generalisability, validity and reliability.

4.3. Learning Dimensions

The presented results in the category of learning dimension, reported by seven studies [7,27,29–32,34], illustrate one main focus of the current research in the field of regular compulsory school- and curriculum-based outdoor education programmes.

According to the results on learning dimensions, students particularly seem to benefit in terms of an improved academic performance in several subjects, improved skills in transferring the knowledge gained to real life situations. In addition, two studies [27,34] mentioned possible benefits on aspects of students' learning motivation, i.e., learning as fun and a desire to learn. Considering that learning motivation can be an important factor for academic success [35], and some studies in outdoor education settings [36–38] have already analysed motivational aspects of short-term interventions, this could possibly be a promising approach for future research.

The methodological quality for studies reporting on learning dimensions, however, is rated as moderate [7,27,29,30,32] except for one study which is rated as low [31]. Due to the methodological weaknesses, the reported results have to be considered with caution. However, they are in concordance with different literature reviews and meta-analyses concerning general outdoor education. Waite, Bølling and Bentsen [1] mentioned that regular udeskole enhances learning outcomes.

Rickinson et al. [2] highlighted the benefits of school grounds/community projects on students' science process skills as well as the impact of fieldwork and visits on students' long-term memory and higher order learning. Furthermore, Cason and Gillis [19] found an average effect size of 0.61 (n = 10; SD = 1.527) of outdoor adventure programmes on adolescents' grades and Hattie et al. [3] mentioned that "adventure programs enhance general problem solving competencies", understood as a subcategory of academic performance (ES = 0.45; n = 23; CI = 0.23 to 0.67).

Taking into account these indications and respective methodological shortcomings, more high quality-studies are needed to further examine possible effects of regular outdoor classes on students learning dimensions.

4.4. Social Dimensions

The presented results in the category of social dimension, reported by nine studies [6,7,23,28,30–32,34], illustrates another main focus of the current research regarding regular compulsory school- and curriculum-based outdoor education programmes.

According to the results in social dimensions, students seem to benefit in terms of their development of social competencies and social relations such as self-esteem, self-confidence, trusting relationships, and the sense of belonging [6,7,27,30,32,34]. One study [23] also reported that students mentioned perceived positive programmes effects, however, with a temporal shift of approximately eight months. Furthermore, three studies reported positive effects on students' attitudes and behaviour patterns towards the environment [30–32]. One study [28] mentioned negative effects on students' environmental attitudes. The methodological quality for studies reporting on social dimension is rated as moderate [6,7,27,30–32] except for two studies rated as high [23,34]. Despite the methodological weaknesses, the reported results are in concordance with conclusions by Waite, Bølling and Bentsen [1]: Forest schools, as well as udeskole programmes, can promote students' social relations, interpersonal skills, and social competencies. Furthermore, Rickinson et al. [2,5] summarised that fieldwork and visits "can lead to individual growth and improvements in social skills (. . .) and improve attitudes towards the environment" while school grounds and community projects can foster students' sense of belonging, relationships and community involvement.

Similar to our demands regarding learning dimensions, there is also a strong need for more high quality-studies to further examine possible effects of regular outdoor classes on students' social dimensions.

4.5. Additional Dimensions

The research on students' physical activity, mental health and action regulation behaviour is underrepresented in comparison to results on students' learning and social dimensions. Only two case studies [4,7] with moderate to low methodological quality, reported positive effects on students' PA. Only one case study [33] with moderate methodological quality mentioned gender differences with respect to action regulation behaviour. Furthermore, only one quasi-experimental study [5], with a high methodological quality reported positive effects of regular outdoor classes on boys' mental health. Therefore, the presented results of PA, mental health and action regulation behaviour can at most be interpreted as first indications. However, taking results from related publications into account, these indications can be partly supported. In detail, Rickinson et al. [2] showed in their review that school grounds and community projects can be beneficial for children's exercise. Additionally, Waite, Bølling and Bentsen [1] mentioned that forest school and udeskole projects increased students' PA and motor-skills. Regarding students' mental health, Cason and Gillis [19] found an average effect size of 1.047 (n = 12; SD = 0.459) for adolescents' clinical scales (e.g., depression and anxiety) regarding outdoor adventure programming.

More high quality-studies are therefore needed to further examine these first indications of the effects of regular outdoor classes on students' PA, mental health and action regulation behaviour, especially when considering an increasing inactivity [39], as well as a rising number of diagnosed mental health disorders in school children [40].

4.6. Strengths and Limitations

There are four main strengths in this systematic review. First, we strictly referred to a search protocol and design according to the PRISMA Guidelines and applied several online databases for literature research. Secondly, the chosen inclusion criteria allowed for the consideration of a wide range of studies concerning study design, country, target group and reported outcomes. Thirdly, two reviewers independently screened the literature and assessed the methodological quality of the included studies and, fourthly, we applied the CCEERC Quantitative Research Assessment Tool as well as the JBI Checklist for Qualitative Research to rate the studies' methodological quality.

However, we only evaluated studies published in English and German in peer-reviewed journals and listed in the used online databases, but no grey literature or reports. We therefore cannot rule out the existence of relevant studies in other languages or studies published elsewhere. Furthermore, we observed that several included, as well as excluded, articles were weak in respect of the internal structure and given information. Hypothesising that this is a wide spread practice, this could also mean that other valuable research results had not been properly published in peer-reviewed journals, and were therefore not eligible for inclusion in this systematic review.

These limitations are in concordance with the critique on systematic reviews in education, as described in the methods chapter. Therefore, we cannot claim to have delivered an all-embracing solution to the questions we have asked. We have not "eliminate(ed) bias" nor have we "present(ed) an 'objective' version of the truth, but" we have "attempt(ed) to minimise bias" in the field [41].

5. Conclusions

To conclude, the number of identified studies on regular compulsory school- and curriculum-based outdoor education programmes is relatively low. In addition, these 13 evaluated studies show wide heterogeneity in respect of the aims, participant groups, learning environments, methods used and reported effects, and the methodological quality is, on average, moderate. However, tendencies were found which indicate that regular compulsory school- and curriculum-based outdoor education programmes can advance students in the physical, psychological, learning and social dimensions.

To further evaluate these indications, more research studies are needed. Thereby, a strong focus on aspects of study design and methodological quality has to be set. Especially randomised-controlled trials, longitudinal studies and studies that are more quasi-experimental with a higher number of participants are desirable for future research. Additionally, the intervention duration should be as long as possible, as it has been shown that longer programmes lead to better effects [2]. Future research should particularly focus on aspects of students' PA and mental health, as we have shown that those are underrepresented in the reviewed literature.

However, these study designs are often difficult to conduct in educational settings, especially as practical "Outdoor Education' strongly depends on the respective teachers" motivation and beliefs, their pedagogical concepts and ideas, and a certain financial support from headmasters/headmistresses and school authorities [1,12]. If practitioners, researchers and policymakers work more closely together in a dialogic relationship and with a strong focus on what is needed, as demanded by Fiennes et al. [20] and Andrews [41], positive changes in school practise can hopefully be realised for students' benefits. This can partly be seen in relationship to a recent OECD report on learning environments in the 21st century. According to the report, innovative learning environments are needed. Specifically, a combination of pedagogical approaches on "guided learning", "action learning" and "experiential learning" that enables self-regulated learning [42]. Although not being the focus in our review, the underlying pedagogical concepts in outdoor education do set focus at least partially on these learning environments [1].

One promising example is the Danish TEACHOUT research project which used a quasi-experimental and longitudinal design to analyse the impacts of regular outdoor teaching on 834 students' PA, well-being, social interaction and learning [43]. First results are to be expected in 2017. In the future, more such high-quality studies should be realised by referring to a rich theoretical background and methodology, as well as informing and including policy and school administration.

Supplementary Materials: The following are available online at www.mdpi.com/1660-4601/14/5/485/s1, PROSPERO Protocol and Search Strategy, Reference Number CRD42016033002.; adapted versions of the CCEERC Quantitative Research Assessment Tool; and the JBI Checklist for Qualitative Research.

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Appendix A

Table A1. Methodological quality assessment for quantitative studies.

Source	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q8	Q10	Q11	Q12	Mean	SD
Mygind [4]	0	0	0	1	1	1	−1	0	−1	1	−1	−1	0.00	0.00
Mygind [27]	0	0	0	1	1	1	0	0	−1	1	−1	−1	0.08	0.79
Martin et al. [29]	0	0	1	1	1	1	0	0	0	1	0	1	0.50	0.52
Moeed et al. [32]	0	0	1	1	−1	−1	−1	−1	−1	−1	−1	−1	−0.50	0.80
Gustafsson et al. [5]	0	0	1	0	1	1	1	0	1	1	1	1	0.66	0.49
Ernst et al. [31]	0	0	1	1	1	1	0	0	0	1	0	−1	0.33	0.65

SD: standard deviation.

Table A2. Methodological quality assessment for qualitative studies.

Source	Q1	Q2	Q3	Q4	Q5	Q7	Q8	Q9	Q10	Mean	SD
Dettweiler et al. [23]	1	1	1	1	1	−1	1	1	1	0.78	0.67
Hartmeyer et al. [6]	1	1	1	1	1	−1	1	1	−1	0.56	0.88
Santelmann et al. [30]	0	0	1	1	1	−1	1	−1	1	0.33	0.86
Moeed et al. [32]	1	1	1	−1	1	−1	1	−1	1	0.33	1
Bowker et al. [35]	1	1	1	1	1	−1	1	1	1	0.78	0.67
Sharpe [33]	1	1	1	1	1	−1	1	0	−1	0.44	0.88
Fiskum et al. [34]	0	0	1	1	1	1	1	−1	−1	0.33	0.87
Wistoft [28]	1	1	1	1	1	−1	1	−1	1	0.56	0.88
Ernst et al. [31]	1	1	1	1	1	−1	1	−1	1	0.56	0.88

SD: standard deviation.

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Article

Stress in School. Some Empirical Hints on the Circadian Cortisol Rhythm of Children in Outdoor and Indoor Classes

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Abstract: This prospective longitudinal survey compared the stress levels of students taught using an outdoor curriculum in a forest, with children in a normal school setting. We were especially interested in the effect outdoor teaching might have on the children's normal diurnal cortisol rhythm. 48 children (mean age = 11.23; standard deviation (SD) = 0.46) were enrolled, with 37 in the intervention group (IG), and 11 in the control group (CG). The intervention consisted of one full school day per week in the forest over the school year. Stress levels were measured in cortisol with three samples of saliva per day. Furthermore, the data allowed for statistical control of physical activity (PA) values. For data analysis, we used a linear mixed-effects model (LMM) with random intercept and general correlation matrix for the within-unit residuals. The LMM yields that IG have expected greater decline of cortisol compared to CG; rate 0.069 μg/L vs. 0.0102 μg/L (log-units/2 h), $p = 0.009$. PA does not show a statistically significant interaction with cortisol ($p = 0.857$), despite being higher in the intervention group ($p < 0.001$). The main effect in our measures was that the IG had a steady decline of cortisol during the school day. This is in accordance with a healthy child's diurnal rhythm, with a significant decline of cortisol from morning to noon. This effect is constant over the school year. The CG does not show this decline during either measurement day. Further research is needed to fully explain this interesting phenomenon.

Keywords: stress; cortisol; physical activity; outdoor learning; mixed effect model

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1. Introduction

Both public debate and epidemiological research show evidence of an increase of stress symptoms and stress-associated diseases over the past decade. This has been identified on an international level [1], but it is also found in Germany specifically [2]. According to a recent statement issued by German health insurance companies [3], 16.2% of all employee sickness-related absences are attributable to mental health disorders, with many of them associated to stress. This is an extreme increase, since only 2% of paid sick leave was attributed to mental health disorders about 40 years ago. The discussion about stress has also reached the school context, at least after a significant school reform in Germany, which reduced high-school duration by one year with almost the same curriculum [4,5].

From a developmental neurobiology perspective, childhood and adolescence can be described as very vulnerable phases in which biological systems develop. Stress experience during this age can influence an individual's response to stressful events for their lifetime, mainly via the effects of an increased activation of the hypothalamic-pituitary-adrenal (HPA) axis, the main biological stress system, on the brain [6]. Stress exposure during childhood might therefore lead to a biologically-based susceptibility to stress-related illnesses later in life [7]. This is also related to lower academic achievement [8]. Measures to reduce stress and to build up stress resilience in schools need to be found.

There is promising research that exposure to green environments has some positive effects on mental health. Green environments can be described as areas with a certain amount of non-built spaces, for instance, public parks, lakes, rivers or forests. 'Green' does therefore not necessarily mean 'green' as a color, but rather stands as a synonym for nature with all its different shapes. In their recent systematic literature review, James et al. found that neighborhood greenness, or vegetation, may affect health behaviors and outcomes, and increased physical activity and social contacts may result in decreasing stress [9]. Their findings accord with a previous systematic literature review by Lee and Maheswaran, who concluded that most studies reported findings that generally supported the view of green environments having a beneficial health effect. However, they found that many studies were limited by poor study design, failure to exclude confounding, bias or reverse causality, and weak statistical associations [10]. On a general level, Roe et al. could associate more green space in deprived urban neighborhoods in Scotland to lower levels of perceived stress and improved physiological stress. This was measured by diurnal secretion patterns of the stress hormone cortisol and a steeper (healthier) diurnal cortisol decline with 104 subjects [11]. A similar correlation of green space and mental health factors seems to hold also for short-term visits of green space. Aspinal et al. found a relationship between green environment, behavior settings and emotions. They investigated the emotional experience of a group of walkers in three types of urban environments, including a green space setting, using mobile electroencephalography (EEG) as a method to record and analyze the $n = 12$ subjects' emotional experience. Their findings showed evidence when moving into the green space zone of lower frustration, engagement, arousal, and higher meditation; respondents showed higher engagement when moving out of it [12]. Certainly in accordance with these findings, Brantman and colleagues investigated the effect of a 90 min walk in a natural environment and found a reduction of blood flow in the subgenual anterior cingulate cortex, a region associated with stress regulation, as well as a reduction of rumination, a cognitive style associated with depression [13]. Interestingly, a lower activation in this brain region during acute social stress was found in individuals who grew up in a rural environment, compared to those who grew up in an urban environment [14]. A systematic literature review [15] compared effects of physical activity in outdoor natural environments with indoor environments on physical and psychological wellbeing. In contrast to being physically active indoors, physical activity in outdoor natural environments—so called green exercise—is associated with a decrease in tension, anger and depression. However, the authors conclude that the methodological quality of evaluated studies is poor and more high quality large-scale studies are needed in this field of research. On this basis, Rogerson, et al. [16] tested 331 participants before and after a 5 km run in four different natural environments, applying appropriate measurements and statistical analyses. Participants' stress and mood improved from pre- to post-run, independent of the specific green environment. The authors concluded that exercise in green environments offers possible benefits to psychological wellbeing.

In a series of studies, van den Berg and her team describe green space as a buffer between stressful life events and health for $n = 4529$ Dutch respondents [17]. They confirm the hypothesis that more time spent in green space is associated with higher scores on mental health and vitality scales, independent of cultural and climatic contexts, by comparing $n = 3748$ observations from four European cities [18]. They also describe gardening as an effective measure to promote neuroendocrine and affective restoration from stress in $n = 30$ active private gardeners [19].

A systematic literature review on the effects of school-based outdoor education programs on students' health, physical activity, social and learning dimensions, however, revealed that only very limited research has been reported so far [20]. Of more than 7800 articles analyzed, only 13 have met the inclusion criteria, of which six are so-called qualitative case studies, and seven apply (mostly poor) quantitative methodology. All studies are consistent in describing at least some positive effects of outdoor teaching to the various variables, health effects, physical activity, social and learning behavior. With respect to stress and mental health in the outdoor teaching context, we could identify only one slightly relevant study in the literature: Gustafsson and his colleagues describe that mental problems decreased in boys, but not in girls, in an outdoor teaching setting when compared to a control condition without outdoor teaching. These statistically significant effects were observed for a "difficulties total score", as well as for "emotional symptoms", "conduct problems", and "hyperactivity". Data were collected with a parent-report questionnaire [21]. Additionally, a promising quasi-experimental study design of the impacts of education outside the classroom on students' physical activity, well-being, and learning has been recently published. This study with n = 834 observations of children aged 9–13 years had been performed in Denmark in the past years, and the results are due in late 2017 [22]. Those results may be able to close some of the above-mentioned gaps in recent research and theory construction in "green exercise".

Taken together, the studies mentioned above give some evidence for a protective effect of a natural environment or outdoor setting on biological stress systems that might be related to mental health, also in the school context. However, to our knowledge, there is no prospective control group study investigating the effect of outdoor teaching on biological measures of stress. Therefore, we conducted the present pilot study, in which we hypothesized that regular intervals of outdoor teaching over the course of one school year will have a stress protective effect and, accordingly, will result in less activation of the HPA-axis, as reflected by a steeper decrease of cortisol secretion over the school day. The normal diurnal cortisol rhythm displays high cortisol values directly after getting up, with a steady decrease over the day [23–25]. However, it has been reported that adolescents with a high score on the Children's Depression Inventory show a reduced decline of morning cortisol, when compared to low scoring children [25]. Thus, cortisol appears to be a fitting measure for stress, with respect to mental health.

In addition, using an explorative approach, we investigated children's physical activity (PA) levels. Previous research indicates that children's PA-levels are consistently higher in outdoor education settings using natural environments, compared to normal indoor settings [26,27]. Since it is known that high PA can lead to higher cortisol levels [28], we needed to control whether the expected PA differences between outdoor and indoor would modulate potential differences in the stress response.

2. Materials and Methods

2.1. Participants, Intervention, and Data Collection

Participants were recruited from 5th grade students from a secondary school (German "Gymnasium") in Heidelberg, Germany. At the time of the study planning, two classes participated in the outdoor teaching program, consisting of one compulsory school day per week in the forest with the regular curriculum, while two classes were run as a traditional class without outdoor teaching. Students from those classes were to serve as control group. However, due to parent demand, the school decided to offer the outdoor teaching to three classes just before the school year, reducing the available number of control participants by 50%. Therefore, we included five students from a 6th grade indoor teaching class into the control group (CG). Finally, we were able to include 48 students into the study, 37 in the IG, and 11 in the CG. However, although we had no drop-outs, due to students being absent during the school year, we were not able to sample data from all students at all time points. Table 1 summarizes the enrolment data.

Table 1. Enrolment data.

	Participants Recruited	Fall	Spring	Summer
Total	48	46	45	46
Intervention	37	35	35	35
Control	11	11	10	11

Regarding the sociodemographic and anthropometric variables, slight differences exist between groups for age, weight and height. This is explained by the inclusion of students from 6th grade in the control group. There is, however, no statistically significant difference between the groups with respect to those variables that could potentially influence the biological measures. Nor was there a statistically significant difference in gender distribution (Table 2). The socio-economic status can be considered similar.

Table 2. Participant anthropometric data.

	Intervention Group	Control Group	Statistics
Age in fall	11.1 years	11.6 years	$p = 0.073$
Weight in fall	35.12 kg	35.67 kg	$p = 0.79$
Height in fall	145.3 cm	148.0 cm	$p = 0.21$
Gender	23 (62%) male 14 (38%) female	7 (64%) male 4 (36%) female	$p = 0.93$

The intervention consisted of one school-day per week in the forest. Thus, the overall mental load for the children is systematically the same, however, differently organized in the intervention. Looking at the respective schedules (Table A1 in Appendix A), two major differences can be seen: (1) the curriculum in IG is taught in cross-disciplinary units on the forest days, whereas it is taught in segments, subject by subject, in the CG; and (2) the pedagogical approach of the outdoor-learning program includes opportunities to be physically active on students' free choice, as well as planned walks to reach specific places in the forest. Due to traditional indoor teaching concepts, such opportunities are rather limited for students within CG. According to the judgement of the school's headmaster and teachers, the mental load of the five included sixth-graders in the CG can be compared to that of the other students in the CG. However, their curriculum is different, while the daily routines are the same.

Examination of the stress reactivity was performed by means of salivary cortisol analyzes with samples taken at three time points (8:30 AM, 10:30 AM, 12:30 PM) over the school year (seasons "fall", "spring", "summer"). Salivary cortisol was determined at the Biopsychology Laboratory at the Technical University Dresden, using a commercially available luminescence immunoassay (IBL, Hamburg, Germany). Due to the high variability of individual cortisol levels, the base-line is defined by the individual morning cortisol values at 8.30 AM, since we were only interested in relative individual cortisol concentrations over the day.

Physical activity of the control and intervention groups was determined by means of acceleration sensors. For this purpose, one Axivity AX3 sensor (Axivity Ltd., Newcastle upon Tyne, UK) was attached to each child's back above the upper point of the posterior iliac crest, with the aid of medical tape. Moderate-to-vigorous physical activity (MVPA) is a reference criterion for determining the recommended physical activity in children and adolescents [29]. As a first step, we converted the raw vector magnitude acceleration data to Actilife-format via an in-house software developed by the University of Southern Denmark. Afterwards, the children's MVPA was analyzed in Actilife v.6.11. 4 (ActiGraph, Pensacola, FL, USA). Cut-points reported by Romanzini et al. [30] have shown a good validity among children and adolescents at every activity level and were used to identify MVPA.

In order to determine long-term effects of the intervention on stress levels and to cross-validate the saliva measures, hair probes had been taken for ex-post analyzes of cortisol levels following the measurement days. However, data from the hair samples are not included in the present paper because of missing data, mostly due to hairs being too short to receive reliable probes.

2.2. Data Analyses

To account for the repeated measurements structure of the data and the complexity of interactions, we fitted linear mixed-effects models (LMMs), using the software package nlme [31,32] in R 3.3.2 (31 October 2016) [33], and JASP [34]. Without interaction terms, the general model for our analyses at time point j and season k = "fall", "spring", "summer" for individual i is:

$$Y_{ijk} = \beta_0 + b_i + \beta_1(season, k) + \beta_2(time.point, j) + \beta_3(group) + \varepsilon_{ijk}, \quad (1)$$

where β_0 is the intercept and the b_i's are the random intercepts being independent zero mean normally distributed random variables. The residuals ε_{ijk} are also zero mean normally distributed random variables with covariance matrix dependent on the situation as described below. Group is an indicator variable showing whether individual i belongs to the intervention or control group. The full factorial model, i.e., including all up to third order interactions between the fixed factors, was checked as a starting point.

With respect to the activity data, we analyzed the time points from 8:30 AM to 10:30 AM ("midmorning"), and from 10:30 AM to 12:30 PM ("noon"), then comparing those sets of moderate-to-vigorous physical activity (MVPA) over the fixed effects. We compared this model to less complex models by removing interactions, and had to include within-individual heteroscedasticity as a weighing factor (power of variance covariate) in order to include adjustment for residual variance dependent on MVPA values [35,36]. The model fit was evaluated using the AIC criterion and likelihood ratio tests (cf. Table S1). The model without 3rd order interaction showed the best fit to the data (cf. Figure S1).

With respect to the cortisol data, we had to include a general correlation matrix for the within-unit residuals. This resulted in a clearly better fit compared to using independent residuals. In order to obtain more symmetric data distributions facilitating assumption of normality, the cortisol values were log transformed. Starting from a full factorial model including the third order interaction between group, time, and season, the third order interaction, and the interaction between time and season, were excluded according to the Akaike information criterion (AIC), which is a measure of the relative quality of statistical models for a given set of data, and likelihood ratio tests. The resulting model showed good fit to the data according to residual plots (cf. Figure S2).

Since physical activity can result in higher cortisol values, we controlled cortisol against physical activity [28]. We therefore set up another set of models to analyze the interactions of MVPA- and cortisol-measures. In a first approach, we tested the accumulated MVPA-values from the subsets 8:30 AM–10:30 AM, and 10:30 AM–12:30 PM, respectively, against the difference of cortisol measures (diff_logCortisol) at 10:30 AM compared to 8:30 AM, and 12:30 PM compared to 10:30 AM, respectively, using a linear mixed effects model with random intercept (2).

$$Y_{ijk} = \beta_0 + b_i + \beta_1(MVPA) + \beta_2(season, k) + \beta_3(time.point, j) + \beta_4(group) + \varepsilon_{ijk}, \quad (2)$$

$$Y_{ik} = \beta_0 + b_i + \beta_1(Sum\_MVPA) + \beta_2(season, k) + \beta_3(group) + \varepsilon_{ik}, \quad (3)$$

Again, we compared the full interaction models to less complex models by removing interactions. The model fit was evaluated using the AIC criterion and likelihood ratio tests. The model without the interaction over time showed the best fit to the data. Additionally, we tested the full accumulated MVPA-values at 12:30 PM (Sum_MVPA) against the overall difference of the cortisol values (delta_logCortisol) in another set of models (3) with varying interactions, including random intercept effects. The inclusion of a general correlation matrix for the within residuals in the model was not necessary according to the AIC criterion (cf. Table S3, Figure S3). Again, the model without interaction over time (season) showed better fit to the data than the full model.

3. Results

3.1. Physical Activity

As could be expected with respect to the school setting, the children in the outdoor classes show higher activity levels than their peers in the school building. Table 3 gives detailed descriptive information on the data, and Figure 1 displays a graphical output.

Table 3. Descriptives-Moderate-to-vigorous physical activity (MVPA) [min].

Group	Time.Point	Season	Mean	SD	*n*
Control	midmorning	fall	10.068	4.996	11
		spring	9.900	5.589	10
		summer	12.458	5.551	6
	noon	fall	7.977	3.414	11
		spring	19.400	13.808	10
		summer	12.417	5.953	6
Intervention	midmorning	fall	21.000	9.655	32
		spring	14.396	6.399	34
		summer	21.333	8.984	24
	noon	fall	30.828	10.640	32
		spring	28.794	10.491	34
		summer	25.427	10.832	24

SD: Standard Deviation.

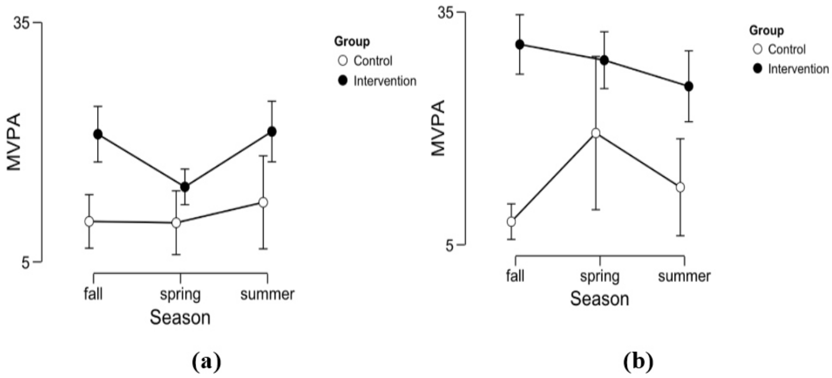

Figure 1. Here, the different moderate-to-vigorous physical activity (MVPA) levels are displayed for (**a**) the accumulated values from 8:30 AM to 10:30 AM, and (**b**), the accumulated values from 10:30 AM to 12:30 PM with respect to seasons and group. The descriptive parameters can be seen in Table 3. The error bars indicate the 95% confidence interval (CI). Inferential analyses reveal that intervention group (IG) are estimated 11:30 min longer in MVPA levels (SE = 2.08) than the control group (CG) ($p < 0.001$) per 2-h time interval (cf. Table A2 in the Appendix B). The difference is especially bigger in the second half of the school day ($p < 0.001$).

The main effect revealed by the linear mixed effect model is as follows: children in the forest group are expectedly 11:30 min longer in MVPA-level per 2-h-unit than their peers back in school ($p < 0.001$), averaged over the whole school year. The difference is especially larger in the second half of the school day (difference: 7:54 min; $p < 0.001$) (cf. Table A3 in Appendix C). Seasonal effects can also be observed. However, the expected time in MVPA in spring and summer is relatively shorter for IG than CG (difference spring: −7:36 min, $p < 0.000$; difference summer: −6:30 min, $p = 0.004$), which is due to a light decrease of time spent in MVPA in IG over the seasons with its lowest value in spring. Meanwhile, time spent in MVPA simultaneously increases in the CG with its highest value in spring. Comparing the means of the accumulated time in MVPA over the three measurement days without accounting for seasonal or diurnal differences shows that IG spent more than twice as much time in MVPA than CG ($M_{IG} = 47.18$ min, $M_{CG} = 23.28$ min; $t\ (51.162) = -7.763$, $p < 0.001$).

3.2. Cortisol Measures

The cortisol measures are log-distributed, as can be seen from the Table A2, and the graphical displays in Appendix B. Thus, we performed the statistical analyses of cortisol with logarithmized data. The linear mixed effect model of the log-cortisol data yields that the intervention group in the outdoors have a statistically significant greater decline of cortisol compared to the control group; rate 0.0102 µg/L + 0.0588 µg/L = 0.069 µg/L vs. 0.0102 µg/L (log-units/2 h, $p = 0.009$) (cf. Figure 2). Moreover, the intervention group has expected lower cortisol levels in spring at the half-year compared to control group, difference: 0.0915 µg/L, $p = 0.050$, which is still statistically significant for the end of the school year, difference: 0.0879 µg/L, $p = 0.052$ (cf. Table A4 in Appendix C).

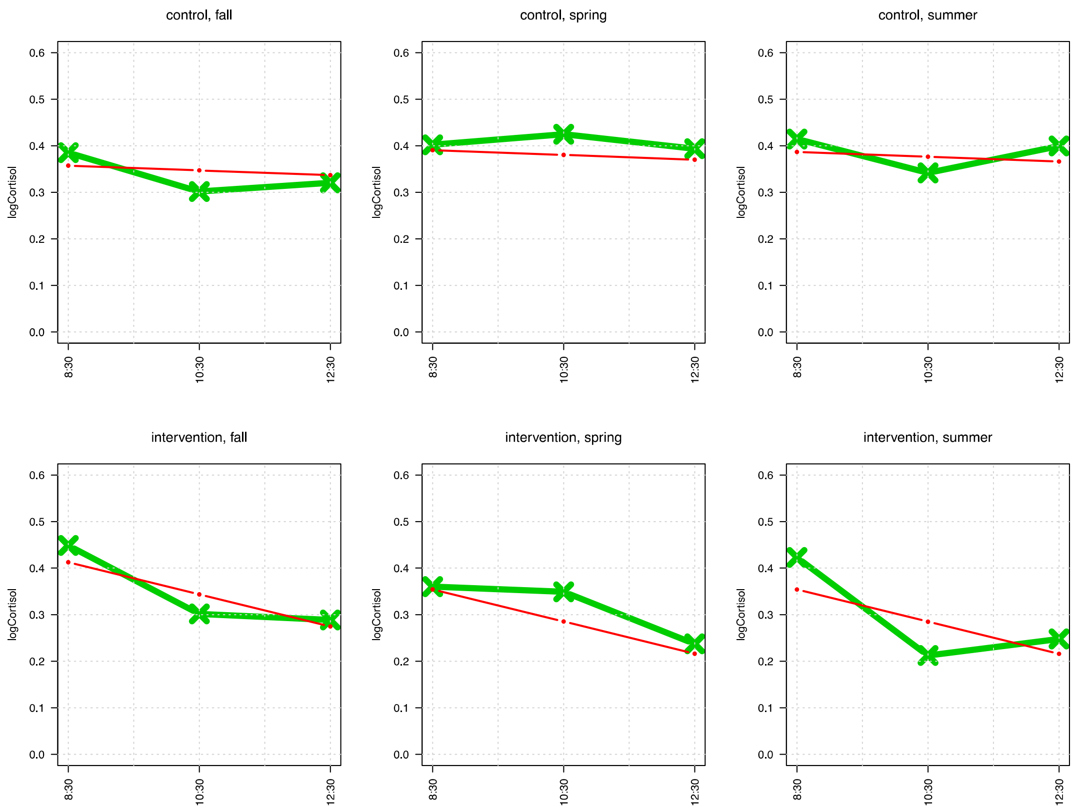

Figure 2. Displayed are the running curves of logCortisol over the day in each season for both groups. The upper panels show the CG values, the lower panels IG values. The green line represents the mean values, the red line connects the calculated values of least squares regression. It can be seen that IG shows, in contrast to CG, a clear decrease of cortisol levels in the course of the school days, but not the CG ($p = 0.009$).

3.3. Interaction of Cortisol Measures and Physical Activity

Both strategies of testing for interaction between the cortisol measures and moderate to vigorous physical activity did not yield statistically significant results. Tables A5 and A6 in Appendix C show the results of the above-mentioned interaction models (2) and (3).

4. Discussion

4.1. General Observations

The present study was conducted to investigate whether regular engagement in an outdoor teaching has a positive effect on stress responses in students, and whether this effect is associated with physical activity in this setting.

The findings of the cortisol measures allow for a straightforward interpretation: In our case, teaching in the forest was associated with a lower cortisol secretion at noon, compared to the control group. Given that the normal diurnal cortisol rhythm displays high cortisol values directly after waking up, which steadily decreases over the day, the lack of such a decrease of cortisol over the school-day in the control group might be regarded as detrimental [23–25]. It has been shown that adolescents with a high score on the Children's Depression Inventory also show a reduced decline of morning cortisol, when compared to low scoring children [25]. It could therefore be argued that the cortisol profile observed in the indoor class is rather similar to profiles observed in individuals prone to develop a stress associated mental disorder, such as depression. However, it is important to remember that cortisol secretion is influenced not only by psychosocial stress, but also by a number of other conditions including physical activity, mental load, or different positive stressors [37]. However, while we can exclude differences in physical activity as a potential factor underlying group differences (see above), we cannot exclude differences in mental load or eustress between groups. Explaining the effect with eustress seems implausible, since the participants in the forest classes did very likely not experience less positive events, and less fun, than those in school. With respect to positive social events, literature suggests that students in outdoor classes experienced more positive encounters during the school day than those in the indoor class [38], which would counter the cortisol effect observed in our study. The mental load is difficult to estimate. In principle, both groups had the same curriculum, with the exception of the five six-graders. However, the individual school subjects had not been absolutely synchronized, nor had the lessons been delivered by the same teachers (cf. Table A1 in the Appendix A). Moreover, literature suggests that students taking part both in short- and long-term outdoor teaching programs likely do not consider the outdoor teaching as "regular school lessons," despite long working hours and a very advanced curriculum [38,39]. Furthermore, they show a higher degree of long-term knowledge retention [38] and emotional connectedness to the curriculum [40,41]. Thus, some of the effect might be attributed to differences in mental load, especially defined by the teaching context.

As mentioned before, the cortisol data are very likely not confounded by the students' physical activity, as the statistically non-significant interaction analyses show. This is interesting in two respects—the "dosage" of physical activity on a "typical" outdoor schooling day seems to be such that children are: (a) comparatively very physically active but without; and (b) are impacted on their biological stress system. Hill et al. [28] report that a statistical increase of cortisol can only be reached at an exercise intensity level of 80% of the VO_2 max for 30 min, which reflects a substantially higher exercise load than the one achieved in our intervention group. After corrections for circadian factors, lower exercise loads than 80% of VO_2 max for 30 min may actually result in a reduction of circulating cortisol [28]. However, VO_2 max related thresholds are relative to the individuals' levels of physical exercise capacity, whereas the accelerometer data in our study are absolute measures of the amount of PA. Both measurement variables are related but not directly comparable. Thus, we cannot explore this further with our data. On the other hand, the lack of interaction between MVPA and cortisol suggests that the observed stress buffering effect of the outdoor teaching setting can be attributed to the specific environment. The natural environment of the forest offers potential—so far unspecified in the educational context—influences on: perception, social aspects, experiences and, specifically, exposure to sunlight [42]. These aspects can be subsumed as a so-called "green effect". There is also some evidence from the literature that such a "green effect" [43–46], together with some learning psychological aspects [39,47], might add to the stress reduction in the intervention group.

With respect to the findings of the children's activity levels in the outdoor vs. the indoor classes, our results directly confirm (in part) previous Danish research by Mygind [26,27]. As reported, the outdoor classes in our study led to statistically significant higher PA levels, compared to the indoor classes. In both Danish studies, the involved students participated in similar regular curriculum-based outdoor education projects. Students' PA was objectively measured during outdoor and indoor learning and compared intra-individually. Students' PA was statistically significant higher during outdoor learning days, compared to traditional indoor school days. We cannot, however, explain the seasonal effects with either the literature or data from our study. From a practical perspective, we would consider those differences as artefacts, and not relevant. However, further research with more measures over the year might support another conclusion.

Taken together, our findings give some preliminary support for the often assumed, but so far empirically unconfirmed, hypothesis that outdoor teaching over regular intervals is beneficial to children's mental and physical health [48], which supports our main hypothesis. This is some of the first research into biological stress factors in the outdoor education context. The results are therefore unique, but have some limitations.

4.2. Limitations

Since the hair cortisol probes could not be reliably analyzed, we were not able to check for mid- or long-term or buffer-effects of the stress reduction in the outdoors, hence stress-resilience. Moreover, three measurement days over one school year provide too coarse a resolution for fully understanding the phenomenon. However, more measurement days were not possible for logistical and school organizational reasons, and more measures would have inevitably led to even less enrolment, and probably more drop-outs. Clearly, more research is needed to understand patterns at a finer level of detail. More insights into students' diurnal cortisol responses could also be realized by testing the IG on normal indoor school days.

Another critical point for the present study is the non- or "quasi"-randomization of the participants into the particular groups. While the group allocation was not done by the experimenter, it was determined by school policies and according to the parents' (and children's) preferences. This might in itself bear some bias which cannot be corrected with statistical methodology. For ethical reasons, we could not test the children for behavioral or mental health disorders. The parents in favor of outdoor teaching against "normal" schooling might have children yielding a certain cluster of psychological straits we are not aware of, and whose statistical prevalence has not yet been researched.

5. Conclusions

The main result of our study is that the children in the forest class show a steady decline of cortisol during the school day which was not observable in the control group. This is in accordance with a healthy child's diurnal rhythm, and its statistically significant decline of cortisol from morning to noon. This effect is constant over the school year. The children in the classroom setting did not show this effect on either measurement day. However, our data gave no empirical hints to explain that interesting phenomenon. Despite the mentioned limitations of the current study, the cortisol data are consistent and valid. Further, the data supports the conclusion that outdoor education had a positive effect on stress responses in children in our intervention group, in contrast to indoor teaching in the control group. These novel findings, interesting as they are, only represent a first step towards a deeper understanding of the "stress in school" phenomenon measured with biological parameters. Larger prospective studies are needed to confirm the results, and to potentially test for consequences of reduced stress exposure in outdoor setting, with respect to mental health in children and adolescents.

Supplementary Materials: The following are available online at www.mdpi.com/1660-4601/14/5/475/s1, Figure S1: Standardized MVPA residuals, Table S1: Model fit parameters MVPA, Figure S2: Standardized logCortisol residuals, Table S2: Model-fit parameters logCortisol, Figure S3: Standardized logCortisol by MVPA interaction residuals, Table S3: Model-fit parameters logCortisol by MVPA interaction.

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Author Contributions: Ulrich Dettweiler, Peter Kirsch and Perikles Simon conceived and designed the study; Christoph Becker collected the data; Ulrich Dettweiler, Peter Kirsch, Perikles Simon and Bjørn H. Auestad analyzed the data; Ulrich Dettweiler wrote most of the paper with substantial contributions from all other authors. All authors proved the final version of the manuscript.

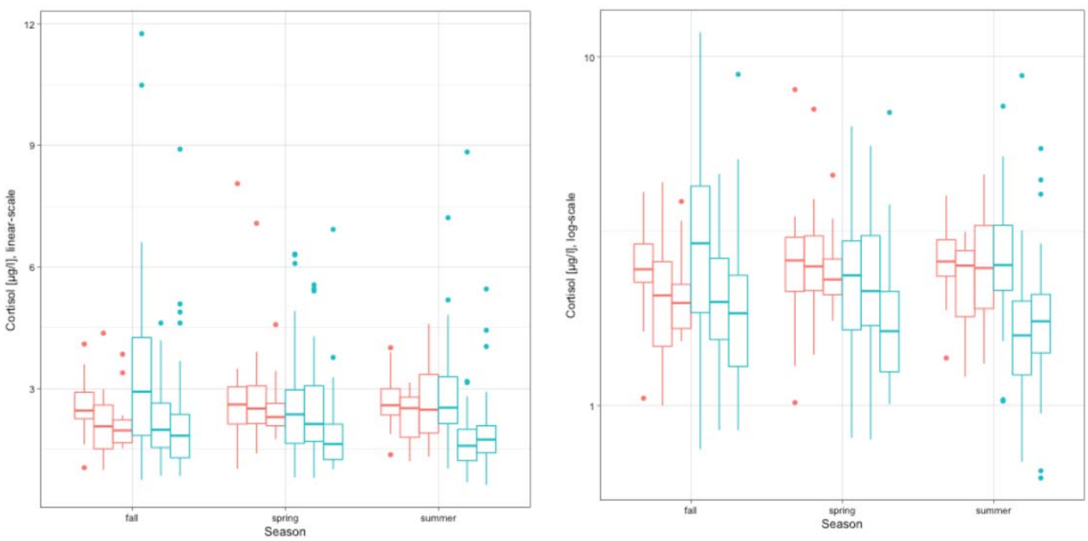
Conflicts of Interest: Peter Kirsch received consulting fees from Biologische Heilmittel Heel. All other authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results. The study has been approved by the internal review board at the University of Heidelberg. Written consent has been given. Data are stored according to international ethical and legal guidelines.

Appendix A

Table A1. School schedule and measurement procedures.

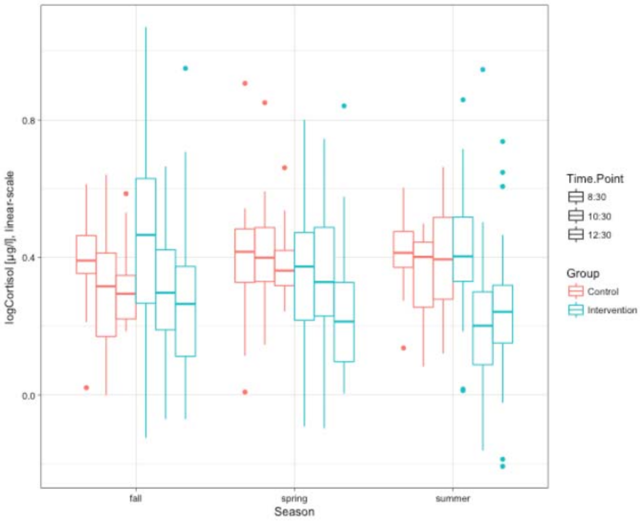
Schedule	Time Data Collection	IG	CG
07.55–08.40	8:30	Meeting at 8.00 and short walk to outdoor "classroom"; preparing for the day	regular class according to curriculum
08.45–09.30		forest class according to curriculum	regular class according to curriculum
09.30–09.45		break	break
09.45–10.30	10:30	continued forest class according to curriculum	regular class according to curriculum
10.35–11.20		continued forest class according to curriculum	regular class according to curriculum
11.20–11.35		break	break
11.35–12.20		continued forest class according to curriculum	regular class according to curriculum
12.25–01.05	12:30	continued forest class according to curriculum	regular class according to curriculum

Appendix B



(**a**) (**b**)

Figure A1. *Cont.*



(**c**)

Figure A1. Cortisol raw values compared to log10-transformed cortisol values. Displayed are (**a**) the raw cortisol values on the three time-points (8:30, 10:30, 12:30) at the three seasons (fall, spring, summer) in both groups (control = red, intervention = green). Panel (**b**) shows the same data with a logarithmized y-axis and (**c**) the logarithmized cortisol-values on a linear y-scale. It can be seen that by log-transforming the y-scale (**b**), some outliers can be eschewed, and that by logarithmizing the cortisol-values (**c**), the variance, skewness, and kurtosis of the data can be reduced (cf. Table A1).

Table A2. Descriptives for logCortisol and Cortisol.

	Cortisol	LogCortisol
Mean	2.445	0.3322
Std. Error of Mean	0.07080	0.01072
Std. Deviation	1.419	0.2150
Variance	2.015	
Skewness	2.413	0.3124
Kurtosis	9.287	0.2787

Appendix C

Table A3. Summary of interaction analyzes MVPA [min].

	Value	Std. Error	DF	*t*-Value	*p*-Value
(Intercept)	9.361	1.59	178	5.889	0.000
IG	11.539	2.08	46	5.547	0.000
S_{spring}	−0.107	1.198	178	−0.089	0.929
S_{summer}	4.664	1.962	178	2.377	0.019
t_{noon}	−0.775	1.107	178	−0.7	0.485
IG:S_{spring}	−7.606	1.703	178	−4.466	0.000
IG:S_{summer}	−6.526	2.249	178	−2.901	0.004
IG:t_{noon}	7.914	1.657	178	4.777	0.000
S_{spring}:t_{noon}	7.767	1.806	178	4.3	0.000
S_{summer}:t_{noon}	−1.261	2.137	178	−0.59	0.556

IG = intervention group, t = time point of the day (morning, midmorning, noon), S = season (fall, spring, summer).

Table A4. Summary of interaction analyzes logCortisol [µg/L].

	Value	Std. Error	DF	*t*-Value	*p*-Value
(Intercept)	0.3675	0.0567	349	6.4843	0.000
t	−0.0102	0.0196	349	−0.5206	0.603
IG	0.1141	0.065	45	1.7555	0.086
S_{spring}	0.0332	0.04	349	0.8302	0.407
S_{summer}	0.0293	0.0392	349	0.7482	0.455
t:IG	−0.0588	0.0225	349	−2.6175	0.009
IG:S_{spring}	−0.0915	0.0459	349	−1.9948	0.050
IG:S_{summer}	−0.0879	0.0451	349	−1.9476	0.052

IG = intervention group, t = time point of the day (morning, midmorning, noon), S = season (fall, spring, summer).

Table A5. Interaction of physical activity on cortisol (segmented 8:30–10:30, 10:30–12:30).

	Value	Std. Error	DF	*t*-Value	*p*-Value
(Intercept)	−0.010	0.064	177	−0.163	0.871
MVPA	0.001	0.004	177	−0.180	0.857
IG	−0.096	0.080	45	−1.192	0.240
MVPA:IG	0.002	0.005	177	0.437	0.662

IG = intervention group, MVPA = moderate-to-vigorous physical activity level, segmented.

Table A6. Interaction of physical activity on cortisol (whole day).

	Value	Std. Error	DF	*t*-Value	*p*-Value
(Intercept)	−0.043	0.082	65	−0.528	0.600
Sum_MVPA	0.001	0.003	65	0.250	0.803
IG	−0.201	0.112	45	−1.790	0.081
Sum_MVPA:IG	0.001	0.003	65	0.389	0.699

IG = intervention group, Sum_MVPA = moderate-to-vigorous physical activity level accumulated over the school day.

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Children's Cortisol and Cell-Free DNA Trajectories in Relation to Sedentary Behavior and Physical Activity in School: A Pilot Study

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The worldwide prevalence of mental disorders in children and adolescents increased constantly. Additionally, the recommended amount of physical activity (PA) is not achieved by this age group. These circumstances are associated with negative impacts on their health status in later life and can lead to public health issues. The exposure to natural green environments (NGE) seems to be beneficial for human health. The compulsory school system offers great opportunities to reach every child with suitable health-related contents and interventions at an early stage. The concept of Education Outside the Classroom (EOtC) uses NGE and sets focus on PA. Therefore, EOtC might be a beneficial educational intervention to promote students health. The association between biological stress markers and sedentary behavior (SB) plus PA is insufficiently evaluated in school settings. This exploratory study aims to evaluate the association between students' cortisol, plus circulating cell-free deoxyribonucleic acid (cfDNA) levels, and their SB, light PA (LPA), and moderate-to-vigorous PA (MVPA). We assessed data from an EOtC program (intervention group [IG], $n = 37$; control group [CG], $n = 11$) in three seasons (fall/spring/summer) in outdoor lessons (IG) in a NGE and normal indoor lessons (CG). SB and PA were evaluated by accelerometry, and cortisol and cfDNA levels by saliva samples. Fitted Bayesian hierarchical linear models evaluated the association between cortisol and cfDNA, and compositional SB/LPA/MVPA. A steady decline of cortisol in the IG is associated with relatively high levels of LPA (posterior mean = −0.728; credible interval [CRI 95%]: −1.268; −0.190). SB and MVPA tended to exhibit a similar effect in the CG. A high amount of cfDNA is positively associated with a relatively high amount of SB in the IG (posterior mean, 1.285; CRI: 0.390; 2.191), the same association is likely for LPA and MVPA in both groups. To conclude, LPA seems to support a healthy cortisol decrease in children during outdoor lessons in NGEs. Associations between cfDNA and SB/PA need to be evaluated in further research. This study facilitates the formulation of straightforward and directed hypotheses for further research with a focus on the potential health promotion of EOtC.

Keywords: cortisol, cfDNA, physical activity, health, outdoor environment, Bayesian inference

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INTRODUCTION

The upsurge in the worldwide prevalence of overweight and obesity in children is anticipated to reach 9.1% in 2020 (1), a high proportion of children do not reach the recommended levels of physical activity (PA) (2, 3), and suffer from mental disorders (4). Chronic stressful events could exert adverse impacts on brain development and result in major mental health-related problems in later life (5, 6). These circumstances require a need for action to improve children and adolescents health perspectives. Successful interventions should therefore consider that (1) PA and exercise during childhood is associated with the development of active lifestyles in later life, improved cognitive functions (7, 8) and, thus, with positive effects on health and prevention of common diseases (9), (2) the exposure to natural green environments (NGE) can have beneficial health effects [see e.g., (10, 11)], and that (3) children spend a substantial share of their waking hours in school. Therefore, the compulsory school system in western countries offers excellent opportunities to reach every child and adolescent with specific interventions focusing on PA in NGE to improve children and adolescents health perspectives.

The present paper aims to address these topics by investigating the relation between biological stress responses and physical activity in students taught in two different school settings: an indoor setting and an outdoor setting in a NGE. Here, we extend our original investigation (12) by increasing our set of dependent variables and introducing circulating cell-free deoxyribonucleic acid (cfDNA) as an innovative biological marker, sedentary behavior (SB) and light physical activity (LPA) as more differentiated measures of physical activity and by applying advanced statistical models to better describe relations between our measures.

Both cortisol and cfDNA are important biomarkers in relation to stress, SB and PA. Thus, the comparison of both cortisol and cfDNA in relation to students' relative levels of SB and PA is a promising approach to investigate students' biological stress response in different school settings. Important findings on cortisol and cfDNA in relation to physical and psychosocial stressful situations are therefore outlined.

Recent studies have focused on exploring the construct of "stress" and its potential negative association with health (13). In fact, an individual's physiological and psychological response, assessed by different stress biomarkers or questionnaire items, could be correlated with several positively, as well as negatively, connoted stimuli. Koolhaas et al. (13) argued that the term "stress" should be restricted to situations of uncontrollability or unpredictability of stimuli which however, must be restricted to "psychological stress" and is not true for so called "physical stress" (14, 15), which can be defined as a loss of homeostasis induced by physical not psychological conditions. Examples of such uncontrollable situations in school are examinations, testimonials, increased mental loads or prolonged social pressure (16, 17). Such stressors can lead to an interruption of the regular circadian cortisol rhythm. The relevance of a normal diurnal cortisol rhythm with high levels of cortisol in the morning and a steady decline until evening has been widely investigated (18–20). Furthermore, several external stimuli could be involved in the disturbance of a normal diurnal cortisol rhythm, for instance, light pollution during nighttime, or continuous changes in waking hour schedules. Moreover, a dysfunctionality in the hypothalamic pituitary adrenocortical (HPA) axis as one primary biological stress system plays a crucial role. In a recent systematic literature review and meta-analysis, Adam et al. (21) reported that a chronic abnormal flat diurnal cortisol rhythm correlated with poor mental and physical health symptoms for various populations. Other experimental studies (22, 23) evaluated the association between cortisol levels and PA, with a particular focus on different PA intensities, as well as the diurnal cortisol rhythm. These studies revealed that high PA intensities ranging from 60 to 80% of the maximal oxygen uptake (VO2 max) (22) or 80% VO2 max (23) for, at least, 30 min resulted in statistically significant higher cortisol levels compared with resting control situations. Interestingly, participants' cortisol levels decreased, although not statistically significant, not only in the resting control groups (CG) but also during low PA intensities of 40% VO2 max. These studies illustrate the potential impact of PA on cortisol levels.

Besides the well-established but also critically discussed stress marker cortisol (24), the circulating cfDNA has garnered more importance as a potential physiological stress marker. Different mechanisms can result in the release of the cfDNA into the human plasma. While an increase in cfDNA levels because of classic cell-death mechanisms would take several hours, or even days, other more rapid mechanisms are related to exercise. Based on plasma samples, the cfDNA is a well-established indicator of the activation of the innate immunity. Various studies have revealed that the innate immunity could be activated by both psychologically (25, 26) and physiologically (27–32) stressful situations. In particular, the cfDNA has been proven to be highly sensitive to physical exercise as a stressor [see (27) for review]. Reportedly, the cfDNA increased with moderate PA below the level of the aerobic–anaerobic transition (29, 33). A recent study (30) reported that cortisol and plasma cfDNA levels positively correlated and both increased in participants under physiological and psychosocial stressful situations. Regarding psychological stress little is known about the reactivity of cfDNA concentrations. To date, only one study has reported that lowering psychological stress in women treated for infertility reduces the plasma cfDNA concentration, a notion that is principally in line with the concept of a stress-associated, sensitive proinflamatory marker (34). Furthermore, current research suggests that major depressive symptoms are associated with elevated levels of cfDNA (35, 36). Cianga et al. (37) studied cfDNA in saliva of immunosuppressed patients. The results indicate that the most important source of DNA in saliva samples are leukocytes that travel from the blood to the oral cavity, where they play an important role in protection against pathogens. The specific cells and tissues, which are involved in psychological induced cfDNA elevation, are still unknown. However, the effect of stress hormones on leukocyte profiles is well-documented in biomedical studies of mammals. This includes glucocorticoid-induced alterations in cell trafficking, or redistribution from blood to other body compartments [reviewed in Davis et al. (38)]. This furthermore indicates an indirect link between cortisol and cfDNA. Higher cortisol values were associated with a greater number of neutrophils (38). In response to infection, tissue injury or exercise, neutrophil glucocorticoids can produce extracellular traps, which are likely to contribute to the pool of cfDNA (39). Most research on the cfDNA is restricted to plasma samples and controlled laboratory settings. However, a study has reported that the cfDNA in the saliva and serum possess a similar half-life time and both follow a first-order clearance model (40). Furthermore, in both body fluids, the cfDNA seems to be predominantly released by cells of the hematopoietic lineage (31, 37). To the best of our knowledge, no research has investigated the association between cfDNA levels based on salivary samples and exercise in an experimental setting, at least, in schools.

SB and PA are relevant factors with different effects in relation to health (3, 41) and in addition potential confounders for cortisol and cfDNA. Therefore, recent research developments with respect to SB, PA and health are outlined. With regard to public health, the relevance of SB and LPA has gained more attention recently. The authors of a recent review (42) suggested that high values in sedentary time correlated with an increased risk of cardio-metabolic disease, decreased fitness, self-esteem, academic achievement, and pro-social behavior for children and adolescent. Very obviously there is also a relation between SB and mental health, particularly depression (43). LPA seems to be beneficial to reduce obesity, overall mortality risk and should be considered for inclusion in PA recommendations (44). Therefore, it is of great importance to consider all parts of human behavior and especially to account for the compositional nature of SB, LPA and moderate-to-vigorous physical activity moderate-to-vigorous PA (MVPA). This approach has been proposed in the recent years (45, 46).

A great responsibility for children's PA and health could be assigned to educational institutions and their schedules. Apparently, students' time in school and its environment play a crucial role. Typically, NGE seem to be beneficial for promoting children's PA (47, 48), mental well-being (47) and cognition (49, 50). The amount of time children being exposed to NGA seems to be important for various health outcomes. Therefore, questions arise how the exposure to NGA and being physically active in NGA can contribute to enhance students PA and stress response during school time.

In a recent systematic literature review (51), we assessed the effects of regular compulsory school and curriculum-based education outside the classroom [EOtC, (52)] programs, focusing on students' health, PA, social, and learning dimensions. EOtC often takes place in both NGE and cultural settings. Students seem to benefit regarding learning and social dimensions. However, only one study reported improved mental health status of boys (53) and two studies (54, 55) reported higher PA levels during days with EOtC compared with regular school days. Unfortunately, the methodological quality of the 13 included studies was mostly moderate or low. Moreover, a recent large-scale study (56) on EOtC reported that the MVPA levels were significantly higher during EOtC compared with regular school days. However, the codependency among students SB, LPA, and MVPA levels remained unclear in this study. Overall, the existing knowledge on effects of EOtC with regard to PA and health is limited, despite the mentioned potentials of this type of teaching setting. Especially in the Scandinavian countries the EOtC approach is widely spread, creating good opportunities for further research (57).

In our recent publication (12), we compared the cortisol levels of students taught by applying an outdoor curriculum in the forest with children taught in the standard school setting. We were primarily interested in assessing the effect of outdoor teaching on children's normal diurnal cortisol rhythms. We reported that students in the intervention group (IG) exhibited a steady decline of cortisol levels during EOtC, whereas no such effect was observed in students in the CG during regular school days; in fact, the effect was independent of students MVPA levels. However, we could not entirely elucidate the differences in students' cortisol levels. We believe that the partial secondary exploitation of the data presented in this study is justified by the new knowledge gained, as we analyzed the cortisol and cfDNA values concerning the compositional nature of SB and PA.

This exploratory, longitudinal analysis aims to evaluate the association between students' cortisol and cfDNA levels and their SB, LPA, and MVPA in outdoor and indoor classroom environments. Based on our previous research, we assumed that different relations exist between the CG and the IG with respect to their cortisol response and PA. Specifically, we hypothesized that a decrease in students' cortisol levels can be explained by their compositional levels in SB, LPA and MVPA and explored if similar relationships exist for students cfDNA response.

MATERIALS AND METHODS

Study Design and Intervention

This exploratory analysis is part of the research project "1 year in the forest–the influence of regular outdoor lessons in a natural environment on biological indicators of stress resilience." The research in the NGE comprised a great complexity concerning measurement procedures and confounding factors. Thus, in this project, we applied a mixed-methods approach in a prospective, longitudinal quasi-experimental design. In addition, functional magnetic resonance imaging, saliva cortisol and saliva cfDNA, three-axis accelerometry, and constructs of the Self-Determination Theory were used as described by Dettweiler et al. (12).

This intervention study was conducted at a secondary school in Heidelberg, Germany. Since the school year 2013–2014, a group of fifth-grade students were taught one compulsory school day per week for the entire schoolyear in a nearby forest. The pedagogical concept of the forest teaching setting was inspired by the Scandinavian udeskole/uteskole approach as well as outdoor education from New Zealand [see (57–59) for further details]. Thus, teachers intended to facilitate student-centered, hands-on, and experimental learning situations in close connection to the NGE. In addition, this change of space within the physical setting of the "classroom" implied different opportunities for problem-solving, co-operation, experimentation, and to be physically active on students' free choice during the lessons. Furthermore, students undertook regular walks to reach specific places in the forest. Of note, the contents of the lessons in the forest setting were highly connected to the formal school curriculum and were taught in cross-disciplinary units on the forest days, including a certain variance concerning the practical relevance and season. Moreover, subject-by-subject teaching was applied on standard school days for both the IG and the CG based on traditional indoor teaching concepts [refer Dettweiler et al. (12) for further information regarding timetables and Von Au (60) for the pedagogical concept].

Participants and Data Collection

We enrolled participants from fifth and sixth grades from the school year 2014–2015. In this school year, three fifth-grade classes had forest teaching, and only one fifth-grade class had regular indoor teaching. Owing to this administrative decision of the school, we could not enroll the same number of fifth-grade students in the IG and CG. Thus, we enrolled students from a sixth-grade regular indoor teaching class into the CG; these students did not participate in the forest teaching setting during their fifth-grade school year 2013–2014. Overall, we enrolled 48 students in this study (IG, 37; CG, 11). As some students were absent during the school year, we could not collect datasets from all 48 students at all-time points in fall, spring, and summer. Furthermore, not all saliva samples provided adequate material for analysis, and accidentally acceleration sensors got lost. Of note, descriptive and enrollment data for participants is presented elsewhere (12).

We collected both samples for saliva cfDNA and cortisol using Salivette™/Cortisol- Salivette™ collection tubes (Sarstedt, Nümbrecht, Germany) at time points 08:30 a.m., 10:30 a.m., and 12:30 p.m. during the seasons fall, spring and summer. All participants were told not to eat 15 min prior every data collection. Saliva cfDNA levels were evaluated using undiluted saliva according to the protocol described elsewhere (61). After centrifuging at 1,600 × g for 2 min (room temperature), the supernatant was transferred into a new collection tube and frozen at −20°C before measurement. In addition, salivary samples for cortisol quantification were frozen at −20°C immediately after the arrival at the Biopsychology Laboratory, Technical University Dresden, and cortisol levels were determined using a commercially available luminescence immunoassay (IBL, Hamburg, Germany). Based on the validation study by Khoury et al. (62), we applied the summary indices peak reactivity (PR) and the area under the curve with respect to increase (AUCi). (For further details regarding the calculation and application of the summary indices, refer to Fekedulegn et al. (63), Khoury et al. (62), and Pruessner et al. (64) and the **Supplementary Material**, section Material and Methods).

We determined both SB and PA of the IG and CG using triaxial Axivity AX3 acceleration sensors (Axivity Ltd., Newcastle upon Tyne, UK). One sensor was attached to each child's back above the upper point of the posterior iliac crest, with the aid of a medical tape (56, 65). The sensors were worn between 08:30 a.m. and 12:30 p.m. during school time. All children were instructed not to re-attach the sensor to their skin once it fell off. All sensors were initialized at 100 Hz and ±8G bandwidth. In addition, we converted the raw vector magnitude acceleration data to ActiLife file format by an in-house software developed by the University of Southern Denmark. Children's PA levels were analyzed using ActiLife v.6.11.4 (ActiGraph, Pensacola, FL). In addition, cut-off points reported by Romanzini et al. (66) were used to distinguish SB, LPA, and MVPA; these cut-off points have been proven to exhibit a good validity among children and adolescents to identify patterns of SB, LPA, and MVPA. However, the validity and comparability of acceleration sensors, as well as applied cut-off points, have been controversially discussed. Therefore, certain differences have to be considered when comparing studies on SB and PA, especially effects of varying epoch lengths, wear time algorithms, and activity cut-points (67–69).

Statistical Analyses

In studies on PA and health, one specific behavior is often analyzed independently from other behaviors. Recent studies focused on this issue and reported that human behavior during a finite time of the day needs to be recognized as a composition that accumulates to 100% of that time. Thus, the components (e.g., sleep, SB, LPA, MVPA) are perfectly codependent and an approach that considers all parts of the composition is recommended to provide reliable evidence on human behaviors related to health (45, 46).

To set up, document and run the Bayesian hierarchical linear models (BHLMs), to evaluate associations between students' cortisol and cfDNA levels, respectively, and their relative time spent in SB, LPA, and MVPA, we applied the software packages ggthemes (70), jagsUI (71), rjags (72), and R2jags (73) in R 3.4.1 (2017-06-30) (74). The usual way to fit regression models with compositional covariates is to apply isometric log-ratio (ilr) or centered log-ratio (clr) transformations on raw values, which is justified as the parts of a composition perfectly correlate and standard regression techniques result in multicollinearity problems. However, the use of ilr or clr transformations poses problems with the interpretation, as the meaning of posterior parameter values remains unclear, especially in hierarchical models. Thus, in the given analysis, a Bayesian ridge regression version suggested by Parnell (75), which accepts raw compositions, was implemented and the raw composition values were transformed into a matrix using a common prior distribution function.

The likelihood for the applied BHLMs reads

$$Y_i \sim N\left(\alpha_{\mathrm{id}_i} + \beta_{\mathrm{cmp}_i}\left(grp_i \times x_{[1:3]i}\right) + \beta_{ssn_i} x_{4_i} + \beta_{gdr_i} x_{5_i} + \beta_{t_i} x_{6_i}, \sigma_y^2\right), \; for\ i = 1, \ldots n$$

where

$$x_{[1:3]i} = \begin{pmatrix} SB_1 & LPA_1 & MVPA_1 \\ SB_2 & LPA_2 & MVPA_2 \\ \ldots & \ldots & \ldots \\ \ldots & \ldots & \ldots \\ SB_n & LPA_n & MVPA_n \end{pmatrix}$$

denotes the matrix of the composition of the three activity behaviors, and **Table 1** presents the prior distributions of parameters in the cortisol and cfDNA models, respectively.

Furthermore, we applied a different set of priors for the respective cortisol and cfDNA models, which is justified to

TABLE 1 | The prior distribution of parameters for Bayesian hierarchical linear models.

Cortisol (BHLM 1 and 3)	cfDNA (BHLM 2 and 4)
$\alpha_{id_j} \sim N(0, \sigma_\alpha^2)$	$\alpha_{id_j} \sim N(0, \sigma_\alpha^2)$
$\beta_{cmp_j} \sim N(\mu_\alpha, 1)$	$\beta_{cmp_j} \sim N(\mu_\alpha, 1)$
$\mu_\alpha \sim N(0, 5)$	$\mu_\alpha \sim N(0, 1^{-6})$
$\beta_{tj} \sim N(0, 5)$	$\beta_{tj} \sim N(0, 1^{-6})$
$\beta_{gdr_j} \sim N(0, 5)$	$\beta_{gdr_j} \sim N(0, 1^{-6})$
$\beta_{ssnj} \sim N(0, 5)$	$\beta_{ssnj} \sim N(0, 1^{-6})$
$\sigma_y^2 \sim \mathcal{HC}(0, 5)$	$\sigma_y^2 \sim \mathcal{HC}(0, 25)$
$\sigma_\alpha^2 \sim \mathcal{HC}(0, 5)$	$\sigma_\alpha^2 \sim \mathcal{HC}(0, 25)$

id, identification of participants; cmp, composition; t, time point (midmorning, noon); gdr, gender (female; male); ssn, season (fall; spring; summer).

(a) address the well-established high-variance cortisol displays (within subjects over the course of the day with higher variance later in the day, within subjects at different seasons, and between subjects and gender) and (b) as to the best of our knowledge nothing is known about children's cfDNA levels in the saliva with respect to the daytime, season, gender, SB, and PA. In this study, we allowed random intercepts (α) for each id, and put a hyper prior to α centered to zero (i.e., inform the prior from the data). In addition, we centered βcmp on μα to tie the slope parameter βcmp to the random intercepts (equivalent to nesting ids in the groups); this is called "alternative hierarchical centering" and is an elegant way to borrow strength (i.e., statistical power) from an individual intercept and group. Putting this prior information on the composition dissolves the problem of collinearity, which is typically addressed in ilr- or clr-transformations, however without changing the scale of the output. Thus, the estimates could be interpreted straightforwardly. Finally, other priors were set to be normally distributed parameters around zero, with vaguely informed standard deviation for cortisol and super-vague informed standard deviation for cfDNA. Hence, the cfDNA model should be considered as a strictly provisional "reference model" (76).

In our analysis, we used log-transformed cortisol and cfDNA measures because of skewness and kurtosis (cf. **Supplementary Table 1** and **Supplementary Figures 3–8**). Furthermore, the Markov chains were set to 50,000 iterations, a burn-in phase of 25,000, and a thinning-rate of 10 were applied.

RESULTS

We fitted different BHLMs to assess the possible impact of the relative amounts of SB, LPA, and MVPA on students' cortisol and cfDNA levels. The BHLMs for cortisol and cfDNA differed between the applied indices PR and AUCi. In the BHLM 2 and 4 (AUCi values), the covariates group (CG/IG), gender (female/male) and season (fall/spring/summer) were included. In the BHLM 1 and 3 (PR values), the covariate time point (midmorning/noon) was additionally included. In addition, we evaluated the model fit by means of the deviance information criterion (DIC). The convergence of the Markov chains were investigated by posterior predictive checks (cf. **Supplementary Figures 7–10** for details; only the results for the respective best fitting BHLMs are presented).

Supplementary Figures 1–6 show the descriptive statistics for the variables cortisol and cfDNA, separated for the overall mean values, PR and AUCi and split by season and group. A stronger decrease of the cortisol values from 08:30 a.m. to 12:30 p.m. can be observed in IG compared to the CG, especially in the season's spring and summer. The cfDNA values show different patterns across the seasons and between the groups, which does not allow for a clear tendency. The values in fall and summer are higher compared to spring in both groups. Furthermore, only in the summer season the values of the CG are clearly higher compared to the IG. No clear correlations were found for cortisol and cfDNA with respect to group and season (cf. **Supplementary Figure 11**).

Supplementary Table 2 shows the descriptive statistics for the variables SB, LPA, and MVPA, separated for seasons and groups. We observed no evident differences between the arithmetic mean and the compositional mean in this study. Most apparent differences were observed in higher relative means of SB for the CG compared to the IG and lower relative means of MVPA for the CG compared to the IG. We neither observed any evident differences in seasons and the relative means of LPA.

Association Between Cortisol PR/AUCi and SB/PA

According to the Markov chain Monte Carlo (MCMC) posterior distributions (cf. **Figure 1** and **Table 2** for summary and **Supplementary Tables 3–4** for details), we observed a strong negative association in the IG for the relative amounts of LPA on the cortisol PR levels (posterior mean = −0.728; lower 95% credible interval [CRI]: −1.268; upper CRI: −0.190). In the CG, tendencies of a negative association were noted between SB and MVPA in the cortisol PR. Regarding cortisol AUCi, we observed the likelihood of a negative association in the IG for LPA and for a negative association in the CG for SB. Considering both posterior mean values and CIs, the IG exhibited stronger associations compared to the CG.

Association Between cfDNA PR/AUCi and SB/PA

Compared with the cortisol PR and AUCi, the results of the cfDNA PR and AUCi were different. In the IG, we observed the likelihood of a positive association between SB and MVPA in the cfDNA PR. In fact, a strong positive association was noted in the IG for SB (posterior mean, 1.285; lower CRI: 0.390; upper CRI: 2.191) and tendencies of a positive association of LPA and MVPA in the cfDNA AUCi. In the CG, tendencies of a positive association were found for LPA and MVPA in the cfDNA AUCi (cf. **Figure 1** and **Table 2** for summary and **Supplementary Tables 5–6** for details).

DISCUSSION

General Observations

We conducted the present study to provide an update on the associations between students' cortisol levels and their physical activity as reflected in the measures of SB, LPA and MVPA as

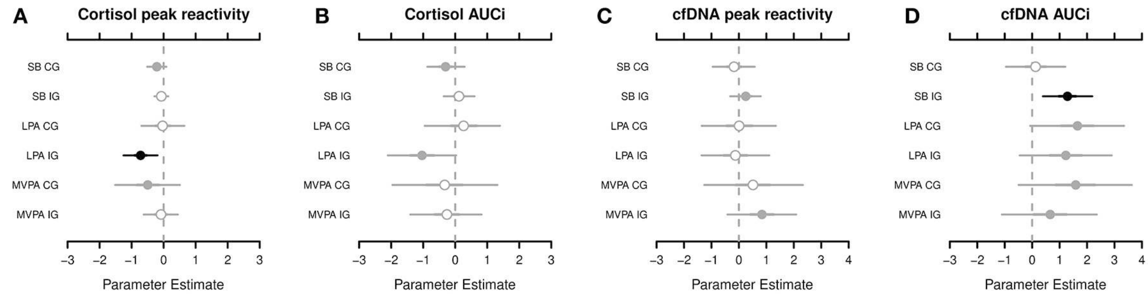

FIGURE 1 | The highest probability density for associations among the cortisol peak reactivity **(A)**, cortisol area under the curve with respect to increase (AUCi) **(B)**, circulating cell-free deoxyribonucleic acid (cfDNA) peak reactivity **(C)**, and cfDNA AUCi **(D)**, respectively, and sedentary behavior (SB), light physical activity (LPA), and moderate-to-vigorous physical activity (MVPA), segregated by groups (CG, control group; IG, intervention group). Shading, whether 50% of credible interval (CRI; gray with open circle), 95% CRI (gray with closed circle), or neither (black) do overlap 0. Black dots indicate of a strong association between the dependent and independend variable. For example, cortisol peak reactivity and LPA in the IG—the adverse effect of the relative amounts of LPA on the cortisol peak reactivity applies to, at least, 95% of children in the posterior distribution.

TABLE 2 | The MCMC output of posterior probabilities.

Variable	Model I	PM	SD	CRI 2.5%	CRI 25%	CRI 50%	CRI 75%	CRI 97.5%	Rhat	ESS
SB (CG)	BHLM 1: cortisol PR	−0.215	0.152	−0.512	**−0.318**	**−0.213**	**−0.112**	0.085	1.001	7,500
LPA (IG)	BHLM 1: cortisol PR	−0.728	0.271	**−1.268**	**−0.908**	**−0.726**	**−0.551**	**−0.190**	1.001	7,500
MVPA (CG)	BHLM 1: cortisol PR	−0.499	0.524	−1.517	**−0.860**	**−0.501**	**−0.141**	0.527	1.001	5,900
SB (CG)	BHLM 2: cortisol AUCi	−0.293	0.298	−0.880	**−0.494**	**−0.291**	**−0.095**	0.297	1.001	7,100
LPA (IG)	BHLM 2: cortisol AUCi	−1.027	0.550	−2.112	**−1.403**	**−1.030**	**−0.661**	0.062	1.001	6,700
SB (IG)	BHLM 3: cfDNA PR	0.242	0.289	−0.329	**0.049**	**0.246**	**0.435**	0.801	1.001	5,400
MVPA (IG)	BHLM 3: cfDNA PR	0.839	0.636	−0.418	**0.416**	**0.839**	**1.269**	2.088	1.001	7,500
SB (IG)	BHLM 4: cfDNA AUCi	1.285	0.464	**0.390**	**0.970**	**1.286**	**1.595**	**2.191**	1.001	7,500
LPA (CG)	BHLM 4: cfDNA AUCi	1.643	0.877	−0.072	**1.058**	**1.652**	**2.232**	3.348	1.001	4,300
LPA (IG)	BHLM 4: cfDNA AUCi	1.231	0.858	−0.455	**0.647**	**1.227**	**1.804**	2.899	1.001	5,300
MVPA (CG)	BHLM 4: cfDNA AUCi	1.574	1.053	−0.492	**0.853**	**1.588**	**2.294**	3.632	1.001	3,600
MVPA (IG)	BHLM 4: cfDNA AUCi	0.649	0.889	−1.102	**0.053**	**0.658**	**1.251**	2.356	1.001	7,500

BHLMs 1–4: variables are presented if lower 25% CRI and upper 75% CRI do not overlap 0; the respective values that do not overlap zero are bold. MCMC, Markov chain Monte Carlo; BLHM, Bayesian hierarchical linear model; SB, sedentary behavior; LPA, light physical activity; MVPA, moderate-to-vigorous physical activity; CG, control group; cfDNA, circulating cell-free deoxyribonucleic acid; IG, intervention group; PR, peak reactivity; AUCi, area under the curve with respect to increase; PM, posterior mean; SD, standard deviation; CRI, credible interval; Rhat, potential scale reduction factor; ESS, effective sample size; the ESS of the posterior distribution differs in relation to the convergence of the MCMC algorithms; the ESS depends on how accurately the proposed model fits the data.

well as associations between students' cfDNA levels and their SB, LPA, and MVPA in outdoor and indoor classroom environments. While interpreting the results of this study, one must consider the character of this exploratory study: the specific school setting in which both (a) the number of available participants is low because of the situation of EOtC in Germany, and (b) the number of possibly uncontrolled confounders is high because of the real-world scenario. However, we believe that our study can provide valuable insights into the EOtC research, health promotion in schools, and the assessment and analysis of cortisol, cfDNA, SB, and PA in the educational setting. In our previous study (12), we reported a statistically significant difference in the measured cortisol levels between the CG and IG; regular teaching in the forest correlated with a lower cortisol secretion at noon compared with the standard indoor teaching, and this association was independent of students' MVPA levels. Considering the compositional nature and, thus, the codependency of students' SB, LPA, and MVPA, we elucidated students' cortisol values during school time in this study. Furthermore, we compared those results with associations between students' cfDNA levels and their SB and PA.

According to the presented posterior distributions of the four BHLMs, the associations between students' cortisol/cfDNA levels and their compositional amount of SB, LPA, and MVPA are diverse. Furthermore, the presented effects with respect to posterior means and credible intervals must be considered as small. First, we could partially confirm our previously reported results (12) reflecting their independence of the analysis methodology, as students' cortisol levels were not affected by the relative amounts of MVPA in the IG. However, in the CG, the relative amount of MVPA is more likely to exert a lowering effect on the cortisol PR; the more active the students were in MVPA levels, the more their cortisol levels seemed to decrease. Two experimental studies (22, 23) reported that human behaviors similar to SB and LPA correlated with declining cortisol levels, which is in concordance with a typical healthy diurnal rhythm. The lowering effect of LPA on cortisol in the IG therefore corroborates Hill et al. (22) and VanBruggen et al. (23), although the specific PA intensities are not directly comparable. Thus, it could be argued that the so-called "green effect" (12, 77) in the forest (positive effects of the NGE on humans' psychological well-being) supports the lowering physiological effect of relatively high LPA levels of cortisol to some extent. Perhaps, this supportive effect could be missing during the regular indoor teaching because of the built environment. The association between SB and cortisol is more likely for the CG but not for the IG. Of note, uncomfortable sitting situations in the forest could result in psychological stress in terms of discomfort or inability to concentrate, and, therefore, potentially be attributed to this missing association in the IG. The validation study by Khoury et al. (62) reported that the PR and AUCi indices exhibit similar results regarding the cortisol increase/decrease. In this study, most associations of SB/LPA/MVPA in cortisol PR/cortisol AUCi, respectively, exhibited similar tendencies; only the tendency for a negative association of MVPA in the CG was not present for the AUCi index. Thus, we assume that our cortisol dataset based on the measurement procedure with three time points (08:30 a.m., 10:30 a.m., and 12:30 p.m.) is not entirely comparable with the time points used previously (62).

Some studies have reported that cfDNA levels already increase with moderate PA below the level of the aerobic–anaerobic transition (29, 33). The results of the cfDNA AUCi posterior distributions suggest that such an association is also more likely in both teaching settings. However, the relative amount of SB in the IG also exhibits a strong positive association with students' cfDNA values, which, perhaps, cannot be easily explained on a theoretical or empirical basis. Furthermore, the deviance values in both cfDNA PR/AUCi analyses are rather high compared with the respective cortisol values (cf. **Supplementary Tables 3–6**), which is an indication that the cfDNA MCMCs present a worse convergence compared with the cortisol MCMCs. Regarding the cfDNA, the log-likelihood is lower, and the data deviate more substantially from the models assumptions compared with cortisol. Thus, a strong positive association of SB in students' cfDNA could be likely attributed to an overestimation in the model. Regarding cfDNA results, similar tendencies have been observed between the applied PR and AUCi indices for SB and MVPA in the IG. Both indices, PR and AUCi, seem to be stable for cortisol, whereas the results for the cfDNA are more diverse regarding the PR and AUCi; this could be potentially explained by the factor "time point." As one's cortisol secretion follows a time-dependent diurnal rhythm (with expected high values in the morning and a steady decline from the noon to the evening), both validated indices account for the variation of cortisol over the period of the school day. Regarding the cfDNA, the AUCi index seems to better account for the less time-dependent and more PA-related secretion, which could be illustrated with nearly two times as much deviance for the cfDNA PR compared with the AUCi (cf. **Supplementary Tables 5–6**). In general, our analysis with the applied PR and AUCi indices was optimized for cortisol with its time-dependent diurnal rhythm and is therefore more appropriate to be used for cortisol compared with cfDNA.

Owing to the underlying pedagogical concept in the forest (60), students might have more breaks between phases of SB, LPA, and MVPA. Recent research has reported about positive health effects of breaks during extended periods of SB (78–81). The possible relevance of the number of interruptions is an interesting phenomenon to be evaluated in future research concerning students' SB and PA in school. According to the pedagogical concept of the present intervention (60), it could be hypothesized that students in the forest have more freedom to choose whether they want to sit, walk, or run. During the normal indoor teaching, students frequently have to sit still for the entire 45 min in each lesson. Perhaps, the hypothesized freedom of choice could result in better physiological reactions within students' adaptive systems. These aspects warrant further investigations. In future large-scale, prospective studies, also aspects of the sunlight exposure (82, 83) and further possible confounders should be considered.

Limitations and Future Directions

This pilot study has an exploratory character that is especially based on the relatively low number of available participants along with the mentioned high number of uncertainties in children's cortisol and cfDNA levels in relation to their SB and PA in indoor and outdoor teaching. Because of non-existing previous research in this field, a meaningful sample size calculation to determine desirable statistical power was not possible before data collection. The promising approach of retrospective design calculation (84) to inform the interpretation of gained results should be considered in general. However, this approach was not applicable in our study because of the lack of reliable external information regarding the real effect sizes. Moreover, "without relatively large sample sizes we are often precluded from saying anything precise about the size of the effect because the likelihood function is not very peaked in small samples" [(85), p. 63]. Therefore, with the results of this pilot study, gained with Bayesian inference, we do not aim to generalize our findings but rather inform prospective studies. In concordance with our previous analysis (12), three measurement points over 1 school-year provided only limited insight into the complex structure of regular compulsory outdoor lessons, students' levels of measured biological stress parameters, and the respective associations with their SB and PA. However, conducting more measurement days was not feasible for logistical and school organizational reasons. In addition, several acceleration sensors fell off because of warm weather conditions during the study time point summer, which resulted in the loss of PA data. Furthermore, we were not able to conduct a long-term cortisol analysis, as collected hair samples could not be analyzed [discussed in (12)]. Therefore, we were restricted to the measurements of saliva cortisol during the three time points at 8:30 a.m., 10:30 a.m., and 12:30 p.m. and need to assume that the salivary cortisol as an HPA axis biomarker reflects psychological and physiological induced stress with sufficient validity (24, 86). Furthermore, the allocation of students to the CG and IG was performed per the school policies and parents' choice. Thus, students could not be randomly allocated to a group by the experimenter, possibly implying certain bias. Hence, the overall small number of participants in this pilot study must be considered, and extensive, prospective studies are warranted to investigate further the tendencies explained in this study. In addition to cortisol and cfDNA the measurement of IGs should be considered in normal indoor settings. Furthermore, questionnaire-based assessment of students' mental well-being needs consideration.

CONCLUSIONS

The most important finding is that despite little difference in LPA between the CG and the IG, relative long time spent in LPA in an outdoor teaching setting seems to be strongly associated with a decline of cortisol levels, whereas no such decrease can be observed in the indoor setting. This is of great importance for educational practice, as the combination of PA and the outdoor environment during EOtC seems to be beneficial for students stress response. Additionally, the observed relative amount of sedentary time is lower and that of MVPA higher in the outdoor teaching setting in a NGE compared with the indoor setting. That implies possible health benefits for students during EOtC. Additionally, a more clinically controlled study will elucidate children's cfDNA values in relation to SB and PA, as well as to cortisol. Furthermore, in future research concerning students' PA and health in school, the co-dependency of SB, LPA and MVPA should be taken into account. Future studies on EOtC can build on the gained knowledge to apply informed priors.

DATA AVAILABILITY

The datasets for this study can be found in the **Supplementary Material**.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of and was approved by the ethics committee of the Medical Faculty Mannheim, University of Heidelberg, Germany. The approval code is 2014-585N-MA. All subjects gave written informed consent in accordance with the Declaration of Helsinki.

AUTHOR CONTRIBUTIONS

PK, PS, and UD conceived and designed the study. CB collected the data. PS, SS, and EN developed tools to prepare the cfDNA-salvia probes which were statistically analyzed by CB and UD. CB wrote the paper with substantial contributions from all other authors. All authors proved the final version of the manuscript.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fpubh.2019.00026/full#supplementary-material

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