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School lunch and short-term effects on children's cognitive functions – Results from randomized crossover intervention studies

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Summary

Studies indicate that eating lunch temporarily impairs some aspects of adults' cognitive functioning. Studies of the short-term effects of lunch on child cognition are rare. This thesis provides the results of two randomized crossover intervention studies, which provide an initial insight into this topic. The Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) and the Cognition Intervention Study Dortmund Continued (CoCo) examined the short-term effects of school lunch on the cognitive performance of children during afternoon lessons.

CogniDo PLUS (n=215) investigated the short-term effects of school-lunch on children's executive functions (EF) in the early afternoon (45 minutes after finishing lunch) and whether the postprandial cortisol increase mediates putative lunch effects on EF performance. The cognitive parameters task switching, updating working memory, and inhibition were tested using a computerized test battery. Saliva samples were used to assess cortisol directly before lunch and again at the beginning of the cognitive assessment after lunch. The results show that school lunch does not impair children's EF under real-life conditions. The study even indicates beneficial effects of school lunch intake after 45 minutes for the working memory updating. The postprandial cortisol increase in the range observed in CogniDo PLUS does not seem to be related with negative effects on the performance of EF, but even seem to mediate the beneficial effect of lunch on the working memory updating. **CoCo** (n=154) investigated the hypothesis of potential positive effects of school lunch on cognitive performance in the afternoon (90 minutes after finishing lunch). The measured parameters were task switching, updating working memory, and alertness. The data suggests that school lunch does not seem to have beneficial or detrimental effects on children's cognitive functions in regard to the tests conducted in the early afternoon, since no significant results were shown after 90 minutes after finishing lunch.

In conclusion, this thesis offers first insights into the short-term effects of school lunch on children's cognitive performance in the afternoon. In contrast to findings in adults, the results indicate that children's cognitive performance respective the measured parameters are not impaired by lunch under real-life conditions. The postprandial cortisol increase in the range observed in our sample does not seem to be related with negative effects on EF, but even seems to mediate the beneficial effect of lunch on the working memory updating. However, beneficial effects regarding working memory updating seem to be restricted to a relatively short period of time after eating lunch (i.e. 45 minutes).

Zusammenfassung

Studien an Erwachsenen liefern Hinweise, dass das Mittagessen ausgewählte Parameter der kognitiven Leistungsfähigkeit kurzfristig negativ beeinflusst. Studien, in denen die kurzfristigen Effekte des Mittagessens in der Kindheit untersucht wurden, sind kaum existent. Diese Doktorarbeit stellt Ergebnisse zweier randomisierter Crossover Interventionsstudien dar, die erste Einblicke in diese Thematik geben. In den Studien "Cognition Intervention Study Dortmund PLUS" (CogniDo PLUS) und "Cognition Intervention Study Dortmund Continued" (CoCo) wurden die kurzfristigen Einflüsse eines Schulmittagessens auf die kognitive Leistungsfähigkeit der Schüler im Nachmittagsunterricht untersucht.

In der **CogniDo PLUS** (n=215) Studie wurden kurzfristige Einflüsse des Schulmittagessens auf exekutive Funktionen (EF) von Kindern am frühen Nachmittag (45 Minuten nach dem Essen) untersucht. Zudem wurde getestet, ob ein postprandialer Cortisolanstieg potentielle Effekte des Mittagessens vermittelt. Die Parameter Aufgabenwechsel, Aktualisierung des Arbeitsgedächtnisses und Inhibition wurden mittels einer computerbasierten Testbatterie erfasst. Speichelproben zur Messung des Cortisols wurden direkt vor dem Essen und den kognitiven Tests abgegeben. Die Ergebnisse geben Hinweise darauf, dass die EF bei Kindern, (unter realen Bedingungen) nicht durch das Mittagessen beeinträchtigt werden. Es zeigte sich sogar ein positiver Effekt in der Aktualisierung des Arbeitsgedächtnisses 45 Minuten nach dem Essen. Der gemessene postprandiale Cortisolanstieg hatte keine negativen Auswirkungen auf die EF. Dieser scheint bei Kindern sogar eine vermittelnde Rolle bei der Verbesserung der Aktualisierung des Arbeitsgedächtnisses zu spielen.

Die **CoCo** Studie (n=154) untersuchte die Hypothese, dass das Mittagessen positive Effekte auf die kognitive Leistungsfähigkeit bei Kindern im Verlauf des Nachmittags hat (90 Minuten nach dem Essen). Dazu wurden die kognitiven Parameter Aufgabenwechsel, Aktualisierung des Arbeitsgedächtnisses und Alertness gemessen. Die Daten zeigen weder einen positiven noch einen negativen Einfluss des Essens auf die gemessenen Parameter nach 90 Minuten.

Insgesamt liefert diese Arbeit erste Einblicke in die kurzfristigen Effekte einer Mittagsmahlzeit auf die kognitive Leistungsfähigkeit bei Kindern am Nachmittag. Im Gegensatz zu Erwachsenen deuten die unter realen Bedingungen gemessenen Ergebnisse – bezogen auf die erhobenen Parameter – keine Verschlechterung der kognitiven Leistung bei Kindern durch das Mittagessen an. Der gemessene postprandiale Cortisolanstieg scheint nicht mit negativen Effekten in Verbindung zu stehen, sondern im Gegenteil den positiven Effekt des Essens auf die Aktualisierung des Arbeitsgedächtnisses zu vermitteln. Allerdings gelten diese positiven Effekte des Essens nur für eine kurze Zeitspanne nach dem Essen (d.h. 45 Minuten).

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Abbreviations

CF	Cognitive functions
CoCo	Cognition Intervention Study Dortmund Continued
CogniDo	Cognition Intervention Study Dortmund
CogniDo PLUS	Cognition Intervention Study Dortmund PLUS
EF	Executive functions
GABA	Gamma-aminobutyric acid
GI	Glycemic Index
IQ	Intelligence Quotient
IQR	Inter Quartile Range
ISI	Interstimulus interval
L	Lunch
LNAA	Large neutral amino acids
PET	Positron emission tomography
NL	No lunch
RQ	Research question
RT	Reaction time
RSI	Response stimulus interval
TMT	Trail Making Task

1 General Introduction

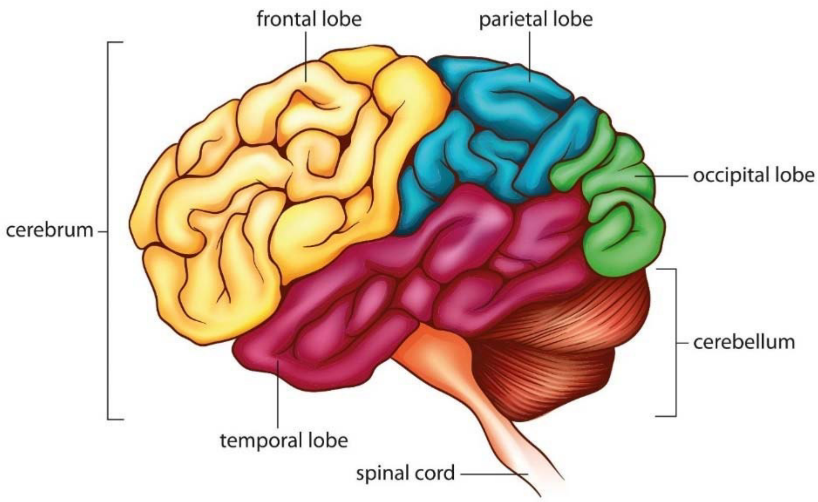
1.1 Overview

1.1.1 Brain development and cognitive functions

The brain is one of the most complex organs in the human body. It undergoes substantive changes in structure and functional organization in the span of a human lifetime [1]. Most of the brain development takes place prenatally, and within the first two years after birth the crucial changes occur [2]. Although the brain has reached 80 % of its adult mass after 2 years, it continues to develop in particular concerning the myelination of neurons [3]. At the age of 6 years a child's brain mass has almost reached its adult brain mass (95 %). A growing brain is metabolically highly active. Until around the fourth year of age, the premature brain tissue utilizes 4 times as much glucose as that of an adult. Glucose utilization remains at a high rate until an age of 9 to 10 years and then declines to the values of an adult during adolescence [3].

The different regions of the brain do not mature uniformly. For example, regions associated with more basic functions such as sensory and motor skills develop earlier than regions like the frontal lobes that are involved in the top-down control of behavior [1]. The frontal lobes (Figure 1), which include the prefrontal lobes, are the area that is slowest to fully myelinate. Myelination begins at approximately 6 months and continues throughout childhood and adolescence into adulthood. Spurts in the development of the frontal lobes have been found to occur in the stages from birth to 2 years, 7 to 9 years, and in the mid-teens [4].
Specifically, during the preadolescent period, neuronal rearrangements occur with an overproduction of synapses and receptors then followed by their reduction or competitive elimination. Between roughly 4 and 12 years of age, synaptic density in the frontal lobe decreases by approximately 40 % [5]. The grey matter of the frontal lobe area continues to thicken as part of a process that peaks around puberty [3]. For instance, frontal lobe gray matter reaches its maximum volume at 11 years in girls and 12 years in boys [6]. It is assumed that the maturation of higher cognitive functions occurs coincidentally with a decrease in the number of existing synapses and not with the formation of new synapses, and finally the remaining synaptic connections are straightened. The late maturing areas of the prefrontal cortex are principally involved in higher executive functions [5].

Figure 1: Different areas of the crerbral cortex [7]



1.1.2 Alertness

Alertness is part of attention, a basal cognitive function, which is the basis of many higher functions e.g. inhibition control. Attention can be divided into two domains: an intensity domain i.e. alertness and sustained attention, and a selection domain i.e. focused and divided attention. Alertness again can be subdivided into intrinsic (or tonic) alertness and phasic alertness. Intrinsic alertness is defined as the internal control of arousal in the absence of an external cue [8], whereas phasic alertness reflects the arousal and increased attention activated by an extrinsic stimulus e.g. acoustic sound [9]. Intrinsic alertness can be measured by simple reaction time tasks without preceding warning stimuli. In adolescents alertness has been suggested to be further developed than complex attention functions i.e. selective attention [10].

1.1.3 Executive Functions

Executive functions (EF; also known as executive control or cognitive control) are located in the prefrontal lobes. EF are often termed as 'higher' functions [11] including i.e. mental flexibility, goal directed behavior, the ability to filter disturbances, and calculating the consequences of actions [12]. There is a general agreement that three core EF can be defined [13]: shifting between tasks or mental sets (task shifting), working memory representation updating and monitoring (working memory updating), and inhibition of dominant or prepotent responses (inhibition) [11]. Miyake et al. conducted studies that showed that these three constructs of executive functions are clearly distinguishable, but have underlying similarities [11].

'Task switching'

Task switching (also known as attention switching) is the ability to shift back and forth between tasks or mental sets. It gives the opportunity to react flexibly to new situations and is considered to be an important part of executive control [11,14]. Continuously repeating the same response even when it is no longer appropriate signals pathologic difficulties in shifting mental sets [11]. Shifting between mental sets requires temporal costs, which can be measured with reaction time tasks.

'Updating'

Updating is closely linked to the monitoring of working memory but goes further than the simple maintenance of task-relevant information [11,15]. Updating is the ability to select and update relevant information and to replace old, no longer relevant information with newer, more relevant information [16]. The essence of updating lies in the active manipulation of relevant information in working memory, rather than the passive storage of information.

'Inhibition'

Inhibition (or inhibitory control) reflects one's ability to volitionally inhibit predominant or automatic responses when necessary and suppress habitual, thoughtless reactions. It reflects self-control and enables choosing how to react [11,13].

Altogether these core EF are the basis of superior functions such as reasoning, problem solving, and planning [13]. They are crucial for mental and physical health, cognitive, social and psychological development, and success in school. Consequently, EF are most important for daily performance during school lessons. They are necessary for retaining and transforming short-term memory contents as well as for controlling actions and planning action sequences [17]. EF are even suggested to be more important for school than IQ level [18].

It is known that children's and adolescents' EF are not fully developed [19,20] and decay earlier than low-level functions with increasing age [21]. The maturation of the cerebral cortex, which includes the frontal lobes, continues until late adolescence and is characterized by dynamic changes in metabolism. For example, a child's brain is more dependent on the supply of glucose than the brain of an adult [22]. Therefore, it has been hypothesized that the developing EF might be more sensitive to environmental influences, such as nutrient intake, than the EF in a fully developed adult brain [22,23]. Also, aging research shows that environmental factors, such as physical exercise, influence EF [24].

1.2 Lunch and cognition: short-term effects

1.2.1 Overview

Most studies of the short-term effects of lunch on cognitive functions have focused on adults, but overall few studies have been done and the results are not fully consistent. For example, Craig et al.[25] compared two situations, one in which lunch was consumed and one in which the participants abstained from lunch. Their results showed that the cognitive ability to discriminate between events was significantly impaired following the consumption of lunch, but it did not improve when no food was ingested [25]. In another study Smith & Miles [26] found that the test-lunch impaired their study participants' sustained attention [26], whereas abstaining from lunch had no influence on sustained attention when compared to the pre-lunch condition. In contrast, Kanarek & Swinney [27] even observed positive effects of lunch consumption (compared to no lunch) when the participants were faced with a reading task [27].

Beside from this general effects of eating lunch, studies have examined the influence of lunch size on adults' cognitive performance. It has been observed that a larger lunch than usual increased the error rate in sustained attention tasks in comparison to the error rate after the consumption of a smaller lunch [28,29].

Collectively the existing studies suggest that lunch consumption could impair some of adults' cognitive functions [27,30] and that the lunch size might influence the study results [25].

1.2.2 Studies on meal composition

Carbohydrates

Carbohydrates have been the most commonly investigated dietary component in the context of studies on cognition up to now. Most studies have examined the impacts of an ingested glucose drink on performance in cognitive tests [31–34]. For example, Owen et al. [31] demonstrated that a glucose dose of 25 g following a 2-h fast enhanced working memory performance, and Benton et al. [34] showed that the consumption of a drink with 25 g glucose improved sustained attention [31,34]. However, tests with pure glucose cannot be compared to tests on the effects of a whole meal because sugars or other carbohydrates as part of a mixed meal increase blood glucose levels more slowly than pure glucose [35,36]. Furthermore, the insulin response is different [37].

Recently, the impact of the glycemic index (GI) of a meal on cognitive performance was discussed in a review [38]. The authors cautiously suggested that a low-GI meal may positively influence adults' cognitive functions, but the authors noted that their findings were inconclusive due to differences in study design, study sample (e.g. size, age), time of testing, and the cognitive domain examined [38].

Fat

Smith et al. [39] conducted a study of the impact of a lunch meal's fat content on cognitive performance within 60 minutes after ingestion. They observed that participants who ate a high-fat test lunch responded more accurately but more slowly during attention tasks than the participants who consumed the low-fat lunch. However, these effects were minimal, and conclusions could only be drawn for the attention tasks conducted in the particular study. The authors critically discussed that changes in fat content resulted in different energetic conditions and may interfere with changes of carbohydrate and/or protein content. However, they concluded that their results based on the fat content change seem to be reliable because they obtained different results in an earlier study in which they modified the proportions of carbohydrates and protein. However, a different study confirmed the results showing a decline in alertness (speed and accuracy) after the consumption of a high-fat lunch compared to an isocaloric low-fat lunch [40]. In contrary, results found by Kaplan et al. [41] indicate that a pure fat drink could enhance the attention function in the case of elderly adults. However, the authors did not define which dimension of attention was tested. Additionally, they tested the subjects only 15 minutes after ingestion and this short period of time precedes fat absorption. The authors speculated that an activation of the gut-brain axis probably plays an important role.

Protein

Studies on the effects of protein in meals come to inconclusive results. Diets rich in protein have been associated with decreased positive cognitive effects and increased negative cognitive effects relative to those of carbohydrate-rich diets [42]. Smith et al. [43] found that study participants who consumed a high-protein lunch were more prone to distractions from the target during a focused-attention task in comparison to the participants who consumed a high-carbohydrate lunch [43]. When comparing the effects of protein-rich and carbohydrate-rich snacks on mood and performance, results were diverse and depended on age, gender, and the time of snacking [44]. Study-meals with a high amount of protein still contain carbohydrates, so it is challenging to distinguish if it is the high protein content, the low carbohydrate content, or the energy intake that has an effect. Therefore, Kaplan et al. [41] conducted a study in which they tested the effects of isoenergetic, pure carbohydrate, fat, and protein drinks on healthy elderly adults. The protein drink reduced the subjects' rate of forgetting on a recalling task. However, all three drinks improved memory function, and the authors concluded that the ingestion of energy appears to improve memory, regardless of its source.

1.2.3 Studies in children

Due to the constitutional and metabolic differences between children and adults, results shown in studies of adults are not necessarily transferable to the case of children. Because the brain is the metabolically most active organ in the body it needs to be continually supplied with glucose [3] and due to the rapid growth and the high metabolic rate it can be speculated that a meal has short-term effects particularly on the cognitive functions of children and adolescents. In addition, the acute effects of a meal on performance may vary with the time of day and nutritional status of the person who consumes it [40,42].

Research on meals and child cognition has mainly focused on long-term effects. Until now studies of the short-term effects of meals on children have been almost exclusively focused on breakfast. The main reason for this is the assumption that after an overnight fast glucose reserves are almost depleted and need to be replaced. Indeed, findings from earlier studies suggest that breakfast consumption is positively associated with better cognitive performance, eventhough the conducted studies do not allow drawing firm conclusions [45,46]. Poorly nourished children benefitted more from breakfast than well-nourished children [47]. Also, the effects of breakfast composition, in particular the glycemic index (GI), have been recently examined. Cooper et al. [48] concluded that a low-GI breakfast is more beneficial for adolescents' cognitive performance than a high-GI breakfast or skipping breakfast.

In contrast to breakfast, the effects of lunch on short-term cognitive performance in childhood have been neglected in research up to now [30]. The CogniDo [49] study in Germany was the first study to provide an insight into this topic. The randomized crossover study was conducted to examine the short-term effects of having lunch or skipping lunch on children's basic cognitive functions i.e. tonic alertness, selective attention and visuospatial memory. No short-term effects of lunch on sustained attention or visuospatial memory were found, but there was a significant effect on tonic alertness, in that omission errors were more frequent after skipping lunch as compared to after lunch consumption. It has not yet been examined if and how lunch affects children's EF shortly after ingestion.

1.3 Possible short-term cognitive mechanisms of lunch

1.3.1 Glucose and glycemic load

As mentioned earlier carbohydrates have been the most studied potentially influencing factor in the context of short-term effects on cognitive performance, since glucose is the primary source of energy for the brain. However, results of studies on the glycemic load of meals show inconsistent results, hence the theory that glucose has a significant effect on cognitive performance via the glycemic load lack evidence at present. It is argued that the brain is not completely permeable to blood glucose, but rather the brain's uptake of glucose is compartmentalized and controlled by local demand [50,51].

Although the evidence is not consistent, a number of studies have reported beneficial effects of glucose on cognitive performance. In particular studies which investigated glucose uptake show that an increase of blood glucose levels enhances cognitive functions, such as memory [31,52], or sustained attention [34]. For example, Owen et al. [31] demonstrated that consumption of a glucose dose of 25 g enhanced working memory performance after a 2-h fast and Benton [34] et al. showed that a drink containing 25 g glucose improved sustained attention. However, the effects of oral glucose dosage may differ depending on individual blood glucose resources or the level of depletion [31]. Even though positron emission tomography (PET) scans revealed that increased cognitive functioning is associated with a rapid uptake of glucose from the blood into the brain, it is not clear if the decrease in blood glucose (and increased cognitive performance) is associated with glucose uptake by the brain or in the peripheral tissue [52]. One hypothesis is that glucose provides energy to neurons either by direct transfer from blood via extracellular fluid or by an intermediate transfer through astrocytes [53]. Taken up by astrocytes glucose is converted via glycolysis into lactate and then used as an energy substrate by neurons [54]. The fact that astrocytes and neurons exhibit monocarboxylate transporters, which transport for example lactate, gives support for this hypothesis. Additionally, the synthesis of several neurotransmitters is directly dependent on exogenous glucose, for example, the two excitatory neurotransmitters glutamate and acetylcholine or the inhibitory transmitter, gamma-aminobutyric acid (GABA) [53]. Taken together, no clear-cut relationship between glycemic response, brain glucose, and performance measures could yet be established.

1.3.2 Protein and neurotransmitters

Ingested proteins deliver amino acids, which are precursors of the neurotransmitter serotonin, catecholamines, histamine, glycine, and acetylcholine, for example, tyrosine (dopamine), tryptophan (serotonin) or cholin (acetylcholine) [55,56]. The uptake of neurotransmitter precursors across the blood-brain-barrier works via two specific transport systems, one for choline and the other competitive system for large neutral amino acids (LNAA) like tryptophan, tyrosine, threonine, phenylalanine, methionine and the branched-chain amino acids (leucine, isoleucine and valin) [55].

For the uptake of tryptophan through the blood-brain-barrier the ratio of plasma tryptophan and the sum of the (other) competing LNAA concentrations is important [42,55]. After ingestion of a meal insulin stimulates the tissue uptake of LNAA, raising ratio in favor of tryptophan. Tryptophan access into the brain is increased, and the serotonin production is enhanced. Interestingly a high-protein meal is less insulinogenic than a low-protein meal and thus the tryptophan: LNAA ratio is lower, which handicaps the uptake of tryptophan into the brain [42,55]. That an increased amount of precursor amino acid could enhance the production of the related neurotransmitter has been shown in tyrosin as a precursor of dopamine, which is involved in cognitive functions like working memory updating and task switching [57]. In a study where 2 g of tyrosine or placebo (2 g of cellulose) was administered as powder dissolved in orange juice before testing, it was shown that the intake of tyrosine could enhance the working memory as tested by a demanding 2-back task. It was suggested that high plasma tyrosine leads to high amounts of dopamine in the prefrontal cortex and that dopamine is involved in the signal transduction within updating working memory.

The other fact that there are no feedback systems to keep the plasma concentration of precursors in narrow ranges, like for glucose or osmolarity, emphasizes the specific role of dietary supply of precursors for the neurotransmitter synthesis [55].

Other dietary components that potentially play role in the effects of food on brain activity could be vitamins and minerals serving as cofactors of enzymes which are involved in the neurotransmitter synthesis [58].

1.3.3 Post- lunch dip

A discussed phenomenon in the context of impairment in adult cognitive functioning after lunch is the so-called post-lunch dip. It is a naturally occurring dip around midday which is supposedly related to more than one factor, e.g. individual circadian rhythm or the length of time since waking up [59–61]. It has been suggested that the post-lunch dip may only reflect an endogenous rhythm of alertness. Decrements in performance around midday, which have been shown by some tasks regardless of whether or not lunch was eaten, support this assumption [60]. However, in intervention studies it was observed that lunch exacerbates the naturally occurring dip in adults [25,61]. For example, in an investigation on the ability to discriminate between events, the participants who ate lunch showed an impaired discrimination efficiency in contrast to the participants without lunch [25].

The mechanisms behind the phenomenon that lunch seems to worsen the post-lunch dip are not yet fully understood. Alterations in the synthesis of neurotransmitters based on availability of amino acids or postprandial glucose metabolism have been discussed as contributing factors [62]. Additionally, the time of the day at which a meal is ingested seems to have an important influence. For example, having breakfast often shows improving consequences on cognitive performance [47], while having lunch could worsen post-lunch decrements in cognitive performance [25]. A late afternoon snack may counteract these decrements [27].

In the adults studied a post-lunch dip was observed about 60 to 120 minutes after lunch [25,61,63] indicating that the time span between a meal and the measurement of the cognitive performance could have a relevant influence [61]. However, there are no well-controlled studies assessing the duration of the post-lunch dip [60].

Furthermore, lunch size might be an important factor that affects the post-lunch dip. Craig and Richardson [28] investigated the influence of a heavy lunch on individuals who normally eat a light lunch. The results indicated that a heavy lunch consumption resulted in a deteriorated subjective alertness rating [28].

Since the majority of experiments in this subject area used healthy young men as experimental subjects, the potential effects of gender, age, and nutritional status on cognitive performance following lunch are not yet known [60]. Because studies on the post-lunch dip among children have not yet been found in the literature, it remains open, whether or not children experience a post-lunch dip phenomenon that may be exacerbated by a meal as is suggested for adults.

1.3.4 Cortisol

One hypothesis discusses meal-induced postprandial increases of plasma cortisol as an effect of lunch on cognitive functions. It has been observed among adults that an increased cortisol level induced by psychological stress or pharmaceuticals impaired memory performance [64–66]. Lupien et al. [67] ascertained that working memory is more prone to pharmaceutically induced cortisol increase than declarative memory. Based on their results the authors inferred that a low pharmaceutical cortisol dose improves the processing capacity of working memory, whereas a high dose leads to impairments. It has been suggested that the correlation between working memory performance and changes in glucocorticoid levels after hydrocortisone infusion can be shown with an inverse U-shape curve [65,68]. Abercromie et al. [69] found out that 20 or 40 mg cortisol doses given before testing resulted in fewer commission errors i.e. false alarms in young adults. Particularly because the frontal lobes contain glucocorticoid receptors, it is reasonable that cortisol influences cognitive functions [70].

Adrenal cortisol secretion underlies a circadian rhythm with peak plasma concentrations upon waking up and a nadir around midnight [71]. For both adults and children lunch intake induces an increase in cortisol levels [72–76]. However, the threshold of a physiological postprandial increase in cortisol, which could effectively impair the cognitive functions, is not yet known. It remains to be evaluated whether a postprandial increase in cortisol might mediate lunch-induced impairments of cognitive performance in children.

1.4 Assessment of cognitive functions in children

Neuropsychological assessments are typically administered in well-structured and quiet settings with minimal distractions and are thus unlikely to be representative of home, classroom, or social environments [77].

Principally, the selected cognitive outcome measures should be sensitive enough to detect nutritional effects, and should ideally be standardized for administration in the respective culture in which they are used [4]. However, seeing as cognitive functions develop rapidly in children, it is difficult to devise tasks that are suitable across the developmental spectrum. Floor effects (i.e. the test is too difficult for most children) and ceiling effects (i.e. the test is too easy for most children) in test performance reduce the test sensitivity to detect improvements and impairments. Floor and ceiling effects should therefore be avoided by adjusting the task difficulty to the appropriate level for the test participants (i.e. children). Unsuitable tasks and the resulting difficulties may also lead to frustration and loss of motivation [78]. It has been recommended that for children aged 5 to 12 years batteries of tests should take no longer than one hour to complete [4].

Regarding the measurement methods, computerized tests (often developed on basis of paper and pencil tests) have the advantage of a standardized presentation and exact and detailed response measurement. With adequate adaptations they can be used in studies of children without problems [78].

Measuring EF does involve some difficulties. Inconsistencies between performance on EF measured and real life behavior have often been described. Environmental factors, such as noise, ambient temperature, and lighting may influence performance levels [78]. Special care should be taken to exclude or at least standardize these factors as much as possible, in order to limit the variability among study subjects. To prevent possible circadian effects from influencing performance, testing should be conducted at similar times of the day on each occasion. Conducting a test series of tasks that are not included in the analyses before the actual test can help prevent any so-called warm-up effects (temporary poor performance at the beginning of testing) [78].

2 Objectives and Research Question

The overall aim of this thesis was to investigate the effects of school-lunch on children's cognitive functions during afternoon lessons. The research questions are defined as followed and were investigated in two consecutive studies:

2.1 Research aim 1

As pointed out in the previous chapter, lunch consumption may have an impact on particular cognitive functions in adults. The majority of the tasks conducted showed that lunch consumption impaired the adult's cognitive functions. In studies of children the evidence suggests benefits for cognitive functioning if they eat breakfast. The CogniDo study [49] was the first analysis of the short-term effects of lunch on children's cognitive functions. This study revealed no sign of cognitive impairment after lunch. However, no study exists in which the influence of lunch on EF was tested in children. Because EF are potentially more vulnerable to environmental influences, it is likely that food intake around midday may interact with EF performance in the afternoon.

RQ1: Does school lunch intake have short-term effects on EF in children at the beginning of the afternoon lessons?

RQ2: Does school lunch intake affect cognitive performance in children throughout the course of the afternoon lessons?

2.2 Research aim 2

If lunch consumption were to have an influence, it would be not clear how this effect could be mediated. It is discussed whether a postprandial cortisol increase mediates potential lunch-related changes in cognitive performance. As outlined in chapter 1.3.4. a cortisol level increase induced by psychological stress or pharmaceuticals has been observed to impair memory performance in adults [64–66]. For both adults and children lunch intake increases cortisol levels [72–76]. Another relevant mechanism is the post-lunch dip. It has been observed that for some adults lunch consumption exacerbates this naturally occurring dip around midday and results in cognitive impairments.

RQ3: Does a postprandial cortisol increase mediate potential lunch-related changes in cognitive performance?

RQ4: Does school lunch exacerbate a potential post-lunch dip in children?

3 General Methodology

3.1 General study design

To answer the defined research questions two randomized controlled crossover studies were designed: the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS; see study schedule Figure 2) and the Cognition Intervention Study Dortmund Continued (Coco; see study schedule Figure 3).

Both studies were designed as randomized crossover intervention trials and were conducted in 5th and 6th grade school classes at the same comprehensive school in Gelsenkirchen. Participants were randomly sorted into two groups: On day 1, group 1 did not eat lunch, whereas group 2 ate lunch ad libitum and on day 2 (one week later), vice versa. Both test days started at 9:15 hours during the regular morning breakfast break, and each student ate ad libitum from a standardized selection of breakfast foods. During the regular lunch break at 12:25 hours. after the morning lessons, subjects either received pasta Bolognese ad libitum and an apple (lunch day; L) which the school cafeteria staff prepared as usual, or the subjects ate no lunch and spent the break in a separate room (no lunch day; NL). The amount of pasta consumed was measured by individually weighing each plate before and after the meal. Water was available for the subjects at any time in both test situations. The study schedule was integrated in children's school routine. Between the morning break and the lunch break (9:35 - 12:25 hours.) all participants were asked to refrain from eating and drinking (except for water and unsweetened tea). The NL group was additionally asked to refrain from eating and drinking until the end of the cognitive performance assessment. In order to assess compliance with the study protocol the study staff supervised the children in the schoolyard and classrooms during the breaks. Additionally, the participants filled out a questionnaire regarding their food and beverage consumption at the end of each test day.

Differences between the two studies were the timing of cognitive performance assessment in CoCo compared to that of CogniDo PLUS, the composition of the cognitive tests conducted, and the cortisol assessment, which was only conducted in CogniDo PLUS. In the CogniDo PLUS study the EF parameters task switching, working memory updating, and inhibition were tested at the beginning of the afternoon lessons (approximately 45 minutes after lunch), whereas in the CoCo study task switching, working memory, and alertness were tested later in the afternoon (approximately 90 minutes after lunch). Both test batteries were respectively tried in a pre-test with groups of different children of the same age. These pretests revealed that the tests are appropriate for the children at the chosen age. Saliva samples to measure cortisol were taken only in CogniDo PLUS immediately before lunch and at the beginning of the cognitive assessment in the afternoon. A third sample was taken after the cognitive assessment, but it was not integrated in the analyses because it was not relevant to research question.

Figure 2. Study schedule CogniDo PLUS.

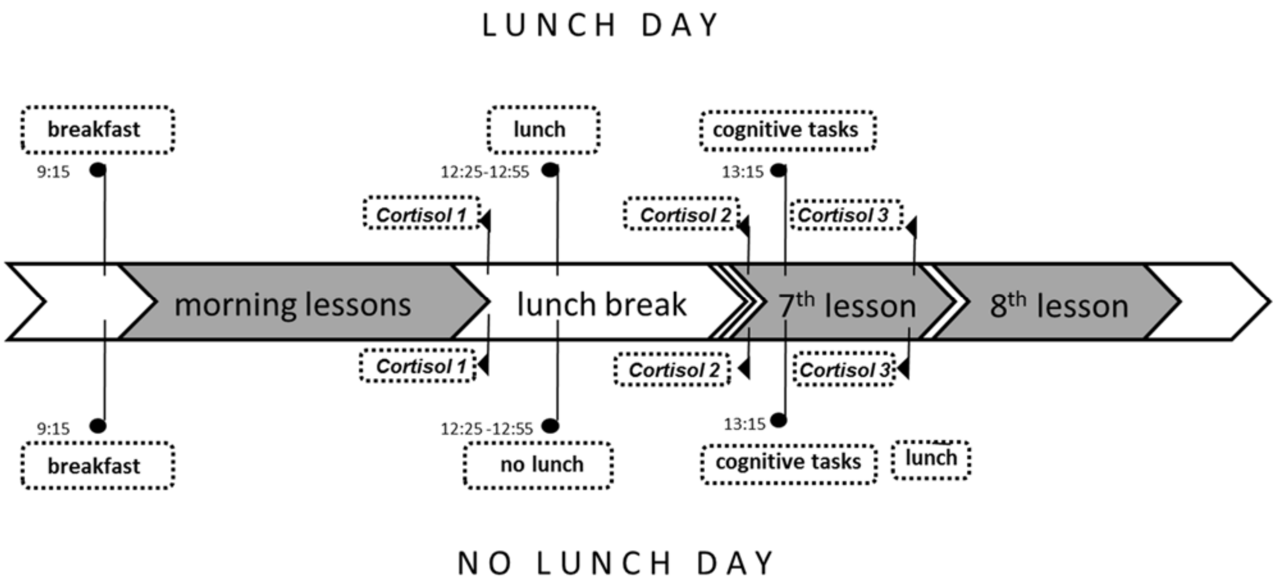
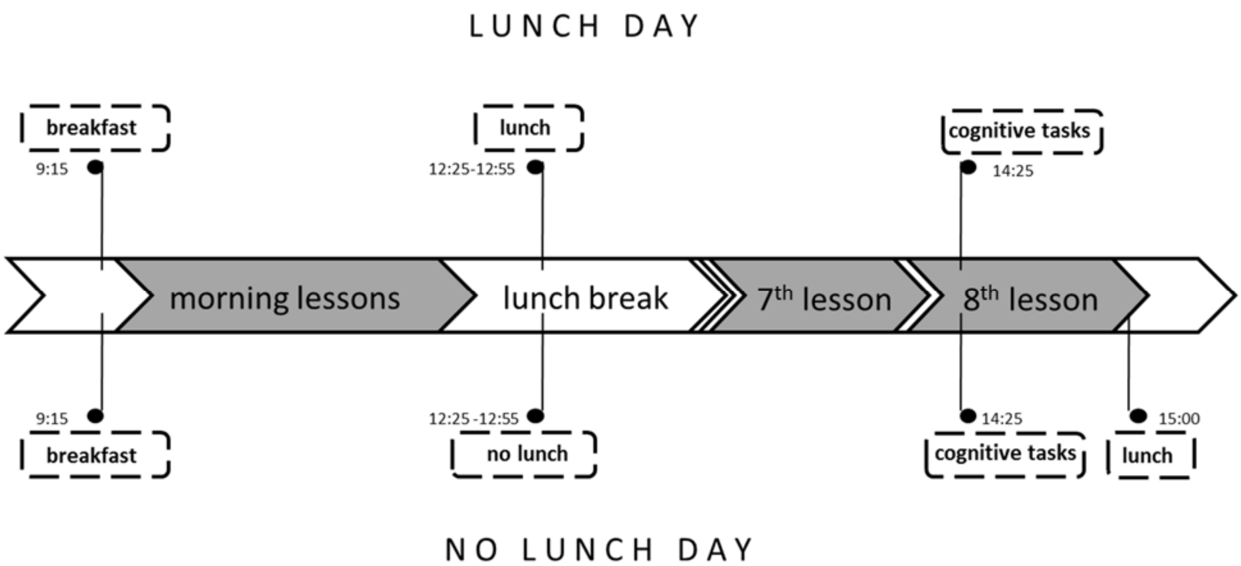


Figure 3: Study schedule CoCo.



3.2 Setting and participants

Both studies were conducted at the comprehensive school Berger Feld in Gelsenkirchen, Germany. This school was selected because it provided the necessary environment for the intervention studies due to its large number of children, who regularly attend lunch at school. The school lunch was prepared at the school's own cafeteria, which allowed for easy communication between the study team and the cafeteria staff. Furthermore, the students at Berger Feld have access to a computer room with a number of computers sufficient for the studies conducted.

In both studies, the participants were recruited from the 5th and 6th grade levels. Any children with a diagnosed learning disorder reported by the class teacher, metabolic disease for which long fasting phases are critical, or on special diets that excluded the lunch meal chosen were excluded. Before the study the class teachers distributed an informational letter with a permission slip in order to receive both parents' and children's consent. On the basis of the written consent forms of parents and children a randomization per class with a block size of 4 participants was conducted in order to assign each participant to one of two study groups. As an incentive, any children who participated on both intervention days received a reward for their participation (a game in CogniDo PLUS and a ball in CoCo).

3.3 Statistical considerations

3.3.1 Analysis of intervention effects in crossover designed studies

The essential benefit of crossover studies compared to those with a parallel group design is that each test person serves as his/her own control. Therefore, any interindividual differences in both the outcome variable and confounders do not bias the study results, i.e. there is no need to question the comparability of the intervention and control group and confounding factors like sex and age can be eliminated from the beginning [79].

In order to guarantee a reliable analysis of crossover studies, a sufficient washout period between the intervention periods is necessary to minimize the chance of a carryover effect, i.e. a possible effect of the treatment is no longer or not as effective during the second intervention/control period. Additionally, the order in which the subject is tested plays an important role and has to be considered in the statistical evaluation. For example, it may be possible that even if treatment A in the first period and treatment B in the second period are the same, the outcome differs due to time effects or getting used to the treatment. An appropriate statistical approach is fundamental to avoid such a confounding, and it is not sufficient to conduct a simple paired t-test [79].

In CogniDo PLUS and CoCo the data were analyzed as recommended by Grizzle [80]. Therefore, the sums of the respective individual outcome variables on the first and second test days were compared between groups in order to examine any potential carryover effects. Appropriate non-normally distributed outcomes were transformed (log, square, root, and reciprocal transformation) and analyzed using unpaired t-test. If transformation did not result in normally distributed sums of parameters, the non-parametric Wilcoxon rank-sum test was used to analyze carryover or intervention effects. Seeing as no carryover effects were observed either in CogniDo PLUS or in CoCo, results from both days were considered for the calculation of the treatment effect. Therefore, individual differences of the particular outcomes of both test days (test day 1- test day 2) were compared between groups (NL-L vs. L-NL) the same way the sums of the outcomes were analyzed. All analyses were performed using the statistical software package SAS 9.2 (SAS Institute, Cary, NC, USA). Values of $P<0.05$ were considered to be statistically significant.

3.3.2 Linear Regression of dose response relationships

To examine a potential mediating effect of single parameters on the lunch effects, a linear regression model was used. In CogniDo PLUS the postprandial cortisol increase and individual lunch size were defined a priori as potential mediators. It was hypothesized that a postprandial cortisol increase may impair cognitive performance. In case of the cortisol increase, two approaches were used. In step 1, a linear regression was used to analyze associations between postprandial cortisol increase (exposition variable) and the change in EF outcomes (outcome variable). These regressions were only conducted in EF outcomes that proved to be affected by eating lunch. If the effects of lunch consumption on EF rely on the increase in cortisol, it implies that lunch effects should be observed in particular in subjects with a high postprandial cortisol increase. This assumption was tested in a second step by performing an additional stratified analysis of lunch effects in subjects with low postprandial cortisol increases versus subjects with high postprandial cortisol increases (using a median-split for postprandial cortisol increase). In CogniDo PLUS and CoCo linear regression analyses between lunch size (exposition variable) and the changes in cognitive performance parameters (performance on lunch day - performance on no lunch day) were conducted for all parameters including age and sex as additional co-variables.

4 Studies and Results

4.1 Study 1: CogniDo PLUS

4.1.1 Summary

Studies indicate that eating lunch impairs some aspects of adults' cognitive functioning. However, the short-term effects of lunch on children's executive functions (EF) have not been examined. The Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) investigated (a) short-term effects of lunch on EF in children and (b) whether the postprandial cortisol increase mediates putative lunch effects on EF performance. 5th and 6th grade students of a comprehensive school in Gelsenkirchen (Germany) participated in the randomized crossover intervention study. On Day 1 of the study, Group 1 did not eat lunch, whereas Group 2 received lunch ad libitum. One week later on Day 2 the groups were treated vice versa. At the beginning of the afternoon lessons, the EF parameters task switching, working memory updating and inhibition were tested using a computerized test battery. Saliva samples were used to measure cortisol directly before lunch and again at the beginning of the cognitive assessment. Of the 215 initially recruited children 21 dropped out of the study due to illness or absence on one of the two test days.

As results lower ratios of false alarms in the working memory updating function were observed when children who ate lunch than for children who had no lunch (8.2 % (lunch) versus 9.4 % (no lunch), $p<0.01$) were seen. Parameters of task switching and inhibition did not differ between children who ate lunch compared to children who had no lunch. Stratification according to postprandial cortisol increase showed that the subgroup with a high increase had lower ratios of false alarms after eating lunch, while false alarm values did not change in the group with a low increase.

In contrast to findings in adults, the results indicate that children's EF are not impaired by lunch under true-to-life conditions. On the contrary, the current study even indicates beneficial effects of lunch intake for the working memory updating. The postprandial cortisol increase in the range observed in our sample does not seem to be related with negative effects on the performance of EF, but even seem to mediate the beneficial effect of lunch on the working memory updating.

4.1.2 Introduction

Considering the increasing numbers of all-day schools in Europe research of potential determinants of children's cognitive performance in the afternoon is a relevant public health issue. Authors of a recent review [30] concluded that lunch can impair some aspects of cognitive functioning in adults. However, appropriate studies of children were not identified. The results of studies of adults are not necessarily transferable to children due to the constitutional and metabolic differences of children and adults (e.g., rapid brain growth, high metabolic rate).

The randomized crossover Cognition Intervention Study Dortmund (CogniDo) [49] recently provided the first insights on the effect of lunch on the cognitive performance of children and found no detrimental short-term effects of lunch on children's basal cognitive functioning (alertness, selective attention, visual-spatial memory). For one parameter (omission errors in tonic alertness tests) the CogniDo study suggests beneficial short-term lunch effects. However, lunch effects on higher cognitive functions, such as executive functions (EF), still need to be examined. EF is an umbrella term for a set of higher-order cognitive processes which govern low-level cognitive functions and are localized in the frontal lobes [11]. EF are most important for daily performance, and during school lessons EF are necessary for retaining and transforming short-term memory contents as well as for controlling actions and planning action sequences [17].

It is known that EF are not fully developed in children and adolescents [19] and decay earlier than low-level functions with increasing age [21]. Aging research shows that lifestyle factors such as physical exercise influence EF [24]. The maturation of the cerebral cortex (including the frontal lobes), which takes place until late adolescence, is characterized by dynamic changes of metabolism. For example, children's brains are more dependent on the intake of glucose in comparison to adult brains [22]. Therefore, it has been hypothesized that the developing EF might be more sensitive to environmental influences (including nutrient intake) than the EF in a fully developed adult brain [22,23]. This study refers to three often postulated executive functions, which are responsible for the following abilities: to inhibit prepotent responses ("inhibition"), to monitor and update information ("working memory updating"), and mental task switching [11,17].

In regard to the reasons for negative effects from lunch intake on adult cognitive performance the so-called post-lunch dip is discussed. It is suggested that a naturally occurring dip in performance around midday seems to be exacerbated by lunch intake [25,59,81]. Little is known about the exact mechanisms how lunch intake could worsen the post-lunch dip. However, different metabolic explanations have been suggested. One explanation might be that the post-lunch increases in blood glucose lead to a hypoglycemic state and a decrease in arousal Other discussed mechanisms assume that an increase in blood glucose after lunch leads to the parasympathetic initiation of an insulin surge or that lunch induce changes in the level of serotonin [63]. The occurrence of this lunch-induced performance dip for EF in children remains to be examined.

Another possible mechanism that would explain how lunch could impair cognitive functions is a meal-induced increase of plasma cortisol. Adrenal cortisol secretion underlies a circadian rhythm with peak plasma concentrations upon waking up and a nadir around midnight [71]. For both adults and children lunch intake induces an increase in cortisol levels [72–76]. It has been observed that an increased cortisol level induced by psychological stress or pharmaceuticals impair memory performance in adults [64–66,82]. Particularly because the frontal brain lobes contain glucocorticoid receptors, it is reasonable that cortisol has an influence on cognitive functions [70]. However, the threshold of a physiological postprandial increase in cortisol, which could effectively impair the cognitive functions, is not known for children. It remains to be evaluated whether a postprandial increase in cortisol might mediate lunch-induced impairments of cognitive performance in children.

In line with CogniDo, the objectives of the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) were to investigate (a) short-term effects of lunch intake on EF in children in the early afternoon and (b) whether a postprandial cortisol increase mediates potential lunch-related changes in cognitive performance.

4.1.3 Methods

4.1.3.1 Study design and participants

This randomized, open-label 2x2 crossover trial also took place in an all-day comprehensive school in Gelsenkirchen, Germany [49]. The research period spanned 6 weeks from beginning of November until the middle of December 2013. Each subject participated on two study days, which took place on the same weekday with one week in between.

The participants were recruited from the 5th and 6th grade levels. Children with a diagnosed learning disorder (reported by the class teacher), metabolic disease (where long fasting phases are critical), or on special diets (which excludes the lunch meal) were excluded. Out of the total of 323 students in the 5th and 6th grades 215 students (67 %) with informed written parental consent were eligible to participate in the study. Each participant was randomly assigned to one of two study groups: on Day 1 of the study, Group 1 did not eat lunch (no lunch, NL), whereas Group 2 received lunch ad libitum (lunch, L) before the assessment of cognitive performance. One week later on Day 2 of the study, Group 1 was in the L condition and Group 2 in the NL condition. Of the 215 children with written consent, 21 dropped out of the study due to illness or absence on one of the two individual test days, which resulted in complete cognitive performance data for 194 children. The children, who participated on both study days, each received a patience game as reward for their participation.

The study was approved by the Ethics Committee of the University of Bonn and registered at clinicaltrials.gov (NCT02082444). All assessments were made in accordance to the Declaration of Helsinki.

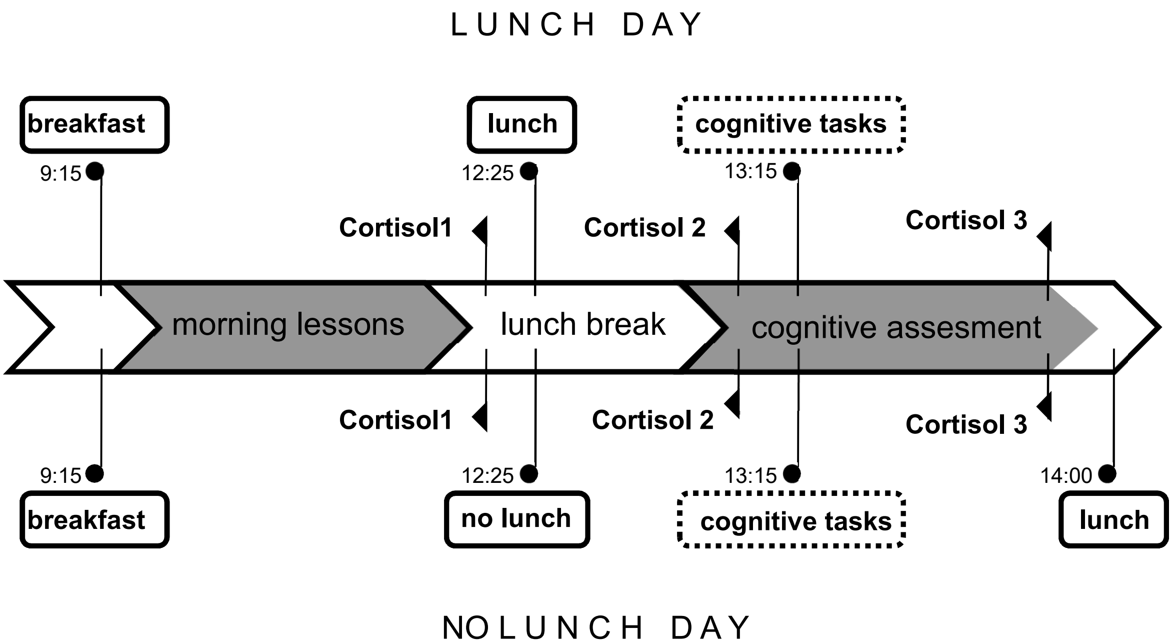
4.1.3.2 Study schedule

The study design was integrated in the regular school-day routine and corresponded to the study schedule of the previous crossover study CogniDo [30], but differed in the integration of additional cortisol measurements (Figure 4).

Both test days started at 9:15 hours during the regular morning breakfast break with a standardized breakfast for each subject (wholemeal bread with margarine, poultry salami or Gouda cheese and carrot sticks) ad libitum. During the regular lunch break at 12:25 hours after the morning lessons, subjects either received pasta Bolognese ad libitum and an apple (L) prepared as usual by the kitchen staff in the school canteen, or the subjects had no lunch in a separate room (NL). The amount of consumed pasta was measured by individually weighing each plate before and after the meal. Water was available for the subjects at any time in both test situations. After finishing lunch all students (L and NL) had a common break until 13:10 hours (app. 15-20 min). At the beginning of the regular afternoon lessons (at 13:15 hours) the assessment of cognitive performance took place in the school's computer room. After completing these tests participants who had no lunch at the regular time received their lunch (14:00 hours).

Between the morning break and the beginning of cognitive performance tests (from 9:35 hours to 14 hours), all participants were asked to refrain from eating (except the lunch at the L condition) and drinking (except for water and unsweetened tea). In order to assess whether the students followed this requirement (for a protocol analysis) the participants filled out a questionnaire regarding their food and beverage consumption at the end of each intervention day. Additionally, the study staff supervised the children in the schoolyard and classrooms during the breaks. Saliva samples were collected to examine the saliva-cortisol status two times on each test day: before lunch (at approx. 12:20 hours) and before the computerized cognitive assessment (at approx. 13:10 hours).

Figure 4: Schedule of the intervention day for lunch group and no lunch group in the CogniDO PLUS study.



4.1.3.3 Cognitive assessment

For the assessment of executive functions (EF) a computerized test battery consisting of 3 tasks designed by the Institute of Working, Aging and Learning (ALA Institute) in Bochum, Germany was used. At the beginning of the cognitive assessment each task was explained to the group, and the participants were able to practice each task once in a training mode. After the training phase and a 5-minute break with low (physical) exercise the actual testing began. Subjects were requested to respond as quickly as possible without sacrificing accuracy. With regard to reaction times (RT) (working memory updating and inhibition) plausible data were defined as values within the range of quartile 1 minus 1.5 times the Inter Quartile Range (IQR) and quartile 3 plus 1.5 times the IQR [83]. For the "ratios of errors" a predefined limit of < 50 % error rate was defined as plausible since the expected error rate would be 50 % if a subject is choosing his/her reaction completely by random. So, exclusion of error ratios > 50 % was intended to reduce the risk of a systematic error in observations from single subjects as these values indicate a high risk that the task instructions were misunderstood (e.g. if a subject always pushed a button when no reaction was attended).

The tasks were applied in the following order:

Task switching (switch)

With an alternative version of the Trail Making Task (TMT) we measured the subjects' ability to switch between two tasks. This task was presented in 3 sections - the first two sections (section one: numbers, section two: letters) in a non-switch condition and the third section in a switch condition (letters and numbers mixed) (Figure 5):

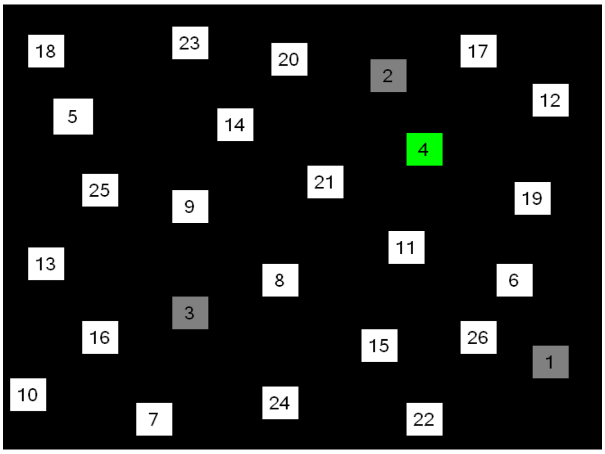
First section, numbers, non-switch: Black numbers from 1 to 26 in white squares were presented in an irregular order on a black computer screen. The children were asked to click the numbers in ascending order with the mouse curser. The starting point was a square with the number 1, which was marked green. The squares turn green after a correct answer and red after a false answer as a form of feedback, and the correctly processed squares fade out. The maximum time limit to finish the task was 3 minutes.

Second section, letters, non-switch: This section had the same format as the numbers section, but used letters from A to Z instead of numbers.

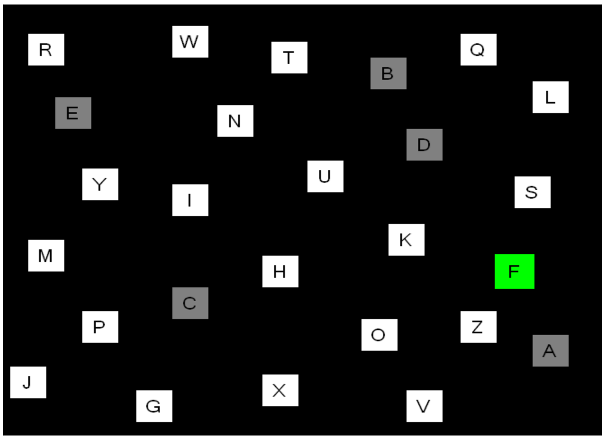
Third section, switch: The 26 squares contained numbers from 1 to 13 and letters from A to M. The children were asked to alternately click numbers and letters in ascending order (i.e. 1-A-2-B-3-C…).

The outcomes consisted of the total reaction time for numbers (for items 1-25), total reaction time for letters (for items 1-13) and switch-costs, i.e. the processing time of the third (switch) section minus the first section (numbers; nonswitch) minus the difference between the first 13 items of the first and second (letter; nonswitch) section. To eliminate any implausible data we excluded all subjects with negative switch costs.

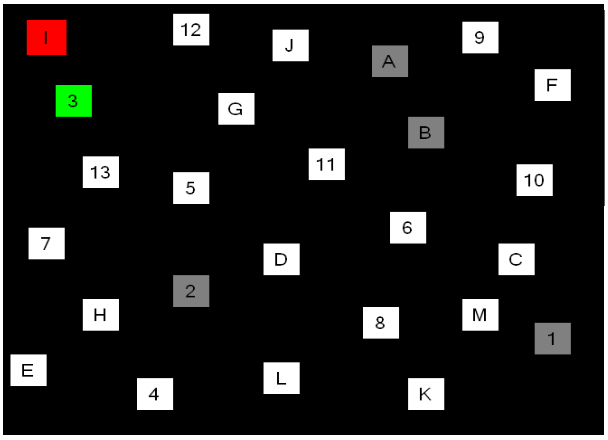
Figure 5: Cognitive assessment: Switch. Screenshots of the sections.



1) First section, numbers, non-switch: children were asked to click the numbers in ascending order with the mouse curser.



2) Second section, letters, non-switch: same format as the first section, but used letters from A to Z instead of numbers.



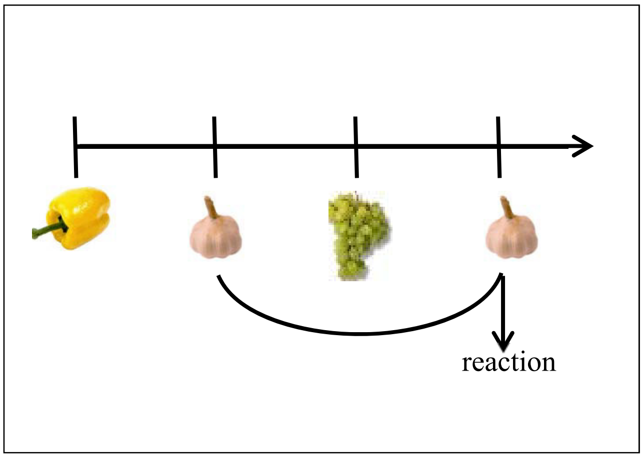
3) Third section, switch: children were asked to alternately click numbers and letters in ascending order (i.e. 1-A-2-B-3-C…).

Working memory updating (2-back task)

In order to assess the function of constant monitoring and adding or deleting of working memory contents, we used the n-back task in a 2-back condition. Participants were asked to monitor a sequence of 106 consecutive trials (pictures of fruits and vegetables) presented in the middle of a white screen. When the current picture matched, the picture presented 2 trials before (n-2), the participant was instructed to press a pre-defined key on the computer keyboard with the index finger (Figure 6). The stimuli were presented for 500 ms with an interstimulus interval (ISI) of 2100 ms and a maximal reaction time of 1400 ms. No feedback was given. Of the 106 pictures shown, 21 were targets (same picture as 2 trials before).

The outcomes were ratios of missings (no reaction while reaction was required), ratios of false alarms (reaction while no reaction was required) and mean reaction times. Plausible measurements were defined as mean of ratio of incorrect answers and false alarms < 50 % and reaction times > 172.8 ms and < 864.7 ms.

Figure 6: Cognitive assessment: 2-back task. Scheme of the task.



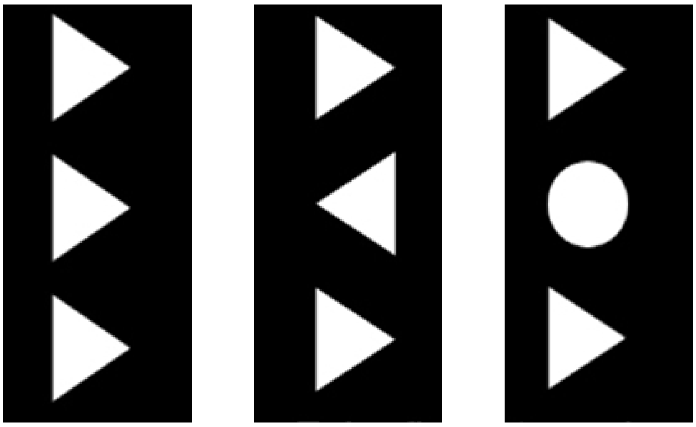
Inhibition (flanker task)

The flanker task was used to assess the ability to inhibit a prepotent response. In each trial three superposed triangles were presented to the participants. The upper and lower triangles (flankers) were pointing in the same direction but independent from the middle triangle (target). Each trial was categorized as compatible-go, incompatible-go or no-go (Figure 7). Compatible-go trials were those during which flankers and the target were pointing in the same direction, incompatible-go trials were those when flankers and target were pointing in opposite direction. During the no-go trials a circle replaced the target. The participants were supposed to press the buttons left or right according to the direction of the target or not react in the case of a no-go condition. To induce flanker-target conflict, the flankers were presented individually for 100 ms and remained together with the target for another 300 ms. The maximum reaction time was 1100 ms, the response stimulus interval (RSI) was 1200 ms (varying +/- 20 %). In go-trials the feedback "faster" was shown when there was no reaction during the maximum reaction time or when the reaction time was > 600 ms. The feedback stimulus started 500 ms after the response (or after 1100 ms in no-reaction trials) and lasted 300 ms. In total, this task consisted of 102 items (32 no-go, 35 incompatible-go, 35 compatible-go).

The outcomes were the difference between the ratio of incorrect reactions in compatible-go and incompatible-go trials (difference error rate), the difference between mean reaction times of compatible-go and incompatible-go trials (rt slowing) and the mean count of false alarms (participants pressed the button instead of showing no reaction). Reactions for which a negative reaction time was detected (i.e. the subject gave the reaction before the trial showed up) were excluded.

Plausible data were defined as follows: ratio of incorrect reactions in compatible-go trials < 50 %, ratio of incorrect reactions in incompatible-go trials < 50 %, count of false alarms < 16 (less than 50 %), reaction times of compatible-go trials > 138.9 ms and < 438.6 ms, reaction times of incompatible-go trials > 201.2ms and < 514.0 ms.

Figure 7: Cognitive assessment: Inhibition. Screenshots of the trials.



compatible-go | incompatible-go | no-go

4.1.3.4 Cortisol measurement

Saliva samples were taken at three times on each intervention day: directly before lunch (at approx. 12:20 hours, T1) and at the beginning of the cognitive assessment (i.e. approximately 45 minutes after starting lunch (at approx. 13:10 hours), T2; see Figure 4). For this analysis samples T1 and T2 were taken into account in order to consider postprandial cortisol changes before the cognitive tests as a potential mediator of putative lunch effects on cognitive performance. Under supervision participants collected their own saliva using Salivette collection devices (Sarstedt, Nuembrecht, Germany). The samples were stored in a cool box for the transport and frozen (-18°C) until they were analyzed. For the analyses the Salivettes were thawed and centrifuged at 3000 rpm for 2 minutes in order to obtain a clear fluid. Free saliva cortisol levels were measured using the RE62011 immunoassay (IBL, Hamburg, Germany). One third of the samples were analyzed two-fold and the remainder one-fold.

4.1.3.5 Statistical analysis

All analyses were performed using the statistical software package SAS 9.2 (SAS Institute, Cary, NC, USA). $P<0.05$ was considered as statistically significant.

The parameters of the three cognitive tasks and the cortisol status at the beginning of the cognitive assessment (T2) as well as the difference of the cortisol status before the lunch break and before cognitive assessment (T2-T1) were used as outcome variables. All outcome variables were interval-scaled. As recommended by Grizzle [80] the sums of the respective outcome variable were compared between groups using the parametric unpaired t-test for normally distributed data (including normal distribution after transformation) and the non-parametric Wilcoxon rank-sum test for non-normally distributed data to examine potential carry-over effects. Appropriate non-normally distributed outcomes were transformed (log, square, root and reciprocal transformation) and analyzed using unpaired t-test. If transformation did not result in normally distributed parameters, the non-parametric Wilcoxon rank-sum test was used to analyze carry-over or intervention effects. As no carry-over effects were observed, results from both days were considered for the calculation of the treatment effect. Therefore, individual differences of the particular outcomes of both test days (test day 1- test day 2) were compared between groups (NL-L vs. L-NL) the same way the sums of the outcomes were analyzed. Descriptive data (sex, lunch size and eating a refectory lunch) were analyzed by Chi-squared test or Fisher's Exact Test. Before the statistical analyses the raw data were checked for plausibility as described above. For cortisol, all available samples of T1 and T2 were analyzed.

To examine the potential mediating effect of the postprandial cortisol increase on potential lunch effects on the individual cognitive performance two approaches were used: In step 1, a linear regression was used to analyze associations between postprandial cortisol increase and the EF outcomes which were affected by eating lunch. If effects of having lunch on EF rely on the increase in cortisol, it implies that lunch effects should be observed in particular in subjects with a high postprandial cortisol increase. This assumption was tested in a second step performing an additional stratified analysis of lunch effects in subjects with low vs subjects with high postprandial cortisol increases (using a median-split for postprandial cortisol increase).

In addition, linear regressions between the lunch size and EF outcomes and between lunch size and cortisol increase were conducted (model 1 unadjusted and model 2 adjusted for sex and age).

4.1.4 Results

Participants

Data from cognitive assessment were not available for 2 subjects for the task switching and flanker task. For the 2-back data from 1 subject were missing.

Implausible data in cognitive performance were excluded separately for each of the 3 tasks. For the switch task 91.7 % of the participants were included, for the 2-back task 57.5 % and for the flanker task 65.6 %. Characteristics of the excluded and included subjects for each task are shown in Table 1. In the 2-back task the excluded subjects were slightly older (11.7 years) than the included (11.4 years). The ratio of boys was higher among excluded (76.8 %) than included (45 %) subjects. Participants' mean consumption of the pasta bolognese was 368 g (+/- 154 g) (range: 55-920 g).

Table 1: Characteristics of included and excluded schoolchildren participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) (Switch and Flanker: n=192; 2-back: n=193)

	Switch		**2-back**		**Flanker**	
	Included (176)	excluded (16)	Included (111)	Excluded (82)	included (126)	excluded (66)
Age, mean +- s.d. years	11.5 (0.76)	11.8 (0.74)	11.4 (0.73)	11.7 (0.79)	11.6 (0.76)	11.5 (0.78)
Female, n (%)	76 (43.2)	5 (31.3)	61 (55)	19 (23.2)	56 (44.4)	24 (36.4)
Regular lunch [a], n (%)	158 (89.8)	14 (87.5)	100 (90.1)	73 (89)	110 (87.3)	62 (93.9)
Pasta consumption (median P25/P75) [g]	345 (265/418)	373 (315/573)	335 (235/405)	375 (315/430)	343 (275/415)	360 (245/430)

[a] Defined as consuming lunch at the school cafeteria regularly by subscription

General lunch effects on cognitive functions

The ratio of false alarms in working memory updating (2-back task) was significantly lower in the L than in the NL condition ($p=0.01$, Table 2). For inhibition (flanker task) the students tended to make fewer errors in the L condition than in the NL condition, but this difference was not statistically significant (difference of error rate $p=0.16$) and disappeared with additional exclusion of subjects who did not follow the study protocol ($n=26$, per protocol analysis) whereas the intervention effect of the ratio of false alarms in working memory updating remained in the per protocol analysis (data not shown). No lunch effects were observed for the other parameters of cognitive performance.

The linear regression of lunch size and EF generally showed no association between lunch size and EF except for a (non-significant) trend between switch costs and lunch size (ß=-18.51 and $p=0.083$ in not adjusted model and ß=-18.95 and $p=0.078$ in the adjusted model)., i.e. the children with higher lunch sizes tended to have lower switch costs. The linear regression of lunch size and postprandial cortisol increase revealed postprandial cortisol increase were positively associated with lunch size (not adjusted model: ß=0.0025 and $p=0.0081$ adjusted model: ß=0.0024 and $p=0.0113$).

Table 2: Effects of no lunch vs. lunch on examined parameters of executive functioning in schoolchildren (10-12 years) participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS)

Task	Main outcome	No Lunch			Lunch			p-value
		Median	25th	75th	Median	25th	75th	
Switch (n=176)	switch costs[a] [ms]	27633	15017	39117	24844	15256	37326	0.34 [b]
	visual search letters[c] [ms]	26462	22893	32361	27121	23424	32002	0.47 [d]
	visual search numbers [ms]	45639	40439	51548	46392	38836	52987	0.91 [d]
2-back (n=111)	ratio of missings [%]	28.6	23.8	35	28.6	19	38.1	0.82 [d]
	ratio of false alarms [%]	9.4	4.7	21.2	8.2	3.5	17.6	**0.01** [d]
	rt [e] [ms]	513.3	443.5	598.5	529.2	432.5	596	0.87 [d]
Flanker (n=126)	rt slowing [ms]	67.3	52.9	89.5	73.1	44.7	90.5	0.18[d]
	difference error rate [%]	0.14	0.06	0.22	0.11	0.06	0.23	0.16[b]
	mean count of false alarms [N]	1.0	0.0	4.0	1.0	0.0	3.0	0.24[d]

[a] switch costs = (mean rt switch task) – (mean rt number task) – (mean rt first 12 reactions of letter task); [b]Two sample t-test; [c] first 12 reactions; [d] Wilcoxon rank-sum test; [e] reaction time

Postprandial cortisol and cognitive functions

The cortisol samples were collected before lunch (T1=12:20 hours) and again about 45 minutes after starting lunch (before cognitive assessment at 13:10 hours=T2). Cortisol levels at T2 were significantly higher on the L day and also the cortisol levels between T1 and T2 increased significantly in comparison to the NL day (p<0.0001,Table 3). On the NL day the cortisol levels decreased from T1 to T2 (p<0.0001) while levels increased on lunch (p<0.001).

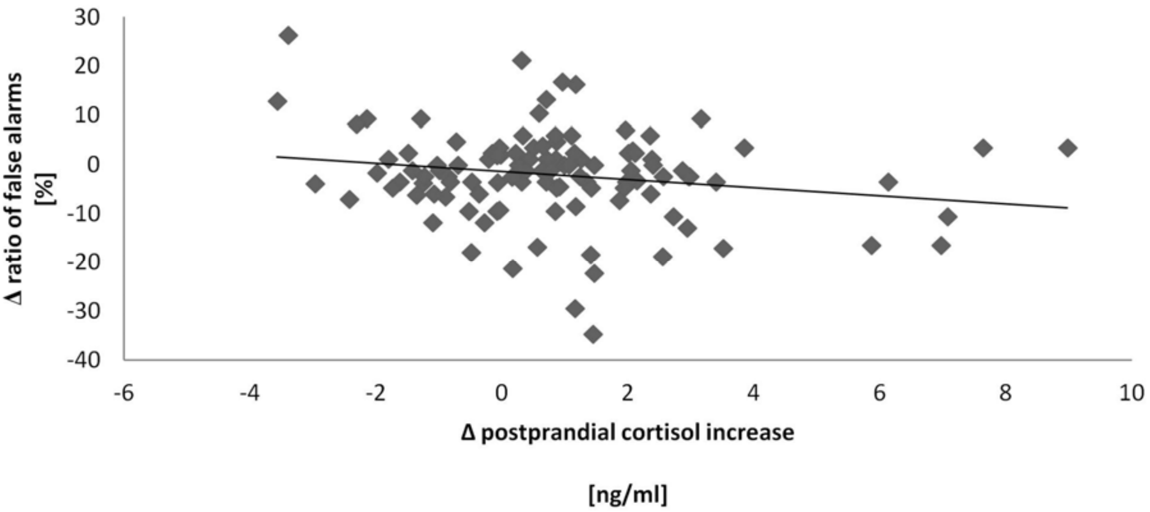
Table 3: Saliva cortisol levels of schoolchildren participating in the Cognition Intervention Study Dortmund PLUS before lunch and changes over time (n=187)[a]

	No Lunch			**Lunch**			
Cortisol	**Median**	**25th**	**75th**	**Median**	**25th**	**75th**	**p-value[b]**
T2 [ng/ml][c]	1.21	0.76	1.78	1.88	1.13	2.99	**<0.0001[d]**
Change between T1 and T2[e]	-0.38[f]	-1.06	0.05	0.27[f]	-0.46	1.09	**<0.0001[g]**

[a] 5 missings; [b] p-value of lunch-effect; [c] T2=Cortisol level at 13:05 (ng/ml); [d] Wilcoxon rank-sum test;
[e] Change between T1 and T2=Difference of cortisol level T2 at 13:05 hours – T1 at 12:15 hours (ng/ml);
[f] Wilcoxon signed rank test;
[g] Two sample t-test

The linear regression revealed a significant negative linear association between postprandial cortisol increases and the ratio of false alarms in the 2-back task, i.e. decreasing ratios of false alarms for increasing levels of postprandial cortisol increase (ß=-0.83 and p=0.04) (Figure 8). The linear regression of cortisol on the remaining EF outcomes revealed a significant association only with the visual search for numbers within the task switching (ß=893.5 and p=0.04). However, the lunch intervention had no influence on this outcome.

Figure 8: Linear regression between postprandial cortisol increase (considering cortisol changes in fasting condition, i.e. cortisol change Lunch minus cortisol change No Lunch) and the difference of the ratio of false alarms of the 2-back task (p=0.04) (n=108)



Stratification according to postprandial cortisol increases (median cut, high vs. low) generally confirmed the results of the pooled analysis, as no differences were observed for parameters of the switch and flanker task (data not shown). In the 2-back task a significant lower ratio of false alarms after lunch was observed only in students with a high postprandial cortisol increase (p=0.03, Table 4).

Table 4: Effects of no lunch vs. lunch on the main outcomes of the 2-back task after stratification for high or low postprandial saliva cortisol increase in schoolchildren participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) (n=108)

			No Lunch			**Lunch**			**p-value**
Test	**Variable**	**Cortisol increase**	**Median**	**25th**	**75th**	**Median**	**25th**	**75th**	
2-back	ratio of missings [%]	high (n=50)	28.57	28.81	33.33	28.57	19.04	40.00	0.678 [a]
		low (n=58)	28.57	19.05	38.1	28.57	19.05	38.1	0.642 [a]
	ratio of false alarms [%]	high (n=50)	9.41	5.88	22.35	8.24	3.53	16.47	0.033 [b]
		low (n=58)	11.18	4.7	19.28	8.24	3.53	17.86	0.232 [a]
	rt [ms]	high (n=50)	518.64	460.37	586.25	530.63	418.06	595.07	0.832 [a]
		low (n=58)	487.73	441.80	604.18	517.94	432.50	602.91	0.820 [a]

[a] Wilcoxon rank-sum test; [b] Two sample t-test with (square) transformed data; [c] reaction time.

4.1.5 Discussion

The present study revealed that there was no evidence for a lunch-related dip in cognitive performance of children. In contrast, levels of false alarms in the working memory updating task were lower when lunch was eaten compared to the test condition in which lunch was omitted. After meals children responded less frequently to non-targets, to which they should have not responded. Because no effect of meals was observed in the inhibition task, it could be hypothesized that the decrease of false alarms is not the consequence of an improvement of inhibition, i.e. of less premature responses. Rather the results may suggest that meals improved working memory updating, i.e. they helped children to distinguish non-targets more clearly from targets. The measured postprandial cortisol increase did not seem to cause declines in performance of EF. On the contrary, the results suggest that those subjects with a high postprandial cortisol increase could experience a beneficial effect on the working memory updating function, i.e. they correctly disregarded non-targets. This analysis confirms that effects of having lunch are indeed only observed if lunch results in high increases of cortisol levels. Therefore, effects of having lunch on children's updating of the working memory seem to be – at least partly – mediated by increasing levels of cortisol after lunch.

Until now, the impact of no lunch or lunch on cognitive functioning has primarily been investigated in adults [25–27,63]. The outcomes of these studies are not fully consistent, but point to impairments of some parameters of cognitive functioning after eating lunch in comparison to the functioning in a pre-lunch condition. Smith and Miles [26] observed no post-lunch changes in selective attention in 48 students but did observe impairments in the ability to maintain attention and reaction times on new stimuli in contrast to pre-lunch testing. In another study it was shown that lunch impaired the ability to discriminate [25]. In contrast, Karnarek and Swinney [27] observed an improvement in reading ability after lunch compared to the no lunch condition, whereas no effects were seen in working memory or sustained attention. A review [84] concluded that lunch seems to attenuate sustained attention more than briefer tasks of selective attention.

To the best of the knowledge of the authors, the previous CogniDo [49] study was the first investigation on the cognitive effects of lunch in children. In line with findings from the current study the CogniDo study neither revealed negative nor general beneficial effects of lunch on parameters of general cognitive performance, such as the visuospatial subsystem within the working memory, or on selective attention. While the CogniDo study suggested an improvement of tonic alertness (as reflected in a lower rate of omissions), the present study revealed an effect on short-term memory and it's updating by using executive functioning tests. Although cognition tasks of the 2 studies partially measured different parameters, a summary of results from both studies suggests that – in contrast to adults – lunch does not seem to impair short-term cognitive functioning in children, but might even improve single cognitive parameters.

A discussed phenomenon of the implied impairment in cognition in adults after lunch is the so called post-lunch dip, i.e. a naturally occurring dip at midday, which is supposedly related to more than one factor, e.g. individual circadian rhythm or length of time since sleep [59–61]. It was observed that lunch exacerbates the naturally occurring dip in adults [25,63]. The mechanism behind this phenomenon is not yet understood, but e.g. alterations in the synthesis of neurotransmitters by availability of amino acids or postprandial hypoglycemia have been discussed as potential factors [62].

In adults the post-lunch dip is assumed to be maximally 60-120 minutes after starting lunch [25,61,63]. In both studies (the present and the previous) cognitive tests were carried out about 60 minutes after starting and 40 minutes after the subjects finished lunch. No lunch-induced dip was seen in children. However, it cannot be ruled out that a post-lunch impairment might occur in children later in the afternoon.

Another possible mechanism explaining the effect of lunch on EF may be related to meal-induced postprandial increases of plasma cortisol [30]. Adults exhibited impaired memory performance after cortisol increases induced by psychological stress or pharmaceuticals [64–66]. Lupien et al. [67] ascertained that working memory is more prone to pharmaceutically induced cortisol increase than declarative memory. From their results they inferred that a low pharmaceutical cortisol dose improves the processing capacity of working memory, whereas a high dose leads to impaired working memory performance. A quadratic function (inverse U-shape curve) is suggested between performance on the working memory task and changes in glucocorticoids levels after hydrocortisone infusion [65,68]. Abercromie et al.[85] found out that a dose of 20 or 40 mg cortisol given previous testing caused fewer errors of commission i.e. false alarms in 18-33 year old adults. This finding was reproduced in the present study since the false alarms in the flanker task were reduced by a physiological raise of cortisol after lunch.

In this study lunch induced a cortisol increase in children, with larger lunch sizes causing increasing cortisol responses (not adjusted model: ß=0.0025 and p=0.0081 adjusted model: ß=0.0024 and p=0.0113). After dividing the subjects into two groups based on postprandial cortisol increase (high and low), the improved working memory updating results after lunch remained only in the high postprandial increase group. We assume that physiologically induced cortisol increase after lunch could improve the working memory updating function in children reflecting the beneficial effect of lunch on working memory updating. No conclusions can be drawn, however, about the amount of the increase or the exact level at which improvements or impairments occur. Micha et al.[86] considered the cortisol response in a study on the impact of GI/GL on cognitive performance in children. It was observed that a high GI breakfast increases the blood glucose and cortisol levels which results in a better performance on a virgilance task. They hypothesized that higher blood glucose levels could activate the hypothalamic–pituitary–adrenal axis in stressful situations (i.e. test situation) which lead to higher cortisol levels and higher tension of the participants before the tests, and thus better performance on tasks were information processing is tested. However, more studies in children are needed to investigate the effect and metabolism pathway of postprandial cortisol levels on cognitive performance in children.

Several characteristics of the study design need to be discussed. CogniDo PLUS was not conducted under clinical conditions, but tested the students under real-life conditions in their school environment. In this setting students are exposed to different influences (e.g. their peers), which might be an explanation for the high count of implausible outcomes. Due to the limiting values in the inhibition and working memory updating task, about 35-42 % of the students were excluded per task because of an error rate over 50 % or an implausible reaction time (see definition of plausible data in ‘methods’). We suppose that the high error rate resulted from accuracy speed trade-off because the excluded participants partially showed relatively short reaction times. It could be assumed that these students suppressed accuracy for speed in order to “win” a speed contest with their seat neighbors. However, only a classroom setting mirrors the real-life conditions in school and allows conclusions about everyday school life compared to purified clinical conditions. The schedule of the intervention was embedded in the “normal” school day with EF tasks performed at the usual start of the afternoon lessons. Therefore, the transferability of our results is given. However, the effect on cognitive performance later in the afternoon remains to be evaluated.

For practical reasons (e.g. no extra medical staff, no cortisol increase because of the stress of blood sampling) postprandial cortisol was not assessed with blood sampling but instead from saliva samples. However, saliva cortisol levels are a valid reflection of the respective unbound hormone in blood [82]. Unfortunately, an evaluation of the analyzed cortisol data is not possible because of a lack of consensus reference values for children's saliva measurement [87]. Nevertheless, absolute values of cortisol were of minor importance in the current study because only the individual changes of cortisol levels of the subjects were considered. Using the Grizzle model for the analyses of intervention effects, it was not possible to perform an interaction test between cortisol and working memory updating as part of a pathway analysis. However, we tested the modifying effect of postprandial cortisol indirectly via stratified analysis of lunch effects and also through linear regression between postprandial cortisol increase and change in working memory updating.

Although absolute lunch size was not associated with most EF parameters, more information would have been needed e.g. about regular individual lunch size in order to draw any conclusions between meal size and cognitive performance. It was not possible to get this information due to organizational reasons, but this question would be interesting for future studies. The present study (as well as CogniDo) stands out due to the crossover design, which eliminates variations between the subjects and reduces bias.

In conclusion and in contrast to findings in adults, the results indicate that lunch does not impair children's EF in real-life conditions. For the parameter reflecting working memory updating the current study even indicates beneficial effects of lunch intake. Taken together, CogniDo and CogniDo PLUS reveal that eating lunch during the school day has no negative effects on cognition in children. For some tasks school lunch could even have partially positive influence on cognitive performance of some children in the afternoon, but these observations need to be further investigated in future studies.

4.2 Study 2: CoCo

4.2.1 Summary

Studies about effects of school lunch on children's cognition are rare; two previous studies (CogniDo [49], CogniDo PLUS [88]) generally found no negative effects of lunch on children's cognitive performance at the end of lunch break (i.e. 45 min after finishing lunch), but suggested potential beneficial effects for single parameters. Therefore, the present study investigated the hypothesis of potential positive effects of school lunch on cognitive performance at early afternoon (90 min after finishing lunch). A randomised, crossover intervention trial was conducted at a comprehensive school with fifth and sixth grade students. Participants were randomised into two groups: On day 1, group 1 did not eat lunch, whereas group 2 received lunch ad libitum. On day 2 (1 week later), group 2 did not eat lunch and group 1 received lunch ad libitum. The cognitive parameters task switching, working memory updating and alertness were tested using a computerised test battery 90 min after finishing the meal. Of the 204 recruited children, fifty were excluded because of deviations from the study protocol or absence on one of the 2 test days, which resulted in 154 participants. Data showed no significant effects of lunch on task switching, working memory updating and alertness (P values between 0.07 and 0.79). The present study suggests that school lunch does not seem to have beneficial effects on children's cognitive functions regarding the conducted tests at early afternoon. Together with our previous studies, we conclude that school lunch in general has no negative effects on cognitive performance in children. However, beneficial effects seem to be restricted to a relatively short time period after eating lunch.

4.2.1 Introduction

Considering the extension of all-day schools in Europe, knowledge about potential effects of school lunch on children's cognition is becoming increasingly important. Short-term lunch effects on cognition were primarily examined in adults with equivocal results, until now. Although sustained attention and the ability to discriminate have been shown to be worsened after lunch [25,26], other cognitive aspects were improved (reading ability) [27] or did not change (selective attention) [63]. One explanation for negative lunch effects on cognitive performance is the post-lunch dip – a naturally occurring nadir in performance at midday. Studies in adults have shown that this dip is worsened by lunch [25,59]. Therefore, it could be hypothesised that skipping lunch could result in an alleviation or prevention of this post-lunch dip. However, these results were obtained in adults and are not necessarily transferable to children due to constitutional and metabolic differences between children and adults (e.g. still rapid brain growth, high metabolic rate in children). Until now, studies in children about the effects of meals on cognition have mainly concentrated on breakfast [45]. A body of research work has shown short-term benefits for cognitive performance when children eat breakfast instead of skipping it [45]. However, other studies showed that poorly nourished children benefit more than well-nourished children [47]. Although no definitive conclusions can be drawn about short-term benefits of breakfast for cognitive performance in children in general, there is at least a noticeable indication for a beneficial influence [45,89,90]. Studies about school lunch and short-term effects on children's cognition are rare [30]. To the best of our knowledge, there are only two crossover, intervention trials from our group, which provided first insights into the impact of school lunch on cognitive performance in children at the end of lunch break (CogniDo [49], CogniDo PLUS study [88]). Both studies did not reveal negative effects of lunch on several parameters of cognitive functions (CF) (i.e. task switching, working memory updating, inhibition, alertness, selective attention, block span) when determined about 45 min after finishing lunch. For single parameters such as omission errors in the alertness task and false alarms in the updating task (regarding the working memory) results even point to beneficial effects of lunch at the end of lunch break. Even though no lunch-related cognitive impairment was observed in these two studies, it has to be considered that a post-lunch dip in children could appear as the afternoon progresses. In adults, a post-lunch dip was observed about 60–120 min after lunch [25,61,63], indicating that the time span between a meal and the measurement of cognitive performance could have a relevant influence [61]. Therefore, the objective of the Cognition Intervention Study Dortmund Continued (CoCo) was to investigate the potential positive effects of school lunch on cognitive performance in children at early afternoon (90 min after finishing lunch instead of 45 min in the previous intervention studies). In order to provide comparability with both previous studies, cognitive tests that proved to be the most sensitive were chosen – that is, the alertness task from the first study (CogniDo) [49] and task switching and working memory updating task from the second study (CogniDo PLUS) [88]. On the basis of the previous results of these two studies, we hypothesise that children will perform better on lunch day than on no lunch day.

4.2.2 Methods

4.2.2.1 Study design and participants

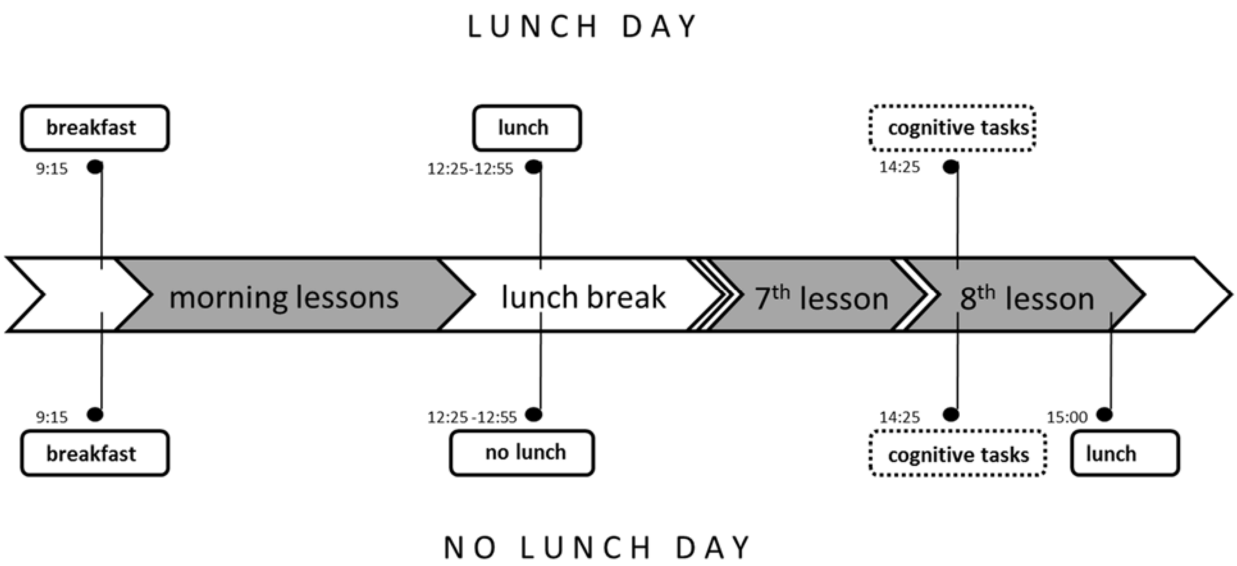
Similar to the previous studies, the CoCo study was conducted as a randomised, open-label, 2 × 2 crossover intervention trial. The same all-day comprehensive school in Gelsenkirchen, Germany, was chosen for the experiment. In total, the field period spanned 19 weeks between October 2014 and March 2015 including 3 weeks of holidays. Each subject had to participate on two study days with 1 week in between on the same weekday. The participants were recruited from the fifth and sixth grades (twelve classes). The students of the sixth grade in the present study had already participated in the previous CogniDo PLUS [88] study (as fifth grade students). Children with diseases with potential consequences of fasting and children on special diets, who were not allowed to eat the study meal, were excluded from participation. Children with a diagnosed learning disorder reported by the class teacher were allowed to participate, but were excluded post hoc for the analyses. Out of 324 students, 204 provided informed written consent to participate. A cluster randomisation per class with a block size of four participants was conducted to assign participants to one of two study groups: on day 1 of the study, group 1 did not eat lunch (no lunch, NL), whereas group 2 received lunch ad libitum (lunch, L); 1 week later, on day 2 of the study, group 1 was in the L condition and group 2 in the NL condition. All children, who participated on both intervention days, received a ball as reward for their participation. The study was approved by the Ethics Committee of the University of Bonn and registered at clinicaltrials.gov (NCT02344056). All assessments were made in compliance with the Declaration of Helsinki.

4.2.2.2 Study schedule

The study design was integrated in the regular school routine and corresponded to the study schedules of the previous studies (CogniDo [49], CogniDo PLUS [88]), but differed in the time interval between lunch break and assessment of cognitive performance (Figure 9). On both test days, a standardised breakfast (wholemeal bread with margarine, poultry salami or Gouda cheese and carrot sticks) ad libitum was offered to the students in the regular morning break at 09.15 hours. During the regular lunch break, starting at 12.25 hours, the subjects either received lunch ad libitum (pasta with or without Bolognese sauce as desired) and an optional apple in the school canteen prepared as usual by the kitchen staff (L), or the subjects skipped lunch and stayed in a separate room (NL). Water was available at any time in both test situations. The amount of pasta was individually weighed before and after the meal ± 5 g. After lunch, all students (L and NL) had their common break (until 13.20 hours) and the regular seventh lesson

(13.25–14.10 hours). At the beginning of the eighth lesson (14.15 hours), the assessment of cognitive performance took place in the school's computer room at about 14.25 hours. After completing these tests (about 15.00 hours), participants in the NL condition received their lunch. Between the morning break and the lunch break (09.35– 12.25 hours), all participants were asked to refrain from eating and drinking (except for water and unsweetened tea). The NL group was additionally asked to refrain from eating and drinking until the end of the test day (15.00 hours). In order to assess compliance with the study protocol, the study staff supervised the children in the schoolyard and classrooms during the breaks. In addition, the participants filled out a questionnaire regarding their food and beverage consumption at the end of each intervention day.

Figure 9: Schedule of the intervention day and timing for lunch group and no lunch group in the CoCo study.



4.2.2.3 Cognitive assessment

For the assessment of CF, a computerised test battery consisting of three tasks (ALA Institute) was used. Before starting, students had to pass a training phase with a task-by-task explanation by the study personnel and a short practise period. After this training and a 5-min break with low physical exercise, the actual cognitive testing began. Subjects were requested to respond as quickly as possible without sacrificing accuracy. The cognition tasks were applied in the following order: task switching, working memory updating and tonic alertness. After finishing the cognitive testing for two-thirds of the study sample, the school laptops had to be replaced by desktop computers for school intern reasons. The remaining participants were tested on the new computers. Consequently, the participants were either tested on the laptops or on the desktops. Therefore, calibration of the new computer was not necessary as the analyses of the intervention effects considered the individual differences between two tests days.

Task switching (switch)

The assesment of task switching in the CoCo-Study was processed exactly like the task switching in CogniDo PLUS (see chapter 4.1.3.3).

The outcomes were total reaction time for numbers (for items 2–26), total reaction time for letters (for items 2–13) and switch costs – that is, the processing time of the third section (switch; items 2–26) minus the first section (numbers; non-switch; items 2–26) minus the difference between the first twelve items of the second section (letter; non-switch; items 2–13) and the first twelve items of the first section (numbers; non-switch; items 2–13). To eliminate implausible data, we excluded all subjects with negative switch costs.

Working memory updating (2-back task)

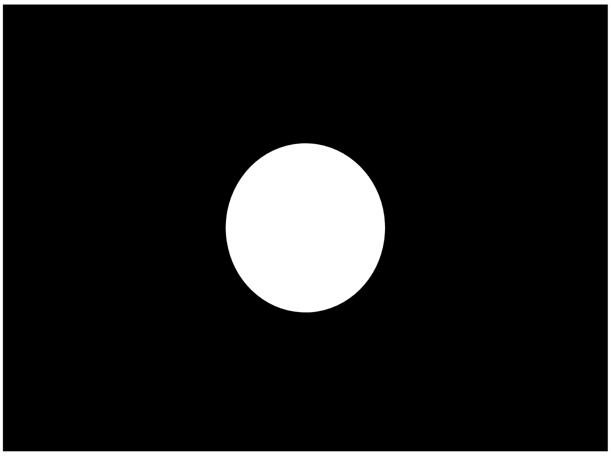
The 2-back task in the CoCo-Study was processed exactly like the 2-back task in CogniDo PLUS (see chapter 4.1.3.3).

The outcome variables were the ratio of missings (no reaction while reaction was required), the ratio of false alarms (reaction while no reaction was required) and the mean reaction time while reaction was required. Plausible measurements were defined as ratios of missing ≤ 50 %, false alarms ≤ 50 % and reaction times, between quartile 1 minus 1.5 times the inter- quartile range (IQR) and quartile 3 plus 1.5 times the IQR (i.e. ≥ 196.935 and ≤ 850.975ms).

Tonic alertness

To measure the level of tonic alertness, we used a simple reaction task. A white fixation cross was presented in the middle of a black screen. In a response stimulus interval of 3300 ms (± 20 %), a circle followed the cross and the subjects were supposed to press a predefined button as soon as the circle appears (maximal reaction time 1500 ms). The test included fifty items. The outcome variables were the mean reaction time (ms), the deviation of reaction time (ms), the number of omission errors (no reaction after appearance of the circle within 1500 ms) and the number of commission errors (reactions during the presence of the fixation cross). Plausible measurements were defined as reaction times ≥ 140.9 and ≤ 492.58 ms.

Figure 10: Cognitive assessment: Alertness. Screenshot of the task.



Children were instructed to press a predefined key as soon as the white circle appears on the screen.

4.2.2.4 Statistical analysis

All analyses were performed using the statistical software package SAS 9.2 (SAS Institute). $P<0.05$ was considered as statistically significant. Before conducting the statistical analyses, the raw data of all cognitive tasks were checked for plausibility. With regard to reaction times (working memory updating and alertness), plausible data were defined as values within the range of quartile 1 minus 1.5 times the IQR and quartile 3 plus 1.5 times the IQR [83]. For the 'ratios of false alarms/missings', a pre- defined limit of < 50 % error rate was defined as plausible as the expected error rate would be 50 % if a subject is choosing his/her reaction completely in random. Therefore, exclusion of error ratios ≥ 50 % was intended to reduce the risk of a systematic error in observations from single subjects as these values indicate a high risk that the task instructions were misunderstood (e.g. if a subject always pushed a button when no reaction was attended). Only plausible data were included in the analysis. The parameters of the three cognitive tasks were used as outcome variables. All outcome variables were interval scaled. As recommended by Grizzle [80], the sums of the two individual values of the particular outcomes variables of test days 1 and 2 were compared between groups using an unpaired t-test for normally distributed data (including normal distribution after transformation) and the Wilcoxon's ranked sum test for non- normally distributed data to examine potential carry-over effects. Non-normally distributed outcomes were transformed (log, square, root or reciprocal transformation) and analysed using unpaired t-test. If transformation did not result in normally distributed parameters, the non-parametric Wilcoxon's ranked sum test was used to analyse carry-over or intervention effects. As no carry-over effects were observed, results from both days were considered for the calculation of the treatment effect. In addition, linear regression analyses between the lunch size and the change in cognitive performance parameters (performance on lunch day and performance on no lunch day) were conducted for all parameters, including age and sex as additional covariables. For all parameters of the alertness task, for the parameter visual search of numbers in the task-switching task and the parameter ratio of false alarms (2-back task), the linear regression revealed non-normally distributed residuals (the association with lunch size was not significant for all of these parameters). As the linear regression might not be meaningful for these parameters, we decided not to present these results in Table 6.

4.2.3 Results

Participants

Of the 204 included participants, nineteen participants (one class) had to be excluded because of a 30-min delay of lunch on their 2nd test day. In addition, thirty-one children were absent on one of the 2 test days, resulting in 154 participants with complete data. Characteristics of the sample stratified by study groups L-NL (n=72), that is, having lunch on the 1st test day and skipping it on the 2nd test day, and NL-L (n=82) are shown in Table 5. There were no differences in age, sex or consumed amounts of the study meal between both groups. The majority of subjects regularly ate lunch at the school refectory (88.7 % in L-NL and 95.1 % in NL-L) with no significant difference between the groups (p=0.23).

Table 5: Characteristics of school children participating in the Cognition Intervention Study Dortmund Continued (CoCo) (n=154)

	L-NL (n=72)	NL-L (n=82)	p-value
Age, mean (SD) [years]	11.3 (0.7)	11.4 (0.6)	0.31 [a]
Female, n (%)	36 (50)	31 (37.8)	0.13 [b]
Regular lunch [c, d], n (%)	63 (88.7)	78 (95.1)	0.23[b]
Meal consumption (median; $P25^{th}/75^{th}$) [g]	360 (275.0/525.0)	375 (275.0/505.0)	0.87 [b]

L, lunch day. NL, no lunch day. Group NL-L skipped lunch during the first period, group L-NL skipped lunch during the second period. [a]Two sample t-test. [b]Chi-squared test/Fisher's Exact Test. [c] Defined as consuming lunch at the school refectory regularly by subscription. [d] missing data from 1 subject

Lunch effects on cognitive functions

Statistical analyses revealed no significant effects of lunch on the examined CF of task switching, working memory updating (2-back task) and alertness (P between 0.07 and 0.79, Table 7). The time for visual search of letters on the task- switching task showed a trend to slightly increase after having lunch (p=0.07). After additional exclusion of subjects who did not follow the study protocol (n=67, per protocol analysis), this trend disappeared (data not shown). The linear regression analysis revealed a significant negative association between the lunch size and the change in visual search of letters (β=-9.3, p=0.03 adjusted) in the task-switching task (Table 6).

Table 6: Linear regression of lunch weight on the change of main outcome variables of cognitive performance in school children (10-12 years) participating in the Cognition Intervention Study Dortmund Continued (CoCo) (adjusted for sex and age; only valid models)

Task	**Outcome**		**Lunch weight**	
		Intercept	**ß-value**	**p-value**
Switch	switch costs	-6754.0	10.966	0.226
	visual search of letters [a]	3635.7	-9.344	0.029
2-back	ratio of missings	23.3	-0.002	0.835
	reaction time	4.3	0.004	0.947

[a] first 12 reactions

Table 7: Effects of no lunch vs. lunch on cognitive performance in school children (10-12 years) participating in the Cognition Intervention Study Dortmund Continued (CoCo)

Task	Main outcome	No lunch			lunch			p-value
		Median	25th	75th	Median	25th	75th	
Switch (n=139)	switch costs[a] [ms]	23928	14990	38298	21475	13367	31864	0.26 [c]
	visual search letters[c] [ms]	26255	22579	31270	26958	22927	33928	0.07 [c]
	visual search numbers [ms]	43967	38007	50548	43854	38220	50782	0.36 [c]
2-back (n=87)	ratio of missings [%]	28.6	19.0	35.0	28.6	19.0	38.1	0.25[c]
	ratio of false alarms [%]	7.1	2.4	17.6	7.1	3.5	14.1	0.63 [c]
	reaction time [ms]	519.0	445.8	584.8	518.5	457.3	615.1	0.36 [c]
Alertness (n=148)	mean reaction time [ms]	306.0	273.1	343.9	314.9	270.9	357.0	0.12[c]
	deviation of reaction time [ms]	117.7	92.5	173.2	128.6	91.4	179.7	0.53[c]
	count of omission errors [N]	0	0	1	0	0	1	0.75[c]
	count of commission errors [N]	3	1	5	2	1	5	0.79[c]

[a] switch costs=(mean rt switch task) – (mean rt number task) – (mean rt first 12 reactions of letter task- mean rt first 12 reactions of number task); [b] first 12 reactions; [c] Wilcoxon rank-sum test

4.2.4 Discussion

The present study revealed no evidence for a lunch-related improvement or decline of cognitive performance in school- children in the early afternoon, about 90 min after finishing lunch. Although our previous studies CogniDo [49] and CogniDo PLUS [88] suggested slight improvements of single cognitive parameters shortly after lunch, the current study did not prove our hypothesis of potentially beneficial cognitive effects of lunch in the afternoon. Interestingly, the linear regression even indicated beneficial effects of lunch size as the individual change between lunch day and no lunch day decreased with larger lunch sizes for the parameter visual search letters. However, more information was needed for interpreting this result – for example, deviation from regular individual lunch size. It was not possible to obtain this information because of organisational reasons, but this question would be interesting for future studies.

In the earlier CogniDo study [49] (n=105), the participants made significantly more omission errors in the tonic alertness task on the no lunch day compared with the lunch day (p=0.03). The CogniDo PLUS study [88] (n=195) suggested slightly lower levels of false alarms in the task regarding working memory updating when lunch was eaten compared with the test condition in which lunch was omitted (p=0.01) – that is, after eating lunch, children responded less frequently to non-targets, to which they should not have responded. Both tasks (tonic alertness and working memory updating) of the previous studies were also conducted in the present study without any hints at beneficial effects. A potential explanation for these divergent study results could be the difference in the time span between lunch and cognitive testing, which was about 45 min after finishing lunch in the former studies, but twice as long with 90 min in the current study. A comparison between the variable values of the present study and the previous CogniDo [49] and CogniDo PLUS [88] studies shows that the values are located in the same data range. Even though the data were not conducted from the same probands, they seem to be comparable. Considering these results, it might be hypothesised that children's cognitive performance may slightly increase immediately after lunch and may not improve when the fasting period is extended into the early afternoon.

Although the reasons for differences in lunch effects depending on the interval until cognitive testing could not be examined in our studies, one plausible explanation may be the course of blood glucose levels. Glucose levels increase in the early postprandial period, but might have been on a decrease at the time when cognitive performance was tested in the present CoCo study [91]. It could be speculated that an increase in blood glucose is beneficial for cognitive performance, whereas a decrease in glucose might attenuate this effect despite higher levels than in the fasting condition. Studies that investigated glucose uptake showed that the resulting increase in blood glucose levels enhances CF such as memory [31,52] or sustained attention [34]. For example, Owen et al. [31] demonstrated that a glucose dose of 25 g enhanced working memory performance following a 2-h fast, and Benton et al. [34] showed that 25 g glucose as a drink improved sustained attention. However, effects of oral glucose dosage may differ depending on blood glucose resources or the level of depletion of glucose reservoirs, for example, in the liver [31]. Furthermore, tests with pure glucose consumption may not simply mirror the effects of a whole meal as applied in our study. Sugars or other carbohydrates as part of a mixed meal increase blood glucose levels more slowly than pure glucose [35,36]. Therefore, future studies should assess the role of lunches differing in their glycaemic response on cognitive performance in children and adolescents.

Another influencing factor on cognition could be the post- lunch dip phenomenon that may relate to the timing of the meal and the interval until measurements. In adults, a decline in cognitive capabilities was observed in a wide range of about 60–120 min after lunch [25,61,63]. In our previous studies, cognitive parameters were measured 45 min after lunch, without any evidence of a post-lunch dip. It remains an open question whether a meal-enhanced post-lunch dip in children exists as has been suggested for adults. As a post-lunch dip was mostly seen in sustained attention tasks [28,29], it might especially be detectable in the alertness task (which includes testing for sustained attention). As our results did not show a significant difference between the L and the NL condition, it could be assumed that there might not be a post-lunch dip in children. However, to answer this question conclusively, further studies will be needed.

There are several characteristics of the CoCo study design that need to be discussed. The study was not conducted under clinical conditions, but tested the students in real-life conditions in their classroom setting. Accordingly, factors apart from lunch such as environmental stress (e.g. noise, peer group actions) might have influenced individual cognitive performance [92]. Especially auditory distraction can have detrimental effects [93,94] on the cognitive performance in children. Even though the study team tried to keep the children in the testing room as quiet as possible, environmental disturbances could partly have masked acute individual effects of lunch. Clinical studies might be more suitable to clearly identify isolated lunch effects, but do not allow any conclusions on the practical meaning of these results in children's everyday life. In addition, the crossover design of the CoCo study should have minimised potential effects of individual confounding in the total sample. To counteract a possible learning effect, we used a parallel version of the cognitive task in which the task switching and the 2-back sequences differed (task switching task differed in position of the items, 2-back task in sequence of fruit and vegetable items). In addition, we conducted a training phase immediately before the actual testing to ensure that every subject understands the tests already before the first assessment. The task-switching task and the 2-back task, which were already used in our previous study, were originally designed for adults. Although we adapted these tests for children and tested the entire test battery in a pretest with children of the same age in another school in order to avoid a very sophisticated test, the rate of implausible 2-back test results might indicate a floor effect in the same subjects.

In the CoCo study, no pre-lunch performance was measured as opposed to laboratory studies. However, we decided against this practice as we were worried about potential negative effects on the motivation with increasing numbers of tests. If we had included a pre-lunch measurement, the children would have had to complete four tests on 2 test days within a week, and without a pre-lunch testing only two. Negative influences on motivation might not only have impact on the results of the cognitive tasks, but could also result in a high rate of dropouts. Therefore, we decided, for this study, to focus on the after-lunch condition.

Another limitation of the CoCo study is that it was not possible to use a double-blind design with a placebo condition. This leaves the possibility of subject and experimenter bias and is a common problem in food-based trials. However, the randomised, crossover design eliminates variations between the subjects and reduces bias. In the present study, fasting was the control condition. However, fasting could also be viewed as intervention as the majority of students regularly eat lunch at school. In a recent review, which compared ten studies of adults for the impact of short-term fasting on cognition [95], results were equivocal. Although some studies showed no effects of fasting on cognition, others showed impairments in tasks related to psychomotor speed, mental rotation or executive function. However, these results are not transferable to the current study because of differences in cognitive tasks, fasting periods, time of fasting, time of day and the age of the participants.

Müller et al. [30] concluded that lunch effects in studies of adults might have been modified by the fact that the test meal size was larger or smaller than the usual lunch size. Although the test meal size was assessed in the CoCo study, it was not possible to assess probable differences with respect to the individual usual lunch size as well. Hence, future studies on cognitive effects of lunch should assess the usual size of lunch to examine the impact of deviation of the test lunch from habitual eating lunch size (smaller or larger) on cognitive performance.

A considerable number of participants did not completely comply with the study protocol (n=67). Reasons for this behaviour were not enquired, but it could be assumed that it was difficult for the children to restrain from eating during this time period, especially when it was explicitly forbidden. Apart from the questionnaire at the end of the test day and supervision in the school yard, children had opportunities for hidden snacking. Thus, it could not be fully ruled out that all participants who did not comply with the protocol were detected. In addition, it has to be mentioned that the sixth grade students of this intervention already participated as fifth grade students in the previous intervention. Therefore, they may have been familiar with two of the three cognitive tasks and were probably less motivated. However, any such effects should have been minimised as the differences between the test days were analysed in the crossover design.

In conclusion, the present study confirmed the results of our previous studies [30,88] that school lunch does not seem to impair children's CF regarding the conducted tests on task switching, working memory updating and alertness. Although the previous studies [49,88] pointed to slight improvements in single cognitive parameters by lunch shortly after the meal, the current study did not indicate positive effects of lunch on cognition after a prolonged interval until early afternoon.

5 General Discussion

5.1 Research aims

In this chapter, the results are summarized and discussed with respect to the research questions (see chapter 2).

Research aim 1

This section addressed the question of potential short-term effects of school lunch on children's EF as measured at the beginning of afternoon lessons (**RQ1**) or in the course of the afternoon lessons (**RQ2**). To investigate this research aim two randomized crossover intervention studies, CogniDo PLUS [88] (RQ1) and CoCo [91] (RQ2) were conducted (see chapter 2.1).

RQ1 (CogniDO PLUS) examined the cognitive parameters task switching (switch), updating working memory (2-back task), and inhibition (flanker task) in 195 pupils on two different days, on which each of the pupils were tested once: 45 minutes after having lunch (lunch; L) and after not having lunch (no lunch; NL). Data analyses revealed that levels of false alarms in the 2-back task were lower in the test situation when lunch was eaten than those when lunch was not eaten, i.e. children responded better to non-targets after eating lunch. No significant influence of lunch consumption was shown in the switch or flanker tasks. However, in the flanker task the students tended to make fewer errors in the L condition than in the NL condition. Taken together these results suggest that lunch could have a beneficial, but at best a small, influence on working memory around 45 minutes after meal consumption.

RQ2 (CoCo), an extension of CogniDo PLUS [88], examined cognitive effects in children 90 minutes after L and NL. The tasks were the same as those in CogniDo PLUS except for the flanker task, which was replaced by a tonic alertness task. Unlike CogniDo PLUS no significant differences between the lunch situations were observed either in the switch or 2-back task 90 minutes after lunch. Also, the alertness task did not reveal any significant differences, a finding which is in contrast to the first study of the workgroup's CogniDo-series, the CogniDo study [49]. In that study the participants showed significantly fewer omission errors in the alertness task in the lunch condition compared to the no lunch condition when tested 45 minutes after lunch.

A potential explanation for these divergent results could be the difference in the time span between lunch and cognitive testing, which was about 45 minutes in the former studies, but 90 minutes in the latest CoCo study [91]. It might be hypothesized that children´s cognitive performance may increase immediately after lunch and may decrease or remain constant if the fasting period is extended into the early afternoon. Vaisman et al. [96] reported similar findings after they conducted a breakfast intervention study of 11- to 13-year-old school children. At baseline (before the test phase began) the children were asked if they regularly skip or eat breakfast and the subjects' cognitive performance were tested. Subsequently, two thirds of the children received a standardized breakfast at school for 14 days, after which their cognitive performance was tested again. The results indicate that routinely eating breakfast at home 2 hours prior to cognitive challenges does not improve cognitive functions in 11- to 13-year-old children. However, food ingestion at school 30 minutes prior to testing did improve the test results compared to those of the children who eat at home and the children who do not eat breakfast. Vaisman et al. [96] suggest that the time between eating and the measurement of cognitive abilities may influence the outcome of studies that investigate the effects of breakfast on behavior.

Another possible explanation for the differing results between the CogniDo studies [88,91] may be the changing course of blood glucose levels. In the early postprandial period glucose levels tend to increase and then decrease. It could be hypothesized that such a rise in blood glucose is favorable for cognitive performance, while a decline may diminish this favorable effect despite levels higher than those in the fasting condition. Studies that investigated pure glucose uptake showed that the resulting increase of blood glucose levels enhances cognitive functions such as memory [31,52] or sustained attention [34]. However, tests with pure glucose consumption may not mirror the effects of a whole meal such as those applied in the present studies. Furthermore, this explanation is speculative since blood glucose responses to the test meal were not examined in CogniDo PLUS [88] and Coco [91]. Additionally, the results from CogniDo PLUS [88] and Coco [91] studies were not directly compared with statistical analyses, so no definitive statements could be made.

Moreover, the statistical power could be another reason for these partially divergent results. Since the CoCo study [91] had fewer participants than CogniDo PLUS [88], the statistical power may have not been sufficient enough to detect small differences such as the case of CogniDo PLUS [88].

The findings in the recent study are not in line with findings in adults, which point to impairments of some cognitive functioning parameters after eating lunch (see chapter 1) [25,26,63]. Various reasons for these differences are conceivable and could be addressed in future studies. For instance, hormonal and metabolic differences between children and adults may have a significant influence.

In the case of adults it is indicated that lunch size plays an important role for cognitive performance. Studies of adults showed that a larger test lunch size than the habitual lunch size could impair cognitive performance [25,28,29]. In the two intervention studies presented, which were conducted at a school setting, actual lunch size was measured and evaluated in the regression analyses. In CogniDo PLUS [88] (median of lunch size: 335-345 g for the respective task) and CoCo [91] (median of lunch size: 360- 375 g for the respective intervention sequence) linear regressions of lunch size and the outcomes were conducted. In CogniDo PLUS [88] a (non-significant) trend was found between switch costs and lunch size (ß=-18.51 and p=0.083 and ß=-18.95 and p=0.078 adjusted for sex and age), i.e. the children with a large lunch tended to have lower switch costs. In CoCo [91], there was a negative association between lunch size and performance in the visual search for letters in the switch task (p=0.03 and ß=-9.3 unadjusted and adjusted), i.e. children who ate a larger lunch performed faster. These results suggest that a bigger lunch might have favorable, although small effects on children's cognitive performance. These findings contrast the findings in adults, where a large lunch was shown to negatively impact cognitive performance. However, more information is needed to interpret these results e.g. a deviation from regular individual lunch size in the CogniDo studies [25,28,29].

However, it could be possible that the children's eating behavior during the testing was different than usual because of the unusual situation of the intervention and an awareness of the fact that the lunch weight was measured. Since the volume of lunch was ad libitum and not standardized no valued statement could be made about the lunch size of the school lunch and its effects on children's cognitive performance. Additionally, due to organizational reasons it was not possible to assess probable differences between the individual usual lunch size and the study lunch size in both studies, which would be important for interpreting the results [25,28,29] and interesting for future studies.

In conclusion, no negative effects of school lunch were observed in the conducted studies- neither 45 nor 90 minutes after lunch, but small positive effects were indicated by improvements in the updating working memory 45 minutes after lunch. However, it is questionable whether or not this effect is relevant to daily performance at school. Because most values of the cognitive variables measured in CogniDo PLUS [88] and CoCo [91] were in the same range, it could be assumed that there may be no large difference in cognitive lunch effects between the times of testing. However, more studies are necessary to be able to answer these questions conclusively.

Research aim 2

The second research aim addressed possible mechanisms for a lunch-related short-term change in performance, i.e. whether or not a postprandial cortisol increase is a mediator for potential lunch-related changes in cognitive performance (**RQ3**) or if school lunch exacerbates a potential post-lunch dip in children (**RQ4**).

RQ3 was investigated as part of the CogniDo PLUS study [88]. Saliva samples were collected before lunch (T1=12:20 hours) and again about 45 min after starting lunch (before cognitive assessment at 13:10 hours=T2). The results showed that lunch induced a cortisol increase, whereas in the NL condition cortisol levels decreased from T1 to T2. These findings are congruent with the work of other authors, who found that lunch induced a cortisol increase in children as in adults [72–76].

In CogniDo PLUS [88] a significant negative linear association was found between the postprandial cortisol increase and the ratio of false alarms in the 2-back task (p=0.04), i.e. a higher postprandial cortisol increase was associated with fewer false alarms. It can be assumed that a physiological increase in cortisol, as observed in the CogniDo PLUS study [88], enhances the updating working memory function, whereas studies with high pharmacological doses showed detrimental effects on cognitive performance [64–66]. Support for a hypothesis of a dose-dependent effect on working memory is provided by Lupien et al [67,68], who suggest that a low dose of pharmaceutical cortisol improves the processing capacity of working memory, whereas a high dose causes impairment. In line with this, a quadratic function (inverse U-shape curve) has been suggested between performance on working memory tasks and changes in glucocorticoid levels after a hydrocortisone infusion [68]. Additionally, it has been ascertained that the working memory is susceptible to a (pharmaceutical) increase cortisol because glucocorticoid receptors are present in the area of the brain where the working memory is located [70]. The question remains, to what extent does a physiological food- and meal-induced increase of cortisol enhance the working memory as effectively as a pharmaceutical dose. A study of young adults, in which a dose of 20 or 40 mg of cortisol given previous to testing caused fewer errors in commission i.e. false alarms, showed that even moderate doses of cortisol can affect working memory [69]. Similarly, in the CogniDo PLUS study [88] a physiological increase of cortisol after lunch reduced the number of false alarms in the 2-back task. After dividing the subjects into two groups (high and low postprandial cortisol increase), improvements in working memory updating after lunch were only observed in the high postprandial increase group. So it can be cautiously assumed that a physiologically induced cortisol increase after lunch may improve the working memory updating function in children, which reflects a beneficial mediating effect of cortisol. No conclusions can be drawn, however, about the amount of the increase or the exact level at which improvements or impairments occur.

The magnitude of the cortisol response could depend on the amount of lunch eaten. A linear regression analysis revealed a significant positive relationship between lunch sizes and cortisol responses (not adjusted model: ß=0.0025, p=0.0081; adjusted model: ß=0.0024, p=0.0113). One explanation as to how lunch could influence the cortisol level could be found in the blood glucose response and the GI of the meal [86]. In an intervention study of children, it was observed that a high GI breakfast increased the blood glucose as well as the cortisol level, which resulted in a better performance on a vigilance task. The authors hypothesized that higher blood glucose levels could activate the hypothalamic –pituitary–adrenal axis in stressful situations (i.e. test situations) leading to higher cortisol levels and higher tension in the participants before the tests and thus better performance on tasks where information processing is tested. More studies of children are needed to investigate the metabolic pathway and effect of postprandial cortisol levels on cognitive performance.

Overall, the CogniDo PLUS [88] results suggest that a physiological increase in cortisol induced by lunch could enhance the student's updating working memory functions. However, to support this finding and to learn about the amount of the increase or the exact level at which cognitive improvements or impairments occur, more studies are needed.

RQ4 of the CogniDo PLUS [88] and CoCo [91] study addressed the question whether or not school lunch exacerbates a potential post-lunch dip in children. The post-lunch dip is a circadian phenomenon in adults. It occurs naturally around midday and may be worsened by lunch intake. The dip in performance is assumed to begin approximately one hour after lunch began with data suggesting that performance begins to recover approximately two hours after lunch [25,61,63]. In CogniDo PLUS [88] (as well as in the former CogniDo study [49]) cognitive performance was measured 45 minutes after lunch, and declines in performance were observed when lunch was skipped. Based on these two studies it could not be ruled out that a post-lunch dip might occur in the later afternoon. In the CoCo study [91] cognitive performance was measured 90 minutes after lunch and no detrimental effects of lunch were seen. The assumption that skipping lunch might result in alleviation or the prevention of this post-lunch dip could not be proven, since there were no indications of improved test results when lunch was skipped. Considering that a post-lunch dip in adults was mostly observed in tasks, involving sustained attention, a post-lunch dip was to be expected in the alertness task in the CoCo [91] and CogniDo [49] studies. However, neither of the studies showed a significant decline in performance measured in the alertness task after lunch.

Considering the results of both studies, there was no indication of a post-lunch dip in children in the early afternoon. In children a circadian dip that may occur regardless of food intake cannot be ruled out, since the studies only investigated the effect of lunch. Additionally, no pre-lunch measurement (for example in the morning) was taken. Because the results did not show a significant impairment after lunch in the tasks conducted, it can be assumed that lunch does not exacerbate the phenomenon of a post-lunch dip in the case of children that like seen in adults. However, it cannot be fully ruled out that children do experience a post-lunch dip like adults do. Regarding the reasons for the divergent findings in children and adults, it could be speculated that due to a different metabolic and hormonal status, children are not as susceptible to a lunch-related dip. Another hypothesis is that the new, exciting testing situation may have compensated for a potential dip in performance after lunch.

5.2 Strength and limitations

Several methodological challenges and strengths need to be addressed. Since both studies followed the same methodical procedures, first considerations that apply to both studies are discussed and then noticeable considerations unique to each study.

Crossover design

One strength of both CogniDo studies [88,91] is the crossover design. The advantage of this design is that each subject serves as his or her own control, and thus the influence of confounding covariates is reduced. There is no need to question the comparability between intervention and control group, and confounders such as sex and age can be eliminated from the beginning [79]. Additionally, due to its statistical efficiency fewer subjects are needed than in non-crossover designs [79]. The statistical analysis recommended by Grizzle [80] included the consideration of the order sequence of the treatment and allowed for the detection of a possible carry-over effect (which was not seen in either study). To guarantee a reliable analysis of the intervention effects a washout phase between the intervention periods was ensured. As it was not possible to perform an interaction test between lunch size and single cognition outcome variables with this model, this interaction was tested with linear regression models.

Study environment

Neither CogniDO PLUS [88] nor CoCo [91] was conducted under clinical conditions, but rather in a real-life environment. A clinical setting would have been more suitable to investigate the isolated effect of lunch on children's cognitive performance. However, in such a setting it would not be possible to draw transferable conclusions on children's everyday life. The purpose of these studies was to test the cognitive effects in real-life conditions in order to identify any (school-) lunch related changes in the school setting. In a real-life setting factors other than lunch e.g. environmental stress (noise, peer group actions) might have an influence on cognitive performance [92]. The classroom situation during testing in both studies may be an explanation for the high count of implausible outcomes. Due to the limiting values in the inhibition (CogniDo PLUS [88]) and working memory updating task (both studies), up to 42 % of the students were excluded per task because of an error rate over 50 % or an implausible reaction time. It can be assumed that the high error rate resulted from trade-off between accuracy and speed because the participants who were excluded partially showed relatively short reaction times. Another explanation could be the noise in the classroom since auditory distraction can have detrimental effects [93,94] on the cognitive performance in children. Even though the study team cared to provide a silent testing room, possible environmental disturbances could have partly masked acute individual effects of lunch.

For these reasons a clinical setting may have been better suited to investigate isolated cognitive performance alone. However, a typical clinical study situation is well structured, with one examiner and one test subject within a quiet environment and limited distractions. Therefore, is unlikely to be representative of home, classroom, or social environments as this one-to-one environment is rarely available in real-life settings and could under some circumstances even enhance motivation and performance [77]. Additionally, a clinical setting could mask children's deficits in inhibition, attentive control, and flexibility due to an examiner who provides clear directions [77]. The schedules of the CogniDo interventions were embedded in the typical school day with cognitive tasks performed at the usual start of the afternoon lessons. Therefore, it is possible to transfer the results to school performance in the afternoon lessons. In addition, the crossover design of the studies should have minimized potential effects of individual confounding in the total sample.

Pre-lunch testing

In the recent study design, the study team decided against a baseline pre-lunch measurement. It was expected that an increased number of tests could have potential negative effects on the participants' motivation. If a pre-lunch measurement would have been included, the children would have had to complete 4 tests on 2 test days within 1 week - without a pre-lunch testing only 2. Any negative influence on the motivation might not only have impacted the results of the cognitive tasks, but could have also resulted in a high rate of dropouts. Therefore, this study purely focused on the after-lunch condition, and it was not measured if the children perform better after lunch time as compared to before. Only by comparing the same children once after having lunch and once with the situation of no lunch, could an endogenous change in cognitive performance be ruled out [62].

Food-based considerations

CogniDo PLUS [88] and CoCo [91] were food-based trials with two conditions: lunch and no lunch. In this kind of study, it is a common problem that it is not possible to use a double-blind design, because it is obvious which treatment is administered since there is no placebo for the no lunch condition. This allows for the possibility of subject and experimenter bias. However, the randomized crossover design eliminates variations between the subjects and therefore reduces bias.

Müller et al. [30] concluded that lunch effects in studies of adults might have been modified by the fact that the test meal size was larger or smaller than the usual lunch size. In the present work the lunch weight was assessed. Linear regression analyses pointed to positive associations between lunch size and some parameters of cognitive performance. However, no conclusions could be made as to whether the test lunch was larger or smaller than the children's usual lunches and what influence individual lunch size has on cognitive performance. To be able to draw such conclusions it would be necessary to assess the children's usual lunch weight as an average of several usual days prior to the intervention. This was not possible for organizational reasons. It is possible that due to the test situation and the special attention to the lunch size on the test day that some children ate more or less than their regular lunch size. Hence deviations from usual lunch sizes could be a future focus in studies examing lunch effects on cognitive performance in childhood.

Compliance

A considerable number of participants did not completely comply with the study protocol (CogniDo PLUS [88]: n=26; CoCo [91]: n=67), i.e. they ate something or drank a caloric, sweetened beverage when they were supposed to abstain from eating. Reasons for this behavior were not individually inquired, but it could be assumed that it was difficult for some of the children to refrain from eating for the period of time, especially when it was explicitly forbidden. Particularly in the CoCo study [91], the non-compliance rate was about 2.5 times higher than in CogniDo PLUS [88] which suggest that the longer time interval to the next meal was also hard to comply with. Although the children were required to answer a questionnaire on their food and drink consumption during the day at the end of the test day and although the study personnel supervised schoolyard on the test days, there may have been opportunities for hidden snacking. It could not be fully ruled out that all participants who did not fully comply with the protocol were detected. However, the exclusion of the children with obvious incompliance from the statistical analysis did not alter the results.

Test conditions

Another point to be considered is that the 6th grade students in the CoCo study [91] were the 5th grade students of the CogniDo PLUS [88] study a year earlier, so they were familiar with two of the three cognitive tasks and may have been less motivated. However, any such effects should have been minimized because the analyses only assessed the individual differences between the test days crossover study.

To counteract a possible learning effect, parallel versions of the cognitive tasks were used. In the switching task, the position of the items differed, and in the 2-back task the sequence of fruit and vegetable items changed.

In order to ensure that every pupil understood the tasks, a training phase was conducted immediately before the actual testing session. The switching task and the 2-back task were originally designed for adults. Although they were adapted to children and tested with children of the same age in another school before the field periods of the studies in order to avoid the use of too highly sophisticated tasks, the relatively high rate of implausible results in the 2-back test might indicate a floor effect for some participants. It can be assumed that it was hard for the children to keep up for the duration of the 2-back task. However, to avoid ceiling effects and to be able to detect even small effects of lunch the 2-back task was not modified further, e.g. shortened.

Cortisol

In CogniDo PLUS [88], for practical reasons (e.g. the need for extra medical staff, probable cortisol increase because of the stress of blood sampling) postprandial cortisol was not assessed using blood sampling, but instead with saliva samples. Saliva cortisol levels are a valid reflection of the unbound hormone in blood [82]. Due to a lack of reference values for saliva measurements in children, it was not possible to categorize the cortisol values as high or low [87]. However, because the individual changes of cortisol levels were considered, the absolute values of cortisol were of minor importance in the CogniDo PLUS study [88].

5.3 Practical implications and public health relevance

Throughout the last decade the number of all-day schools in Germany increased. In Germany, like in many other countries, it is mandatory to offer a midday meal in schools with a day-long schedule [97]. With 2.7 million students attending all-day schools in 2014 nationwide, school lunch is an issue of interest for public health [97].

Under these circumstances schools are a perfect setting for prevention measures regarding nutrition. Lunch provides an important and specific portion of the daily nutrient intake. Recommendations for the composition of school lunch are available in Germany such as the "Quality standard for school meals" developed by the German Nutrition Society (DGE) and the concept of the "Optimized Mixed Diet" from the Research Institute of Child Nutrition (FKE). These guidelines were developed to ensure an adequate long-term supply of nutrients. They were not created with the explicit aim to immediately support or improve school performance, although short-term effects are often implicitly expected when a school-lunch program is established. Nevertheless, with the expansion of all-day schools students are challenged to maintain their attention spans and cognitive performance during the afternoon lessons, and the short-term effects of school lunch enter the focus of interest.

The crossover studies conducted and described in this thesis examined the short-term effects of school lunch on children's cognitive performance. The results indicate no negative short-term influence of school lunch on cognitive performance in children as might have been assumed based upon studies in adults. 45 minutes after lunch, a point that usually collides with the start of the afternoon lessons, the updating working memory and alertness function were even significantly improved (CogniDo PLUS [88] and CogniDo [49]), but these results were not reproducible when the time span was doubled to 90 minutes after lunch (CoCo [91]). The positive effects seen after 45 minutes were minimal, and it is questionable whether they would make a considerable difference in school performance. However, overall the results indicate that a lunch-induced worsening of a potential post-lunch dip in children does not seem to exist. This could be an argument in favor of the daily consumption of school lunch. If so, lunch should conform to the existing recommendations for its basic composition (DGE/ FKE). Childhood and adolescence are critical developmental periods, in which a diet of high nutritional quality is particularly important [98].

Overall, considering that the intervention studies presented here are the first to examine lunch and cognitive functioning in children, it is not yet possible to conclude any reliable practical implications. More studies are necessary to prove the effects of lunch regarding the short-term effects on cognitive performance in children. Most adults experience an impairment of cognitive performance after lunch, but it can be argued that such a phenomenon was not observed in the children, at least based on the studies conducted in the recent research. The results of this thesis could contribute to an argument in favor of maintaining and establishing the regular provision of school lunch.

5.4 Conclusion and Perspectives

This thesis and the previous study in the CogniDo project give the first insights into the short-term effects of school lunch on cognitive performance in children during afternoon lessons. The cognitive functions examined were the three executive functions task switching, updating working memory, and inhibition and tonic alertness as a basal cognitive parameter. The results tend to indicate beneficial but small short-term effects of lunch at the beginning of afternoon lessons. The updating of working memory showed to be significantly improved and the inhibition tended to be better. Additionally, the alertness function was significantly improved after lunch in the beginning of the afternoon lessons in a previous study with children making less omission errors [49]. However, these effects were not observed, when the participants were tested later in the course of the afternoon lessons. Nonetheless, the exhibited improvements shortly after lunch were very small with a difference of one percent in the rate of false alarms in the updating working memory task between lunch and no lunch. It is questionable if such minimal effects are relevant for every day performance. Overall, school lunch does not seem to have any negative impact but rather positive consequences shortly after lunch and neither positive nor negative effects in the course of the afternoon.

Since these results are in contrary to studies in adults, in which a lunch worsened the post-lunch dip and thereby the cognitive performance, the results of this work could serve as an argument in favor of school lunch. In this work there was no indication of a post-lunch dip in children in the afternoon. Although a circadian dip in children, which may occur independently from eating, cannot be ruled out, the results indicated no negative short-term consequences of school lunch on cognitive performance in children. Furthermore, the results suggest that the physiological postprandial increase in cortisol might be associated with lunch size and could enhance the function of updating working memory. No conclusions can be drawn, however, about the amount of the increase or the exact level at which improvements or impairments are to be expected. To support this finding more studies will be needed.

Although findings from the two studies within this thesis and from a previous study do not indicate any detrimental short-term effects of school lunch, no definite conclusions can be drawn considering the small number of studies focusing this topic. Cognitive performance results seem to be partially dependent on environmental influences, such as the sensitivity and suitability of the chosen tests [99], the study panel, the nutritional factor tested, the culturally appropriate implementation of the tests, and the setting in which they are applied [4,99]. Therefore, the results need to be interpreted with caution before they are transferred to other groups i.e.

younger children or adolescents. In the real life setting, in which the present studies were conducted, other factors probably had more influence on cognitive performance than lunch.

However, there are few existing studies on the correlation between lunch and cognition in children, and more studies in this important field of public health and nutritional research are needed. The composition of lunch and lunch size may be important for cognitive performance [30]. Consequently, the CogniDo research series' next step is to consider the composition of lunch in the 'Cognition Intervention Study Dortmund Glycemic Index (CogniDo GI)'. The results of this thesis together with the results of the ongoing study and others future studies will contribute towards uncovering the relationships between lunch and cognitive performance in children.

6 References

1. Casey BJ, Galvan A, Hare TA. Changes in cerebral functional organization during cognitive development. *Curr Opin Neurobiol*. 2005;15(2):239-244. doi:10.1016/j.conb.2005.03.012.
2. Casey BJ, Giedd JN, Thomas KM. Structural and functional brain development and its relation to cognitive development. *Biol Psychol*. 2000;54(1-3):241-257. doi:10.1016/S0301-0511(00)00058-2.
3. Benton D. The influence of children's diet on their cognition and behavior. *Eur J Nutr*. 2008;47 Suppl 3:25-37. doi:10.1007/s00394-008-3003-x.
4. Hughes D, Bryan J. The Assessment of Cognitive Performance in Children: Considerations for Detecting Nutritional Influences // The Assessment of Cognitive Performance in Children: Considerations for Detecting Nutritional Influences. *Nutr. Rev*. 2003;61(12):413-422. doi:10.1301/nr.2003.dec.413–422.
5. Fuster JM. Frontal lobe and cognitive development. *Journal of Neurocytology*. 2002;31:373-385. Accessed June 29, 2016.
6. Lenroot RK, Giedd JN. Brain development in children and adolescents: insights from anatomical magnetic resonance imaging. *Neuroscience and biobehavioral reviews.* Updated 2006:718-729.
7. Shutterstock (shutterstock_139691548) processed by kreativrudel Bonn after Oates, John, Karmiloff-Smith, Annette and Johnson, Mark eds. (2012). Developing Brains. Early Childhood in Focus, 7. Milton Keynes: The Open University. https://www.shutterstock.com/pic-139691548.html.
8. Sturm W, Simone A de, Krause BJ, et al. Functional anatomy of intrinsic alertness: evidence for a fronto-parietal-thalamic-brainstem network in the right hemisphere. *Neuropsychologia*. 1999;37:797-805. Accessed June 30, 2016.
9. Sturm W, Longoni F, Fimm B, et al. Network for auditory intrinsic alertness: A PET study. *Neuropsychologia*. 2004;42(5):563-568. doi:10.1016/j.neuropsychologia.2003.11.004.
10. Sturm W. *Manual: Wahrnehmungs- und Aufmerksamkeitsfunktionen: Alertness*. [Kurzbezeichnung WAFA]. Mödling. 2012.
11. Miyake A, Friedman NP, Emerson MJ, Witzki AH, Howerter A, Wager TD. The unity and diversity of executive functions and their contributions to complex "Frontal Lobe" tasks: a latent variable analysis. *Cogn Psychol*. 2000;41(1):49-100. doi:10.1006/cogp.1999.0734.

12. Ardila A. On the evolutionary origins of executive functions. *Brain Cogn*. 2008;68(1):92-99. doi:10.1016/j.bandc.2008.03.003.
13. Diamond A. Executive functions. *Annu Rev Psychol*. 2013;64:135-168. doi:10.1146/annurev-psych-113011-143750.
14. Rogers RD, Monsell S. Costs of a predictable switch between simple cognitive tasks. *Journal of Experimental Psychology: General*. 1995;124(2):207-231. Accessed June 30, 2016.
15. Lehto J. Are Executive Function Tests Dependent on Working Memory Capacity? *The Quarterly Journal of Experimental Psychology Section A*. 1996;49(1):29-50. doi:10.1080/713755616.
16. Morris N, Jones DM. Memory updating in working memory: The role of the central executive. *British Journal of Psychology*. 1990;81:111-121. Accessed June 30, 2016.
17. Tillman C, Brocki KC, Sørensen L, Lundervold AJ. A Longitudinal Examination of the Developmental Executive Function Hierarchy in Children With Externalizing Behavior Problems. *Journal of Attention Disorders*. 2013. doi:10.1177/1087054713488439.
18. Diamond A, Lee K. Interventions shown to aid executive function development in children 4 to 12 years old. *Science*. 2011;333(6045):959-964. doi:10.1126/science.1204529.
19. Thompson-Schill SL, Ramscar M, Chrysikou EG. Cognition without control: When a little frontal lobe goes a long way. *Curr Dir Psychol Sci*. 2009;18(5):259-263. doi:10.1111/j.1467-8721.2009.01648.x.
20. Davidson MC, Amso D, Anderson LC, Diamond A. Development of cognitive control and executive functions from 4 to 13 years: evidence from manipulations of memory, inhibition, and task switching. *Neuropsychologia*. 2006;44(11):2037-2078. doi:10.1016/j.neuropsychologia.2006.02.006.
21. Turner GR, Spreng RN. Executive functions and neurocognitive aging: dissociable patterns of brain activity. *Neurobiol. Aging*. 2012;33(4):826. doi:10.1016/j.neurobiolaging.2011.06.005.
22. Chugani HT. A critical period of brain development: studies of cerebral glucose utilization with PET. *Prev Med*. 1998;27(2):184-188. doi:10.1006/pmed.1998.0274.
23. Pollit E. Does Breakfast Make a Difference in School? *Journal of the American Dietetic Association*. 1995;95(10):1134-1139. doi:10.1016/S0002-8223(95)00306-1.
24. Guiney H, Machado L. Benefits of regular aerobic exercise for executive functioning in healthy populations. *Psychon Bull Rev*. 2013;20(1):73-86. doi:10.3758/s13423-012-0345-4.

25. Craig A, Baer K, Diekmann A. The effects of lunch on sensory-perceptual functioning in man. *Int Arch Occup Environ Health*. 1981;49(105-114). doi:10.1016/j.csm.2004.12.002.
26. Smith AP, Miles C. Effects of lunch on selective and sustained attention. *Neuropsychobiology*. 1986;16(2-3):117-120.
27. Kanarek RB, Swinney D. Effects of food snacks on cognitive performance in male college students. *Appetite*. 1990;14(1):15-27.
28. Craig A, Richardson E. Effects of experimental and habitual lunch-size on performance, arousal, hunger and mood. *Int. Arch Occup Environ Heath*. 1989;61(5):313-319. doi:10.1007/BF00409386.
29. Smith A, Ralph A, McNeill G. Influences of Meal Size on Post-Lunch Changes inPerformance Efficiency, Mood, and CardiovascularFunction. *Appetite*. 1991;(16):85-91. Accessed April 12, 2016.
30. Müller K, Libuda L, Terschlüsen AM, Kersting M. A Review of the Effects of Lunch: On Adults' Short-term Cognitive Functioning. *Canadian Journal of Dietetic Practice and Research*. 2013;74(4):181-188. doi:10.3148/74.4.2013.181.
31. Owen L, Scholey AB, Finnegan Y, Hu H, Sünram-Lea SI. The effect of glucose dose and fasting interval on cognitive function: a double-blind, placebo-controlled, six-way crossover study. *Psychopharmacology (Berl)*. 2012;220(3):577-589. doi:10.1007/s00213-011-2510-2.
32. Sünram-Lea S, Foster J, Durlach P, Perez C. Glucose facilitation of cognitive performance in healthy young adults: Examination of the influence of fast-duration, time of day and pre-consumption plasma glucose levels. *Psychopharmacology (Berl)*. 2001;157(1):46-54. doi:10.1007/s002130100771.
33. Scholey AB, Harper S, Kennedy DO. Cognitive demand and blood glucose. *Physiol Behav*. 2001;73(4):585-592. doi:10.1016/S0031-9384(01)00476-0.
34. Benton D, Brett V, Brain PF. Glucose improves attention and reaction to frustration in children. *Biol Psychol*. 1987;24(2):95-100.
35. Wang SR, Chase HP, Garg SK, Hoops SL, Harris MA. The effect of sugar cereal with and without a mixed meal on glycemic response in children with diabetes. *J Pediatr Gastroenterol Nutr*. 1991;13(2):155-160.
36. Buss RW, Kansal PC, Roddam RF, Pino J, Boshell BR. Mixed meal tolerance test and reactive hypoglycemia. *Horm Metab Res*. 1982;14(6):281-283. doi:10.1055/s-2007-1018995.

37. Ball SD, Keller, Kelly R.: Moyer-Mileur, Laurie J., Ding Y-W. Prolongation of Satiety After Low Versus Moderately High GlycemicIndex Meals in Obese Adolescents. *Pediatrics.* Updated 2003:488-494.
38. Philippou E, Constantinou M. The influence of glycemic index on cognitive functioning: a systematic review of the evidence. *Adv Nutr*. 2014;5(2):119-130. doi:10.3945/an.113.004960.
39. Smith A, Kendrick A, Maben A, Salmon J. Effects of Fat Content, Weight, and Acceptabilityof the Meal on Postlunch Changes in Mood,Performance, and Cardiovascular Function. *Physiol Behav*. 1994;55(3):417-422. Accessed June 30, 2016.
40. Wells AS, Read NW, Craig A. Influences of dietary and intraduodenal lipid on alertness, mood, and sustained concentration. *British Journal of Nutrition*. 1995;74:115-123. Accessed June 30, 2016.
41. Kaplan RJ, Greenwood CE, Winocur G, Wolever TM. Dietary protein, carbohydrate, and fat enhance memoryperformance in the healthy elderly. *Am J Clin Nutr*. 2001;74:687-693. Accessed June 30, 2016.
42. Gibson LE, Green MW. Nutritional influences on cognitive function: mechanisms of susceptibility. *Nutr Res Rev*. 2002;15(1):169-206. doi:10.1079/NRR200131.
43. Smith A, Leekam S, Ralph A, McNeill G. The Influence of Meal Composition on Post-lunch Changes in Performance Efficiency and Mood. *Appetite.* Updated 1988:195-203.
44. Spring B, Maller O, Wurtman J. Effects of Protein and Carbohydrate Meals on Mood and Performance: Interactions with Sex and Age. *Journal of psychiatric Research.* Updated 1982:155-167.
45. Pollitt E, Cueto S, Jacoby ER. Fasting and cognition in well- and undernourished schoolchildren: a review of three experimental studies. *Am. J. Clin. Nutr.* 1998;67(4):779.
46. Edefonti V, Rosato V, Parpinel M, et al. The effect of breakfast composition and energy contribution on cognitive and academic performance: a systematic review. *The American journal of clinical nutrition.* Updated 2014:626-656.
47. Hoyland A, Dye L, Lawton CL. A systematic review of the effect of breakfast on the cognitive performance of children and adolescents. *Nutr Res Rev*. 2009;22(2):220-243. doi:10.1017/S0954422409990175.
48. Cooper SB, Bandelow S, Nute ML, Morris JG, Nevill ME. Breakfast glycaemic index and cognitive function in adolescent school children. *Br. J. Nutr.* 2012;107(12):1823-1832. doi:10.1017/S0007114511005022.

49. Müller K, Libuda L, Gawehn N, et al. Effects of lunch on children's short-term cognitive functioning: a randomized crossover study. *Eur J Clin Nutr*. 2013;67(2):185-189. doi:10.1038/ejcn.2012.209.
50. Gibson EL. Carbohydrates and mentalfunction: feeding or impedingthe brain? *Nutrition Bulletin*. 2007;32(suppl 1):71-83. Accessed June 30, 2016.
51. Westenhöfer J. Kohlenhydrate und kognitive Leistungsfähigkeit. *Akt Ernähr Med*. 2006;31:96-102. doi:10.1055/s-2005-915364.
52. Donohoe RT, Benton D. Cognitive functioning is susceptible to the level of blood glucose. *Psychopharmacology (Berl)*. 1999;145(4):378-385.
53. Messier C. Glucose improvement of memory: a review. *Eur J Pharmacol*. 2004;490(1-3):33-57. doi:10.1016/j.ejphar.2004.02.043.
54. Gilsenan MB, Bruin EA de, Dye L. The influence of carbohydrate on cognitive performance: a critical evaluation from the perspective of glycaemic load. *Br. J. Nutr.* 2009;101(7):941-949. doi:10.1017/S0007114508199019.
55. Wurtman RJ, Hefti F, Melamed E. Precursor control of neurotransmitter synthesis. *Pharmacol Rev*. 1980;32(4):315-335.
56. Hirsch MJ, Growdon JH, Wurtman RJ. Relations between dietary choline or lecithin intake, serum choline levels, and various metabolic indices. *Metabolism*. 1978;27(8):953-960. doi:10.1016/0026-0495(78)90139-7.
57. Colzato LS, Jongkees BJ, Sellaro R, Hommel B. Working memory reloaded: tyrosine repletes updating in the N-back task. *Front Behav Neurosci*. 2013;7:200. doi:10.3389/fnbeh.2013.00200.
58. Greenwood CE, Craig RE. Dietary Influences on Brain Function: ImplicationsDuring Periods of Neuronal Maturation. *Current Topics in Nutrition and Disease.* Updated 1987:159-216. Accessed June 30, 2016.
59. Monk TH. The post-lunch dip in performance. *Clin Sports Med*. 2005;24(2):23. doi:10.1016/j.csm.2004.12.002.
60. Kanarek R. Psychological effects of snacks and altered meal frequency. *Br. J. Nutr.* 1997;77 Suppl 1:18.
61. Craig A. Acute effects of meals on perceptual and cognitive efficiency. *Nutr. Rev.* 1986;44 Suppl:163-171.
62. Smith AP, Kendrick AM. Meals and performance. *Handbook of human performance*. 1992;2:1-23.
63. Smith AP, Miles C. The effects of lunch on cognitive vigilance tasks. *Ergonomics*. 1986;29(10):1251-1261. doi:10.1080/00140138608967238.

64. Kirschbaum C, Wolf OT, May M, Wippich W, Hellhammer DH. Stress- and treatment-induced elevations of cortisol levels associated with impaired declarative memory in healthy adults. *Life Sci.* 1996;58(17):1475-1483.
65. Lupien SJ, Fiocco A, Wan N, et al. Stress hormones and human memory function across the lifespan. *Psychoneuroendocrinology*. 2005;30(3):225-242. doi:10.1016/j.psyneuen.2004.08.003.
66. Bohnen N, Houx P, Nicolson N, Jolles J. Cortisol reactivity and cognitive performance in a continuous mental task paradigm. *Biol Psychol*. 1990;31(2):107-116.
67. Lupien SJ, Gillin CJ, Hauger RL. Working memory is more sensitive than declarative memory to the acute effects of corticosteroids: a dose-response study in humans. *Behav. Neurosci.* 1999;113(3):420-430.
68. Lupien SJ, Maheu F, Tu M, Fiocco A, Schramek TE. The effects of stress and stress hormones on human cognition: Implications for the field of brain and cognition. *Brain Cogn*. 2007;65(3):209-237. doi:10.1016/j.bandc.2007.02.007.
69. Abercrombie HC, Kalin NH, Thurow ME, Rosenkranz MA, Davidson RJ. Cortisol variation in humans affects memory for emotionally laden and neutral information. *Behav. Neurosci.* 2003;117(3):505-516. doi:10.1037/0735-7044.117.3.505.
70. Lupien SJ, Lepage M. Stress, memory, and the hippocampus: can't live with it, can't live without it. *Behav. Brain Res.* 2001;127(1-2):137-158.
71. Stimson RH, Mohd-Shukri NA, Bolton JL, Andrew R, Reynolds RM, Walker BR. The Postprandial Rise in Plasma Cortisol in Men Is Mediated by Macronutrient-Specific Stimulation of Adrenal and Extra-Adrenal Cortisol Production. *The Journal of Clinical Endocrinology & Metabolism*. 2014;99(1):160-168. doi:10.1210/jc.2013-2307.
72. Hershberger AM, McCammon MR, Garry JP, Mahar MT, Hickner RC. Responses of lipolysis and salivary cortisol to food intake and physical activity in lean and obese children. *J. Clin. Endocrinol. Metab.* 2004;89(9):4701-4707. doi:10.1210/jc.2003-031144.
73. van Cauter E, Shapiro ET, Tillil H, Polonsky KS. Circadian modulation of glucose and insulin responses to meals: relationship to cortisol rhythm. *Am. J. Physiol.* 1992;262(4 Pt 1):75.
74. Gibson EL, Checkley S, Papadopoulos A, Poon L, Daley S, Wardle J. Increased salivary cortisol reliably induced by a protein-rich midday meal. *Psychosom Med*. 1999;61(2):214-224.
75. Knoll E, Müller FW, Ratge D, Bauersfeld W, Wisser H. Influence of food intake on concentrations of plasma catecholamines and cortisol. *J. Clin. Chem. Clin. Biochem.* 1984;22(9):597-602.

76. Follenius M, Brandenberger G, Hietter B. Diurnal cortisol peaks and their relationships to meals. *J. Clin. Endocrinol. Metab.* 1982;55(4):757-761. doi:10.1210/jcem-55-4-757.
77. Anderson P. Assessment and development of executive function (EF) during childhood. *Child Neuropsychol*. 2002;8(2):71-82. doi:10.1076/chin.8.2.71.8724.
78. Schmitt JAJ, Benton D, Kallus KW. General methodological considerations for the assessment of nutritional influences on human cognitive functions. *Eur J Nutr*. 2005;44(8):459-464. doi:10.1007/s00394-005-0585-4.
79. Wellek S, Blettner M. On the proper use of the crossover design in clinical trials: part 18 of a series on evaluation of scientific publications. *Dtsch Arztebl Int*. 2012;109(15):276-281. doi:10.3238/arztebl.2012.0276.
80. Grizzle JE. The Two-Period Change-Over Design and Its Use in Clinical Trials. *Biometrics*. 1965;21(2):467. doi:10.2307/2528104.
81. Monk TH, Buysse DJ, Reynolds CF, Kupfer DJ. Circadian determinants of the postlunch dip in performance. *Chronobiol. Int.* 1996;13(2):123-133.
82. Kirschbaum C, Hellhammer DH. Salivary cortisol in psychoneuroendocrine research: recent developments and applications. *Psychoneuroendocrinology*. 1994;19(4):313-333.
83. Tukey JW. *Exploratory Data Analysis.* Addison-Wesley Series in Behavioral Science. Massachusetts; 1977. Quantitative Methods, Reading; 1.
84. Leigh Gibson E, Green MW. Nutritional influences on cognitive function: mechanisms of susceptibility. *Nutr Res Rev*. 2002;15(1):169-206. doi:10.1079/NRR200131.
85. Abercrombie H.C. , Kalin N. H. , Thurow M. E. , Rosenkranz M. A., Davidson R. J. Cortisol Variation in Humans Affects Memory for Emotionally Laden and Neutral Information. *Behav. Neurosci.* 2003;(117):505-516.
86. Micha R, Rogers PJ, Nelson M. Glycaemic index and glycaemic load of breakfast predict cognitive function and mood in school children: a randomised controlled trial. *The British journal of nutrition.* Updated 2011:1552-1561.
87. Michels N, Sioen I, Vriendt T de, Huybrechts I, Vanaelst B, Henauw S de. Children's morning and evening salivary cortisol: pattern, instruction compliance and sampling confounders. *Horm Res Paediatr*. 2012;77(1):27-35. doi:10.1159/000334412.
88. Schröder M, Müller K, Falkenstein M, Stehle P, Kersting M, Libuda L. Short-term effects of lunch on children's executive cognitive functioning: The randomized crossover Cognition Intervention Study Dortmund PLUS (CogniDo PLUS). *Physiol Behav*. 2015;152(Pt A):307-314. doi:10.1016/j.physbeh.2015.09.025.

89. Adolphus K, Lawton CL, Dye L. The effects of breakfast on behavior and academic performance in children and adolescents. *Front Hum Neurosci*. 2013;7:425. doi:10.3389/fnhum.2013.00425.
90. Bellisle F. Effects of diet on behaviour and cognition in children. *BJN*. 2004;92(S2):S227. doi:10.1079/BJN20041171.
91. Schroder M, Muller K, Falkenstein M, Stehle P, Kersting M, Libuda L. Lunch at school and children's cognitive functioning in the early afternoon: results from the Cognition Intervention Study Dortmund Continued (CoCo). *Br. J. Nutr.* 2016;116(7):1298-1305. doi:10.1017/S0007114516002932.
92. Isaacs E, Oates J. Nutrition and cognition: assessing cognitive abilities in children and young people. *Eur J Nutr*. 2008;47 Suppl 3:4-24. doi:10.1007/s00394-008-3002-y.
93. Beaman CP. Auditory distraction from low-intensity noise: A review of the consequences for learning and workplace environments. *Appl. Cognit. Psychol.* 2005;19(8):1041-1064. doi:10.1002/acp.1134.
94. Elliott EM. The irrelevant-speech effect and children: theoretical implications of developmental change. *Mem Cognit*. 2002;30(3):478-487.
95. Benau EM, Orloff NC, Janke EA, Serpell L, Timko CA. A systematic review of the effects of experimental fasting on cognition. *Appetite*. 2014;77:52-61. doi:10.1016/j.appet.2014.02.014.
96. Vaisman N, Voet H, Akivis A, Vakil E. Effect of Breakfast Timing on the CognitiveFunctions of Elementary School Students. *Arch Pediatr Adolesc Med.* 1996;150:1089-1092. Accessed July 1, 2016.
97. Sekretariat der Ständigen Konferenz der Kultusminister der Länder in der Bundesrepublik Deutschland. *Allgemeinbildende Schulen in Ganztagsformin den Ländern in der Bundesrepublik Deutschland- Statistik 2010 bis 2014* -. Berlin; 2016.
98. World Health Organization (WHO) Europe. *Food and nutritionpolicy for schoolsFood and nutritionpolicy for schools: A tool for the development of school nutritionprogrammes in the European Region*. [Programme for Nutrition and Food Security WHO Regional Office for Europe]. Copenhagen; 2006.
99. Jager CA de, Dye L, Bruin EA de, et al. Criteria for validation and selection of cognitive tests for investigating the effects of foods and nutrients. *Nutr. Rev.* 2014;72(3):162-179. doi:10.1111/nure.12094.

7 Appendix

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Short-term effects of lunch on children's executive cognitive functioning: The randomized crossover Cognition Intervention Study Dortmund PLUS (CogniDo PLUS)☆

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HIGHLIGHTS

- Cross-over study to investigate lunch effect on children's executive functions
- Relation of postprandial cortisol increase and possible lunch effects was examined.
- No evidence for post-lunch dip in children
- Results indicate improving of updating function in children after lunch.
- Postprandial cortisol increase was not related with declines in executive functions, but even with reduced false alarms in the working memory updating.

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ABSTRACT

Objective: Studies indicate that eating lunch impairs some aspects of adults' cognitive functioning. However, the short-term effects of lunch on children's executive functions (EF) have not been examined. The Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) investigated (a) short-term effects of lunch on EF in children and (b) whether the postprandial cortisol increase mediates putative lunch effects on EF performance.
Methods: 5th and 6th grade students of a comprehensive school in Gelsenkirchen (Germany) participated in the randomized crossover intervention study. On Day 1 of the study, Group 1 did not eat lunch, whereas Group 2 received lunch ad libitum. One week later on Day 2 the groups were treated vice versa. At the beginning of the afternoon lessons, the EF parameters task switching, working memory updating and inhibition were tested using a computerized test battery. Saliva samples were used to measure cortisol directly before lunch and again at the beginning of the cognitive assessment. Of the 215 initially recruited children 21 dropped out of the study due to illness or absence on one of the two test days.
Results: Lower ratios of false alarms in the working memory updating function were observed when children who ate lunch than for children who had no lunch (8.2% (lunch) versus 9.4% (no lunch), $p < 0.01$). Parameters of task switching and inhibition did not differ between children who ate lunch compared to children who had no lunch. Stratification according to postprandial cortisol increase showed that the subgroup with a high increase had lower ratios of false alarms after eating lunch, while false alarm values did not change in the group with a low increase.
Conclusion: In contrast to findings in adults, the results indicate that children's EF are not impaired by lunch under true-to-life conditions. On the contrary, the current study even indicates beneficial effects of lunch intake for the working memory updating. The postprandial cortisol increase in the range observed in our sample does not seem to be related with negative effects on the performance of EF, but even seem to mediate the beneficial effect of lunch on the working memory updating.

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1. Introduction

Considering the increasing numbers of all-day schools in Europe research of potential determinants of children's cognitive performance in the afternoon is a relevant public health issue. Authors of a recent review [1] concluded that lunch can impair some aspects of cognitive functioning in adults. However, appropriate studies of children were not identified. The results of studies of adults are not necessarily transferable to children due to the constitutional and metabolic differences of children and adults (e.g., rapid brain growth, high metabolic rate).

The randomized crossover Cognition Intervention Study Dortmund (CogniDo) [2] recently provided the first insights on the effect of lunch on the cognitive performance of children and found no detrimental short-term effects of lunch on children's basal cognitive functioning (alertness, selective attention, visual-spatial memory). For one parameter (omission errors in tonic alertness tests) the CogniDo study suggests beneficial short-term lunch effects. However, lunch effects on higher cognitive functions, such as executive functions (EF), still need to be examined. EF is an umbrella term for a set of higher-order cognitive processes which govern low-level cognitive functions and are localized in the frontal lobes [3]. EF are most important for daily performance, and during school lessons EF are necessary for retaining and transforming short-term memory contents as well as for controlling actions and planning action sequences [4].

It is known that EF are not fully developed in children and adolescents [5] and decay earlier than low-level functions with increasing age [6]. Aging research shows that lifestyle factors such as physical exercise influence EF [7]. The maturation of the cerebral cortex (including the frontal lobes), which takes place until late adolescence, is characterized by dynamic changes of metabolism. For example, children's brains are more dependent on the intake of glucose in comparison to adult brains [8]. Therefore, it has been hypothesized that the developing EF might be more sensitive to environmental influences (including nutrient intake) than the EF in a fully developed adult brain [8,9]. This study refers to three often postulated executive functions, which are responsible for the following abilities: to inhibit prepotent responses ("inhibition"), to monitor and update information ("working memory updating"), and mental task switching [4,3].

In regard to the reasons for negative effects from lunch intake on adult cognitive performance the so-called post-lunch dip is discussed. It is suggested that a naturally occurring dip in performance around midday seems to be exacerbated by lunch intake [10–12]. Little is known about the exact mechanisms how lunch intake could worsen the post-lunch dip. However, different metabolic explanations have been suggested. One explanation might be that the post-lunch increases in blood glucose lead to a hypoglycemic state and a decrease in arousal. Other discussed mechanisms assume that an increase in blood glucose after lunch leads to the parasympathetic initiation of an insulin surge or that lunch induce changes in the level of serotonin [13].The occurrence of this lunch-induced performance dip for EF in children remains to be examined.

Another possible mechanism that would explain how lunch could impair cognitive functions is a meal-induced increase of plasma cortisol. Adrenal cortisol secretion underlies a circadian rhythm with peak plasma concentrations upon waking up and a nadir around midnight [14]. For both adults and children lunch intake induces an increase in cortisol levels [15–19]. It has been observed that an increased cortisol level induced by psychological stress or pharmaceuticals impair memory performance in adults [20–22]. Particularly because the frontal brain lobes contain glucocorticoid receptors, it is reasonable that cortisol has an influence on cognitive functions [23]. However, the threshold of a physiological postprandial increase in cortisol, which could effectively impair the cognitive functions, is not known for children. It remains to be evaluated whether a postprandial increase in cortisol might mediate lunch-induced impairments of cognitive performance in children.

In line with CogniDo, the objectives of the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) were to investigate (a) short-term effects of lunch intake on EF in children in the early afternoon and (b) whether a postprandial cortisol increase mediates potential lunch-related changes in cognitive performance.

2. Methods

2.1. Study design and participants

Similar to CogniDo [2], this randomized, open-label 2 × 2 crossover trial also took place in an all-day comprehensive school in Gelsenkirchen, Germany. The research period spanned 6 weeks from beginning of November until the middle of December 2013. Each subject participated on two study days, which took place on the same weekday with one week in between.

The participants were recruited from the 5th and 6th grade levels. Children with a diagnosed learning disorder (reported by the class teacher), metabolic disease (where long fasting phases are critical), or on special diets (which excludes the lunch meal) were excluded. Out of the total of 323 students in the 5th and 6th grades 215 students (67%) with informed written parental consent were eligible to participate in the study. Each participant was randomly assigned to one of two study groups: on Day 1 of the study, Group 1 did not eat lunch (no lunch, NL), whereas Group 2 received lunch ad libitum (lunch, L) before the assessment of cognitive performance. One week later on Day 2 of the study, Group 1 was in the L condition and Group 2 in the NL condition. Of the 215 children with written consent, 21 dropped out of the study due to illness or absence on one of the two individual test days, which resulted in complete cognitive performance data for 194 children. The children, who participated on both study days, each received a patience game as reward for their participation.

The study was approved by the Ethics Committee of the University of Bonn and registered at clinicaltrials.gov (NCT02082444). All assessments were made in accordance to the Declaration of Helsinki.

2.2. Study schedule

The study design was integrated in the regular school-day routine and corresponded to the study schedule of the previous crossover study CogniDo [2], but differed in the integration of additional cortisol measurements (Fig. 1).

Both test days started at 9:15 a.m. during the regular morning breakfast break with a standardized breakfast for each subject (wholemeal bread with margarine, poultry salami or Gouda cheese and carrot sticks) ad libitum. During the regular lunch break at 12:25 p.m. after the morning lessons, subjects either received pasta Bolognese ad libitum and an apple (L) prepared as usual by the kitchen staff in the school canteen, or the subjects had no lunch in a separate room (NL). The amount of consumed pasta was measured by individually weighing each plate before and after the meal. Water was available for the subjects at any time in both test situations. After finishing lunch all students (L and NL) had a common break until 13:10 p.m. (app. 15–20 min). At the beginning of the regular afternoon lessons (at 13:15 p.m.) the assessment of cognitive performance took place in the school's computer room. After completing these tests participants who had no lunch at the regular time received their lunch (14:00 p.m.).

Between the morning break and the beginning of cognitive performance tests (from 9:35 a.m. to 14 p.m.), all participants were asked to refrain from eating (except the lunch at the L condition) and drinking (except for water and unsweetened tea). In order to assess whether the students followed this requirement (for a protocol analysis) the participants filled out a questionnaire regarding their food and beverage consumption at the end of each intervention day. Additionally, the study staff supervised the children in the schoolyard and classrooms during the breaks. Saliva samples were collected to examine the

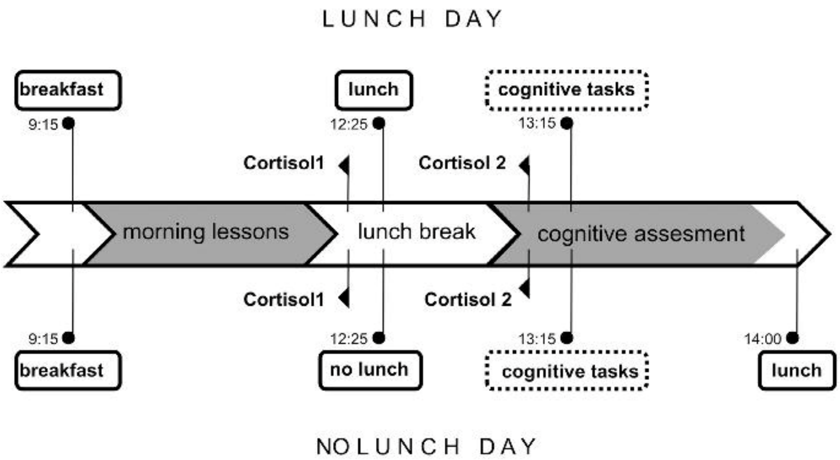

Fig. 1. Schedule of the intervention day for Lunch and No lunch group in the CogniDo PLUS study.

saliva-cortisol status two times on each test day: before lunch (at approx. 12:20 p.m.) and before the computerized cognitive assessment (at approx. 13:10 p.m.).

2.3. Cognitive assessment

For the assessment of executive functions (EF) a computerized test battery consisting of 3 tasks designed by the Institute of Working, Aging and Learning (ALA Institute) in Bochum, Germany was used. At the beginning of the cognitive assessment each task was explained to the group, and the participants were able to practice each task once in a training mode. After the training phase and a 5-min break with low (physical) exercise the actual testing began. Subjects were requested to respond as quickly as possible without sacrificing accuracy. With regard to reaction times (working memory updating and inhibition) plausible data were defined as values within the range of quartile 1 minus 1.5 times the Inter Quartile Range (IQR) and quartile 3 plus 1.5 times the IQR [24]. For the "ratios of errors" a predefined limit of <50% error rate was defined as plausible since the expected error rate would be 50% if a subject is choosing his/her reaction completely by random. So, exclusion of error ratios >50% was intended to reduce the risk of a systematic error in observations from single subjects as these values indicate a high risk that the task instructions were misunderstood (e.g. if a subject always pushed a button when no reaction was attended).

The tasks were applied in the following order:

2.3.1. Task switching (switch)

With an alternative version of the Trail Making Task (TMT) we measured the subjects' ability to switch between two tasks. This task was presented in 3 sections - the first two sections (section one: numbers, section two: letters) in a non-switch condition and the third section in a switch condition (letters and numbers mixed) (Fig. 2):

2.3.1.1. First section, numbers, non-switch. Black numbers from 1 to 26 in white squares were presented in an irregular order on a black computer screen. The children were asked to click the numbers in ascending order with the mouse curser. The starting point was a square with the number 1, which was marked green. The squares turn green after a correct answer and red after a false answer as a form of feedback, and the correctly processed squares fade out. The maximum time limit to finish the task was 3 min.

2.3.1.2. Second section, letters, non-switch. This section had the same format as the numbers section, but used letters from A to Z instead of numbers.

2.3.1.3. Third section, switch. The 26 squares contained numbers from 1 to 13 and letters from A to M. The children were asked to alternately click numbers and letters in ascending order (i.e. 1-A-2-B-3-C…).

The outcomes consisted of the total reaction time for numbers (for items 1–25), total reaction time for letters (for items 1–13) and switch-costs, i.e. the processing time of the third (switch) section minus the first section (numbers; nonswitch) minus the difference between the first 13 items of the first and second (letter; nonswitch) section. To eliminate any implausible data we excluded all subjects with negative switch costs.

2.3.2. Working memory updating (2-back task)

In order to assess the function of constant monitoring and adding or deleting of working memory contents, we used the n-back task in a 2-back condition. Participants were asked to monitor a sequence of 106 consecutive trials (pictures of fruits and vegetables) presented in the middle of a white screen. When the current picture matched the picture presented 2 trials before (n-2), the participant was instructed to press a pre-defined key on the computer keyboard with the index finger (Fig. 3). The stimuli were presented for 500 ms with an interstimulus interval (ISI) of 2100 ms and a maximal reaction time of 1400 ms. No feedback was given. Of the 106 pictures shown, 21 were targets (same picture as 2 trials before).

The outcomes were ratios of missings (no reaction while reaction was required), ratios of false alarms (reaction while no reaction was required) and mean reaction times. Plausible measurements were defined as mean of ratio of incorrect answers and false alarms <50% and reaction times >172.8 ms and <864.7 ms.

2.3.3. Inhibition (flanker task)

The flanker task was used to assess the ability to inhibit a prepotent response. In each trial three superposed triangles were presented to the participants. The upper and lower triangles (flankers) were pointing in the same direction but independent from the middle triangle (target). Each trial was categorized as compatible-go, incompatible-go or no-go (Fig. 4). Compatible-go trials were those during which flankers and the target were pointing in the same direction, incompatible-go trials were those when flankers and target were pointing in opposite direction. During the no-go trials a circle replaced the target. The participants were supposed to press the buttons left or right according to the direction of the target or not to react in the case of a no-go condition. To induce flanker-target conflict, the flankers were presented individually for 100 ms and remained together with the target for another 300 ms. The maximum reaction time was 1100 ms, the response stimulus interval (RSI) was 1200 ms (varying ±20%). In go-trials the feedback "faster" was shown when there was no reaction during the maximum reaction

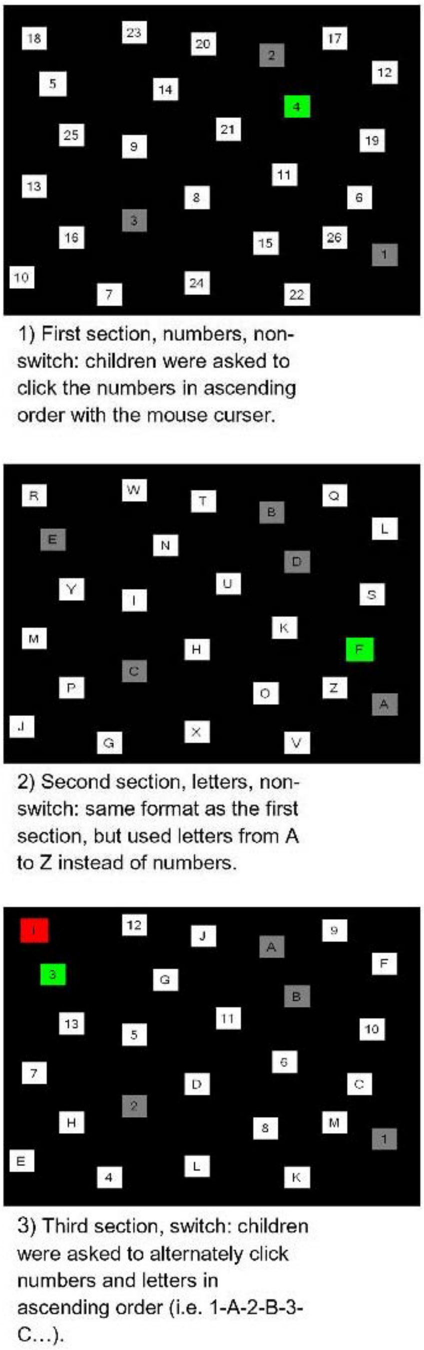

Fig. 2. Cognitive assessment: Switch task was presented in 3 sections — the first two sections (section one: numbers, section two: letters) in a non-switch condition and the third section in a switch condition (letters and numbers mixed). Screenshots of the sections.

time or when the reaction time was >600 ms. The feedback stimulus started 500 ms after the response (or after 1100 ms in no-reaction trials) and lasted 300 ms. In total, this task consisted of 102 items (32 no-go, 35 incompatible-go, 35 compatible-go).

The outcomes were the difference between the ratio of incorrect reactions in compatible-go and incompatible-go trials (difference error rate), the difference between mean reaction times of compatible-go and incompatible-go trials (rt slowing) and the mean count of false alarms (participants pressed the button instead of showing no reaction). Reactions for which a negative reaction time was detected (i.e. the subject gave the reaction before the trial showed up) were excluded.

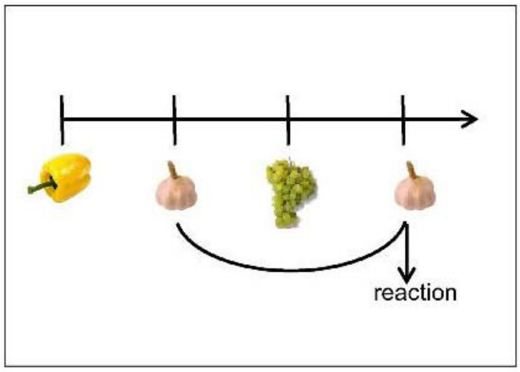

Fig. 3. Cognitive assessment: 2-back task to assess working memory updating. Scheme of the task.

Plausible data were defined as follows: Ratio of incorrect reactions in compatible-go trials <50%,

Ratio of incorrect reactions in incompatible-go trials <50%,
Count of false alarms <16 (less than 50%),
Reaction times of compatible-go trials >138.9 ms and <438.6 ms,
Reaction times of incompatible-go trials >201.2 ms and <514.0 ms.

2.4. Cortisol measurements

Saliva samples were taken at three times on each intervention day: directly before lunch (at approx. 12:20 p.m., T1) and at the beginning of the cognitive assessment (i.e. approximately 45 min after starting lunch (at approx. 13:10 p.m.), T2; see Fig. 1). For this analysis samples T1 and T2 were taken into account in order to consider postprandial cortisol changes before the cognitive tests as a potential mediator of putative lunch effects on cognitive performance. Under supervision participants collected their own saliva using Salivette collection devices (Sarstedt, Nuembrecht, Germany). The samples were stored in a cool box for the transport and frozen (−18 °C) until they were analyzed. For the analyses the Salivettes were thawed and centrifuged at 3000 rpm for 2 min in order to obtain a clear fluid. Free saliva cortisol levels were measured using the RE62011 immunoassay (IBL, Hamburg, Germany). One third of the samples were analyzed two-fold and the remainder one-fold.

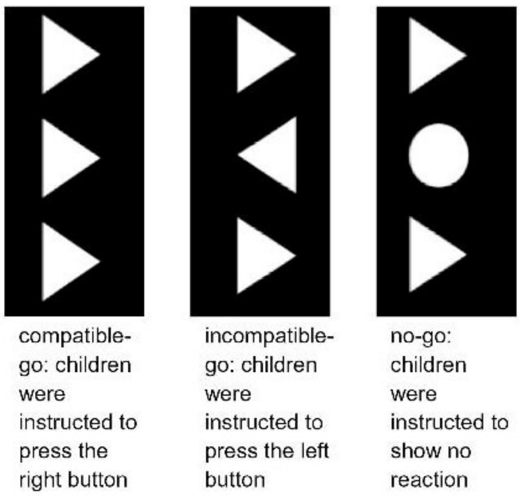

Fig. 4. Cognitive assessment: inhibition. Screenshots of the trials.

2.5. Statistical analyses

All analyses were performed using the statistical software package SAS 9.2 (SAS Institute, Cary, NC, USA). $p < 0.05$ was considered as statistically significant.

The parameters of the three cognitive tasks and the cortisol status at the beginning of the cognitive assessment (T2) as well as the difference of the cortisol status before the lunch break and before cognitive assessment (T2–T1) were used as outcome variables. All outcome variables were interval-scaled. As recommended by Grizzle [25] the sums of the respective outcome variable were compared between groups using the parametric unpaired t-test for normally distributed data (including normal distribution after transformation) and the non-parametric Wilcoxon rank-sum test for non-normally distributed data to examine potential carry-over effects. Appropriate non-normally distributed outcomes were transformed (log, square, root and reciprocal transformation) and analyzed using unpaired t-test. If transformation did not result in normally distributed parameters the non-parametric Wilcoxon rank-sum test was used to analyze carry-over or intervention effects. As no carry-over effects were observed, results from both days were considered for the calculation of the treatment effect. Therefore, individual differences of the particular outcomes of both test days (test day 1–test day 2) were compared between groups (NL–L vs. L–NL) the same way the sums of the outcomes were analyzed. Descriptive data (sex, lunch size and eating a refectory lunch) were analyzed by Chi-squared test or Fisher's Exact Test.

Before the statistical analyses the raw data were checked for plausibility as described above. For cortisol, all available samples of T1 and T2 were analyzed.

To examine the potential mediating effect of the postprandial cortisol increase on potential lunch effects on the individual cognitive performance two approaches were used: In step 1, a linear regression was used to analyze associations between postprandial cortisol increase and the EF outcomes which were affected by eating lunch. If effects of having lunch on EF rely on the increase in cortisol, it implies that lunch effects should be observed in particular in subjects with a high postprandial cortisol increase. This assumption was tested in a second step performing an additional stratified analysis of lunch effects in subjects with low vs subjects with high postprandial cortisol increases (using a median-split for postprandial cortisol increase).

In addition linear regressions between the lunch size and EF outcomes and between lunch size and cortisol increase were conducted (model 1 unadjusted and model 2 adjusted for sex and age).

3. Results

3.1. Participants

Data from cognitive assessment were not available for 2 subjects for the task switching and flanker task. For the 2-back data from 1 subject were missing.

Implausible data in cognitive performance were excluded separately for each of the 3 tasks. For the switch task 91.7% of the participants were included, for the 2-back task 57.5% and for the flanker task 65.6%. Characteristics of the excluded and included subjects for each task are shown in Table 1. In the 2-back task the excluded subjects were slightly older (11.7 years) than the included (11.4 years). The ratio of boys was higher among excluded (76.8%) than included (45%) subjects. Participants' mean consumption of the pasta bolognese was 368 g (±154 g) (range: 55–920 g).

3.2. General lunch effects on cognitive functions

The ratio of false alarms in working memory updating (2-back task) was significantly lower in the L than in the NL condition ($p = 0.01$, Table 2). For inhibition (flanker task) the students tended to make fewer errors in the L condition than in the NL condition, but this difference was not statistically significant (difference of error rate $p = 0.16$) and disappeared with additional exclusion of subjects who did not follow the study protocol ($n = 26$, per protocol analysis) whereas the intervention effect of the ratio of false alarms in working memory updating remained in the per protocol analysis (data not shown). No lunch effects were observed for the other parameters of cognitive performance.

The linear regression of lunch size and EF generally showed no association between lunch size and EF except for a (non-significant) trend between switch costs and lunch size ($ß = -18.51$ and $p = 0.083$ in not adjusted model and $ß = -18.95$ and $p = 0.078$ in the adjusted model), i.e. the children with higher lunch sizes tended to have lower switch costs. The linear regression of lunch size and postprandial cortisol increase revealed postprandial cortisol increase were positively associated with lunch size (not adjusted model: $ß = 0.0025$ and $p = 0.0081$ adjusted model: $ß = 0.0024$ and $p = 0.0113$).

3.3. Postprandial cortisol and cognitive functions

The cortisol samples were collected before lunch (T1 = 12:20 h) and again about 45 min after starting lunch (before cognitive assessment at 13:10 = T2). Cortisol levels at T2 were significantly higher on the L day and also the cortisol levels between T1 and T2 increased significantly in comparison to the NL day ($p < 0.0001$, Table 3). On the NL day the cortisol levels decreased from T1 to T2 ($p < 0.0001$) while levels increased on lunch ($p < 0.001$).

The linear regression revealed a significant negative linear association between postprandial cortisol increases and the ratio of false alarms in the 2-back task, i.e. decreasing ratios of false alarms for increasing levels of postprandial cortisol increase ($ß = -0.83$ and $p = 0.04$) (Fig. 5). The linear regression of cortisol on the remaining EF outcomes revealed a significant association only with the visual search for numbers within the task switching ($ß = 893.5$ and $p = 0.04$). However the lunch intervention had no influence on this outcome.

Stratification according to postprandial cortisol increases (median cut, high vs. low) generally confirmed the results of the pooled analysis, as no differences were observed for parameters of the switch and

Table 1
Characteristics of included and excluded schoolchildren participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) (Switch and Flanker: $n = 192$; 2-back: $n = 193$).

	Switch		2-back		Flanker	
	Included (176)	Excluded (16)	Included (111)	Excluded (82)	Included (126)	Excluded (66)
Age, mean ± S.D. years	11.5 (0.76)	11.8 (0.74)	11.4 (0.73)	11.7 (0.79)	11.6 (0.76)	11.5 (0.78)
Female, n (%)	76 (43.2)	5 (31.3)	61 (55)	19 (23.2)	56 (44.4)	24 (36.4)
Regular lunch[a], n (%)	158 (89.8)	14 (87.5)	100 (90.1)	73 (89)	110 (87.3)	62 (93.9)
Pasta consumption (median P25/P75) [g]	345 (265/418)	373 (315/573)	335 (235/405)	375 (315/430)	343 (275/415)	360 (245/430)

[a] Defined as consuming lunch at the school cafeteria regularly by subscription.

Table 2
Effects of no lunch vs. lunch on examined parameters of executive functioning in schoolchildren (10–12 years) participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS).

Task	Main outcome	No lunch			Lunch			p-Value
		Median	25th	75th	Median	25th	75th	
Switch (n = 176)	Switch costs[a] [ms]	27,633	15,017	39,117	24,844	15,256	37,326	0.34[b]
	Visual search letters[c] [ms]	26,462	22,893	32,361	27,121	23,424	32,002	0.47[d]
	Visual search numbers [ms]	45,639	40,439	51,548	46,392	38,836	52,987	0.91[d]
2-back (n = 111)	Ratio of missings [%]	28.6	23.8	35	28.6	19	38.1	0.82[d]
	Ratio of false alarms [%]	9.4	4.7	21.2	8.2	3.5	17.6	0.01[d]
	rt[e] [ms]	513.3	443.5	598.5	529.2	432.5	596	0.87[d]
Flanker (n = 126)	rt. slowing [ms]	67.3	52.9	89.5	73.1	44.7	90.5	0.18[d]
	Difference error rate [%]	0.14	0.06	0.22	0.11	0.06	0.23	0.16[b]
	Mean count of false alarms [N]	1.0	0.0	4.0	1.0	0.0	3.0	0.24[d]

[a] Switch costs = (mean rt. switch task) − (mean rt. number task) − (mean rt. first 12 reactions of letter task).
[b] Two sample t-test.
[c] First 12 reactions.
[d] Wilcoxon rank-sum test.
[e] Reaction time.

flanker task (data not shown). In the 2-back task a significant lower ratio of false alarms after lunch was observed only in students with a high postprandial cortisol increase ($p = 0.03$, Table 4).

4. Discussion

The present study revealed that there was no evidence for a lunch-related dip in cognitive performance of children. In contrast, levels of false alarms in the working memory updating task were lower when lunch was eaten compared to the test condition in which lunch was omitted. After meals children responded less frequently to non-targets, to which they should have not responded. Because no effect of meals was observed in the inhibition task, it could be hypothesized that the decrease of false alarms is not the consequence of an improvement of inhibition, i.e. of less premature responses. Rather the results may suggest that meals improved working memory updating, i.e. they helped children to distinguish non-targets more clearly from targets. The measured postprandial cortisol increase did not seem to cause declines in performance of EF. On the contrary, the results suggest that those subjects with a high postprandial cortisol increase could experience a beneficial effect on the working memory updating function, i.e. they correctly disregarded non-targets. This analysis confirms that effects of having lunch are indeed only observed if lunch results in high increases of cortisol levels. Therefore, effects of having lunch on children's updating of the working memory seem to be -at least partly- mediated by increasing levels of cortisol after lunch.

Until now, the impact of no lunch or lunch on cognitive functioning has primarily been investigated in adults [26,13,12,27]. The outcomes of these studies are not fully consistent, but point to impairments of some parameters of cognitive functioning after eating lunch in comparison to the functioning in a pre-lunch condition. Smith and Miles [26] observed no post-lunch changes in selective attention in 48 students but did observe impairments in the ability to maintain attention and reaction times on new stimuli in contrast to pre-lunch testing. In another study it was shown that lunch impaired the ability to discriminate [12]. In contrast, Karnarek and Swinney [27] observed an improvement in reading ability after lunch compared to the no lunch condition, whereas no effects were seen in working memory or sustained attention. A review [28] concluded that lunch seems to attenuate sustained attention more than briefer tasks of selective attention.

To the best of the knowledge of the authors, the previous CogniDo [2] study was the first investigation on the cognitive effects of lunch in children. In line with findings from the current study the CogniDo study neither revealed negative nor general beneficial effects of lunch on parameters of general cognitive performance, such as the visuospatial subsystem within the working memory, or on selective attention. While the CogniDo study suggested an improvement of tonic alertness (as reflected in a lower rate of omissions), the present study revealed an effect on short-term memory and it's updating by using executive functioning tests. Although cognition tasks of the 2 studies partially measured different parameters, a summary of results from both studies suggests that — in contrast to adults — lunch does not seem to impair short-term cognitive functioning in children, but might even improve single cognitive parameters.

A discussed phenomenon of the implied impairment in cognition in adults after lunch is the so called post-lunch dip, i.e. a naturally occurring dip at midday, which is supposedly related to more than one factor, e.g. individual circadian rhythm or length of time since sleep [29,11,30]. It was observed that lunch exacerbates the naturally occurring dip in adults [12,13]. The mechanism behind this phenomenon is not yet understood, but e.g. alterations in the synthesis of neurotransmitters by availability of amino acids or postprandial hypoglycemia have been discussed as potential factors [31].

In adults the post-lunch dip is assumed to be maximally 60–120 min after starting lunch [12,30,13]. In both studies (the present and the previous) cognitive tests were carried out about 60 min after starting and 40 min after the subjects finished lunch. No lunch-induced dip was seen in children. However, it cannot be ruled out that a post-lunch impairment might occur in children later in the afternoon.

Another possible mechanism explaining the effect of lunch on EF may be related to meal-induced postprandial increases of plasma cortisol [1]. Adults exhibited impaired memory performance after cortisol increases induced by psychological stress or pharmaceuticals [20–22]. Lupien et al. [32] ascertained that working memory is more prone to pharmaceutically induced cortisol increase than declarative memory. From their results they inferred that a low pharmaceutical cortisol dose improves the processing capacity of working memory, whereas a high dose leads to impaired working memory performance. A quadratic

Table 3
Saliva cortisol levels of schoolchildren participating in the Cognition Intervention Study Dortmund PLUS before lunch and changes over time ($n = 187$)[a].

	No lunch			Lunch			
Cortisol	Median	25th	75th	Median	25th	75th	p-Value[b]
T2 [ng/ml][c]	1.21	0.76	1.78	1.88	1.13	2.99	<0.0001[d]
Change between T1 and T2[e]	−0.38[f]	−1.06	0.05	0.27[f]	−0.46	1.09	<0.0001[g]

[a] 5 missings.
[b] p-Value of lunch-effect.
[c] T2 = cortisol level at 13:05 (ng/ml).
[d] Wilcoxon rank-sum test.
[e] Change between T1 and T2 = difference of cortisol level T2 at 13:05–T1 at 12:15 (ng/ml).
[f] Wilcoxon signed rank test.
[g] Two sample t-test.

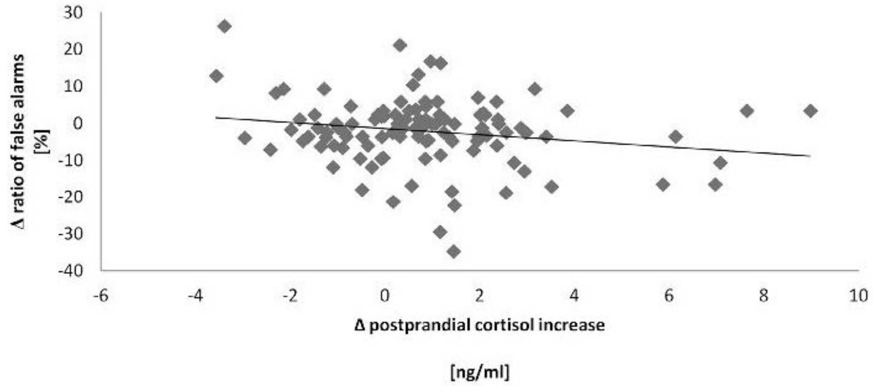

Fig. 5. Linear regression between postprandial cortisol increase (considering cortisol changes in fasting condition, i.e. cortisol change lunch minus cortisol change no lunch) and the difference of the ratio of false alarms of the 2-back task ($p = 0.04$) ($n = 108$).

function (inverse U-shape curve) is suggested between performance on the working memory task and changes in glucocorticoids levels after hydrocortisone infusion [33,21]. Abercromie et al. [34] found out that a dose of 20 or 40 mg cortisol given previous testing caused fewer errors of commission i.e. false alarms in 18–33 year old adults. This finding was reproduced in the present study since the false alarms in the flanker task were reduced by a physiological raise of cortisol after lunch.

In this study lunch induced a cortisol increase in children, with larger lunch sizes causing increasing cortisol responses (not adjusted model: $ß = 0.0025$ and $p = 0.0081$ adjusted model: $ß = 0.0024$ and $p = 0.0113$). After dividing the subjects into two groups based on postprandial cortisol increase (high and low), the improved working memory updating results after lunch remained only in the high postprandial increase group. We assume that physiologically induced cortisol increase after lunch could improve the working memory updating function in children reflecting the beneficial effect of lunch on working memory updating. No conclusions can be drawn, however, about the amount of the increase or the exact level at which improvements or impairments occur. Micha et al. [35] considered the cortisol response in a study on the impact of GI/GL on cognitive performance in children. It was observed that a high GI breakfast increases the blood glucose and cortisol levels which results in a better performance on a virgilance task. They hypothesized that higher blood glucose levels could activate the hypothalamic–pituitary–adrenal axis in stressful situations (i.e. test situation) which lead to higher cortisol levels and higher tension of the participants before the tests, and thus better performance on tasks were information processing is tested. However, more studies in children are needed to investigate the effect and metabolism pathway of postprandial cortisol levels on cognitive performance in children.

Several characteristics of the study design need to be discussed. CogniDo PLUS was not conducted under clinical conditions, but tested the students under real-life conditions in their school environment. In this setting students are exposed to different influences (e.g. their peers), which might be an explanation for the high count of implausible outcomes. Due to the limiting values in the inhibition and working memory updating task, about 35–42% of the students were excluded per task because of an error rate over 50% or an implausible reaction time (see definition of plausible data in 'methods'). We suppose that the high error rate resulted from accuracy speed trade-off because the excluded participants partially showed relatively short reaction times. It could be assumed that these students suppressed accuracy for speed in order to "win" a speed contest with their seat neighbors. However, only a classroom setting mirrors the real-life conditions in school and allows conclusions about everyday school life compared to purified clinical conditions. The schedule of the intervention was embedded in the "normal" school day with EF tasks performed at the usual start of the afternoon lessons. Therefore, the transferability of our results is given. However, the effect on cognitive performance later in the afternoon remains to be evaluated.

For practical reasons (e.g. no extra medical staff, no cortisol increase because of the stress of blood sampling) postprandial cortisol was not assessed with blood sampling but instead from saliva samples. However, saliva cortisol levels are a valid reflection of the respective unbound hormone in blood [36]. Unfortunately, an evaluation of the analyzed cortisol data is not possible because of a lack of consensus reference values for children's saliva measurement [37]. Nevertheless, absolute values of cortisol were of minor importance in the current study because only the individual changes of cortisol levels of the subjects were considered.

Using the Grizzle model for the analyses of intervention effects, it was not possible to perform an interaction test between cortisol and working memory updating as part of a pathway analysis. However, we tested the modifying effect of postprandial cortisol indirectly via stratified analysis of lunch effects and also through linear regression between postprandial cortisol increase and change in working memory updating.

Table 4
Effects of no lunch vs. lunch on the main outcomes of the 2-back task after stratification for high or low postprandial saliva cortisol increase in schoolchildren participating in the Cognition Intervention Study Dortmund PLUS (CogniDo PLUS) ($n = 108$).

			No lunch			Lunch			p-Value
Test	Variable	Cortisol increase	Median	25th	75th	Median	25th	75th	
2-back	Ratio of missings [%]	High (n = 50)	28.57	28.81	33.33	28.57	19.04	40.00	0.678[a]
		Low (n = 58)	28.57	19.05	38.1	28.57	19.05	38.1	0.642[a]
	Ratio of false alarms [%]	High (n = 50)	9.41	5.88	22.35	8.24	3.53	16.47	0.033[b]
		Low (n = 58)	11.18	4.7	19.28	8.24	3.53	17.86	0.232[a]
	rt[c] [ms]	High (n = 50)	518.64	460.37	586.25	530.63	418.06	595.07	0.832[a]
		Low (n = 58)	487.73	441.80	604.18	517.94	432.50	602.91	0.820[a]

[a] Wilcoxon rank-sum test.
[b] Two sample t-test with (square) transformed data.
[c] Reaction time.

Although absolute lunch size was not associated with most EF parameters, more information would have been needed e.g. about regular individual lunch size in order to draw any conclusions between meal size and cognitive performance. It was not possible to get this information due to organizational reasons, but this question would be interesting for future studies. The present study (as well as CogniDo) stands out due to the crossover design, which eliminates variations between the subjects and reduces bias.

In conclusion and in contrast to findings in adults, the results indicate that lunch does not impair children's EF in real-life conditions. For the parameter reflecting working memory updating the current study even indicates beneficial effects of lunch intake. Taken together, CogniDo and CogniDo Plus reveal that eating lunch during the school day has no negative effects on cognition in children. For some tasks school lunch could even have partially positive influence on cognitive performance of some children in the afternoon, but these observations need to be further investigated in future studies.

Conflicts of interest

The authors declare that there are no conflicts of interest.

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References

[1] K. Müller, L. Libuda, A.M. Terschlüsen, M. Kersting, A review of the effects of lunch: on adults' short-term cognitive functioning, Can. J. Diet. Pract. Res. 74 (4) (2013) 181–188, http://dx.doi.org/10.3148/74.4.2013.181.

[2] K. Müller, L. Libuda, N. Gawehn, et al., Effects of lunch on children's short-term cognitive functioning: a randomized crossover study, Eur. J. Clin. Nutr. 67 (2) (2013) 185–189, http://dx.doi.org/10.1038/ejcn.2012.209.

[3] A. Miyake, N.P. Friedman, M.J. Emerson, A.H. Witzki, A. Howerter, T.D. Wager, The unity and diversity of executive functions and their contributions to complex "frontal lobe" tasks: a latent variable analysis, Cogn. Psychol. 41 (1) (2000) 49–100, http://dx.doi.org/10.1006/cogp.1999.0734.

[4] C. Tillman, K.C. Brocki, L. Sørensen, A.J. Lundervold, A longitudinal examination of the developmental executive function hierarchy in children with externalizing behavior problems, J. Atten. Disord. (2013)http://dx.doi.org/10.1177/1087054713488439.

[5] S.L. Thompson-Schill, M. Ramscar, E.G. Chrysikou, Cognition without control: when a little frontal lobe goes a long way, Curr. Dir. Psychol. Sci. 18 (5) (2009) 259–263, http://dx.doi.org/10.1111/j.1467-8721.2009.01648.x.

[6] G.R. Turner, R.N. Spreng, Executive functions and neurocognitive aging: dissociable patterns of brain activity, Neurobiol. Aging 33 (4) (2012) e1–13, http://dx.doi.org/10.1016/j.neurobiolaging.2011.06.005 (826).

[7] H. Guiney, L. Machado, Benefits of regular aerobic exercise for executive functioning in healthy populations, Psychon. Bull. Rev. 20 (1) (2013) 73–86, http://dx.doi.org/10.3758/s13423-012-0345-4.

[8] H.T. Chugani, A critical period of brain development: studies of cerebral glucose utilization with PET, Prev. Med. 27 (2) (1998) 184–188, http://dx.doi.org/10.1006/pmed.1998.0274.

[9] E. Pollit, Does breakfast make a difference in school? J. Am. Diet. Assoc. 95 (10) (1995) 1134–1139, http://dx.doi.org/10.1016/S0002-8223(95)00306-1.

[10] T.H. Monk, D.J. Buysse, C.F. Reynolds, D.J. Kupfer, Circadian determinants of the postlunch dip in performance, Chronobiol. Int. 13 (2) (1996) 123–133.

[11] T.H. Monk, The post-lunch dip in performance, Clin. Sports Med. 24 (2) (2005) e15–e23, http://dx.doi.org/10.1016/j.csm.2004.12.002 (xi-xii).

[12] A. Craig, K. Baer, A. Diekmann, The effects of lunch on sensory-perceptual functioning in man, Int. Arch. Occup. Environ. Health 49 (1981) 105–114, http://dx.doi.org/10.1016/j.csm.2004.12.002.

[13] A.P. Smith, C. Miles, The effects of lunch on cognitive vigilance tasks, Ergonomics 29 (10) (1986) 1251–1261, http://dx.doi.org/10.1080/00140138608967238.

[14] R.H. Stimson, N.A. Mohd-Shukri, J.L. Bolton, R. Andrew, R.M. Reynolds, B.R. Walker, The postprandial rise in plasma cortisol in men is mediated by macronutrient-specific stimulation of adrenal and extra-adrenal cortisol production, J. Clin. Endocrinol. Metab. 99 (1) (2014) 160–168, http://dx.doi.org/10.1210/jc.2013-2307.

[15] A.M. Hershberger, M.R. McCammon, J.P. Garry, M.T. Mahar, R.C. Hickner, Responses of lipolysis and salivary cortisol to food intake and physical activity in lean and obese children, J. Clin. Endocrinol. Metab. 89 (9) (2004) 4701–4707, http://dx.doi.org/10.1210/jc.2003-031144.

[16] E. van Cauter, E.T. Shapiro, H. Tillil, K.S. Polonsky, Circadian modulation of glucose and insulin responses to meals: relationship to cortisol rhythm, Am. J. Physiol. 262 (4 Pt 1) (1992) E467–E475.

[17] E.L. Gibson, S. Checkley, A. Papadopoulos, L. Poon, S. Daley, J. Wardle, Increased salivary cortisol reliably induced by a protein-rich midday meal, Psychosom. Med. 61 (2) (1999) 214–224.

[18] E. Knoll, F.W. Müller, D. Ratge, W. Bauersfeld, H. Wisser, Influence of food intake on concentrations of plasma catecholamines and cortisol, Eur. J. Clin. Chem. Clin. Biochem. (Z. Klin. Chem. Klin. Biochem), 22 (9) (1984) 597–602.

[19] M. Follenius, G. Brandenberger, B. Hietter, Diurnal cortisol peaks and their relationships to meals, J. Clin. Endocrinol. Metab. 55 (4) (1982) 757–761, http://dx.doi.org/10.1210/jcem-55-4-757.

[20] C. Kirschbaum, O.T. Wolf, M. May, W. Wippich, D.H. Hellhammer, Stress- and treatment-induced elevations of cortisol levels associated with impaired declarative memory in healthy adults, Life Sci. 58 (17) (1996) 1475–1483.

[21] S.J. Lupien, A. Fiocco, N. Wan, et al., Stress hormones and human memory function across the lifespan, Psychoneuroendocrinology 30 (3) (2005) 225–242, http://dx.doi.org/10.1016/j.psyneuen.2004.08.003.

[22] N. Bohnen, P. Houx, N. Nicolson, J. Jolles, Cortisol reactivity and cognitive performance in a continuous mental task paradigm, Biol. Psychol. 31 (2) (1990) 107–116.

[23] S.J. Lupien, M. Lepage, Stress, memory, and the hippocampus: can't live with it, can't live without it, Behav. Brain Res. 127 (1–2) (2001) 137–158.

[24] J.W. Tukey, Exploratory Data Analysis, Addison-Wesely, 1977.

[25] J.E. Grizzle, The two-period change-over design and its use in clinical trials, Biometrics 21 (2) (1965) 467, http://dx.doi.org/10.2307/2528104.

[26] A.P. Smith, C. Miles, Effects of lunch on selective and sustained attention, Neuropsychobiology 16 (2–3) (1986) 117–120.

[27] R.B. Kanarek, D. Swinney, Effects of food snacks on cognitive performance in male college students, Appetite 14 (1) (1990) 15–27.

[28] E.L. Gibson, M.W. Green, Nutritional influences on cognitive function: mechanisms of susceptibility, Nutr. Res. Rev. 15 (1) (2002) 169–206, http://dx.doi.org/10.1079/NRR200131.

[29] R. Kanarek, Psychological effects of snacks and altered meal frequency, Br. J. Nutr. 77 (Suppl 1) (1997) S105–S118 discussion 118-20.

[30] A. Craig, Acute effects of meals on perceptual and cognitive efficiency, Nutr. Rev. 44 (Suppl) (1986) 163–171.

[31] A.P. Smith, A.M. Kendrick, Meals and performance, Handbook of human performance, 2 1992, pp. 1–23.

[32] S.J. Lupien, C.J. Gillin, R.L. Hauger, Working memory is more sensitive than declarative memory to the acute effects of corticosteroids: a dose–response study in humans, Behav. Neurosci. 113 (3) (1999) 420–430.

[33] S.J. Lupien, F. Maheu, M. Tu, A. Fiocco, T.E. Schramek, The effects of stress and stress hormones on human cognition: implications for the field of brain and cognition, Brain Cogn. 65 (3) (2007) 209–237, http://dx.doi.org/10.1016/j.bandc.2007.02.007.

[34] H.C. Abercrombie, N.H. Kalin, M.E. Thurow, M.A. Rosenkranz, R.J. Davidson, Cortisol variation in humans affects memory for emotionally laden and neutral information, Behav. Neurosci. 117 (2003) 505–516.

[35] R. Micha, P.J. Rogers, M. Nelson, Glycaemic index and glycaemic load of breakfast predict cognitive function and mood in school children: a randomised controlled trial, Br. J. Nutr. 106 (10) (2011) 1552–1561, http://dx.doi.org/10.1017/S0007114511002303.

[36] C. Kirschbaum, D.H. Hellhammer, Salivary cortisol in psychoneuroendocrine research: recent developments and applications, Psychoneuroendocrinology 19 (4) (1994) 313–333.

[37] N. Michels, I. Sioen, T. de Vriendt, I. Huybrechts, B. Vanaelst, S. de Henauw, Children's morning and evening salivary cortisol: pattern, instruction compliance and sampling confounders, Horm. Res. Paediatr. 77 (1) (2012) 27–35, http://dx.doi.org/10.1159/000334412.

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Lunch at school and children's cognitive functioning in the early afternoon: results from the Cognition Intervention Study Dortmund Continued (CoCo)

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Abstract

Studies about effects of school lunch on children's cognition are rare; two previous studies (CogniDo, CogniDo PLUS) generally found no negative effects of lunch on children's cognitive performance at the end of lunch break (i.e. 45 min after finishing lunch), but suggested potential beneficial effects for single parameters. Therefore, the present study investigated the hypothesis of potential positive effects of school lunch on cognitive performance at early afternoon (90 min after finishing lunch). A randomised, cross-over intervention trial was conducted at a comprehensive school with fifth and sixth grade students. Participants were randomised into two groups: On day 1, group 1 did not eat lunch, whereas group 2 received lunch *ad libitum*. On day 2 (1 week later), group 2 did not eat lunch and group 1 received lunch *ad libitum*. The cognitive parameters task switching, working memory updating and alertness were tested using a computerised test battery 90 min after finishing the meal. Of the 204 recruited children, fifty were excluded because of deviations from the study protocol or absence on one of the 2 test days, which resulted in 154 participants. Data showed no significant effects of lunch on task switching, working memory updating and alertness (*P* values between 0·07 and 0·79). The present study suggests that school lunch does not seem to have beneficial effects on children's cognitive functions regarding the conducted tests at early afternoon. Together with our previous studies, we conclude that school lunch in general has no negative effects on cognitive performance in children. However, beneficial effects seem to be restricted to a relatively short time period after eating lunch.

Key words: Children: Cognitive functions: Lunch: Cognition

Considering the extension of all-day schools in Europe, knowledge about potential effects of school lunch on children's cognition is becoming increasingly important. Short-term lunch effects on cognition were primarily examined in adults with equivocal results, until now. Although sustained attention and the ability to discriminate have been shown to be worsened after lunch(1,2), other cognitive aspects were improved (reading ability)(3) or did not change (selective attention)(4). One explanation for negative lunch effects on cognitive performance is the post-lunch dip – a naturally occurring nadir in performance at midday. Studies in adults have shown that this dip is worsened by lunch(5,1). Therefore, it could be hypothesised that skipping lunch could result in an alleviation or prevention of this post-lunch dip. However, these results were obtained in adults and are not necessarily transferable to children due to constitutional and metabolic differences between children and adults (e.g. still rapid brain growth, high metabolic rate in children). Until now, studies in children about the effects of meals on cognition have mainly concentrated on breakfast(6). A body of research work has shown short-term benefits for cognitive performance when children eat breakfast instead of skipping it(6). However, other studies showed that poorly nourished children benefit more than well-nourished children(7). Although no definitive conclusions can be drawn about short-term benefits of breakfast for cognitive performance in children in general, there is at least a noticeable indication for a beneficial influence(8,6,9).

Studies about school lunch and short-term effects on children's cognition are rare(10). To the best of our knowledge, there are only two cross-over, intervention trials from our group, which

Abbreviations: CF, cognitive functions; CoCo, Cognition Intervention Study Dortmund Continued; L, lunch; NL, no lunch.

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provided first insights into the impact of school lunch on cognitive performance in children at the end of lunch break (CogniDo[11], CogniDo PLUS[12] study). Both studies did not reveal negative effects of lunch on several parameters of cognitive functions (CF) (i.e. task switching, working memory updating, inhibition, alertness, selective attention, block span) when determined about 45 min after finishing lunch. For single parameters such as omission errors in the alertness task and false alarms in the updating task (regarding the working memory) results even point to beneficial effects of lunch at the end of lunch break. Even though no lunch-related cognitive impairment was observed in these two studies, it has to be considered that a post-lunch dip in children could appear as the afternoon progresses. In adults, a post-lunch dip was observed about 60–120 min after lunch[1,4,13], indicating that the time span between a meal and the measurement of cognitive performance could have a relevant influence[13].

Therefore, the objective of the Cognition Intervention Study Dortmund Continued (CoCo) was to investigate the potential positive effects of school lunch on cognitive performance in children at early afternoon (90 min after finishing lunch instead of 45 min in the previous intervention studies). In order to provide comparability with both previous studies, cognitive tests that proved to be the most sensitive were chosen – that is, the alertness task from the first study (CogniDo)[11] and task switching and working memory updating task from the second study (CogniDo PLUS)[12]. On the basis of the previous results of these two studies, we hypothesise that children will perform better on lunch day than on no lunch day.

Methods

Study design and participants

Similar to the previous studies, the CoCo study was conducted as a randomised, open-label, 2×2 cross-over intervention trial. The same all-day comprehensive school in Gelsenkirchen, Germany, was chosen for the experiment. In total, the field period spanned 19 weeks between October 2014 and March 2015 including 3 weeks of holidays. Each subject had to participate on two study days with 1 week in between on the same weekday.

The participants were recruited from the fifth and sixth grades (twelve classes). The students of the sixth grade in the present study had already participated in the previous CogniDo PLUS study (as fifth grade students). Children with diseases with potential consequences of fasting and children on special diets, who were not allowed to eat the study meal, were excluded from participation. Children with a diagnosed learning disorder reported by the class teacher were allowed to participate, but were excluded *post hoc* for the analyses. Out of 324 students, 204 provided informed written consent to participate. A cluster randomisation per class with a block size of four participants was conducted to assign participants to one of two study groups: on day 1 of the study, group 1 did not eat lunch (no lunch, NL), whereas group 2 received lunch *ad libitum* (lunch, L); 1 week later, on day 2 of the study, group 1 was in the L condition and group 2 in the NL condition. All children, who participated on both intervention days, received a ball as reward for their participation.

The study was approved by the Ethics Committee of the University of Bonn and registered at clinicaltrials.gov (NCT02344056). All assessments were made in compliance with the Declaration of Helsinki.

Study schedule

The study design was integrated in the regular school routine and corresponded to the study schedules of the previous studies (CogniDo[11], CogniDo PLUS[12]), but differed in the time interval between lunch break and assessment of cognitive performance (Fig. 1).

On both test days, a standardised breakfast (wholemeal bread with margarine, poultry salami or Gouda cheese and carrot sticks) *ad libitum* was offered to the students in the regular morning break at 09.15 hours. During the regular lunch break, starting at 12.25 hours, the subjects either received lunch *ad libitum* (pasta with or without Bolognese sauce as desired) and an optional apple in the school canteen prepared as usual by the kitchen staff (L), or the subjects skipped lunch and stayed in a separate room (NL). Water was available at any time in both test situations. The amount of pasta was individually weighed before and after the meal ± 5 g. After lunch, all students (L and NL) had their common break (until 13.20 hours) and the regular seventh lesson (13.25–14.10 hours). At the beginning of the eighth lesson (14.15 hours), the assessment of

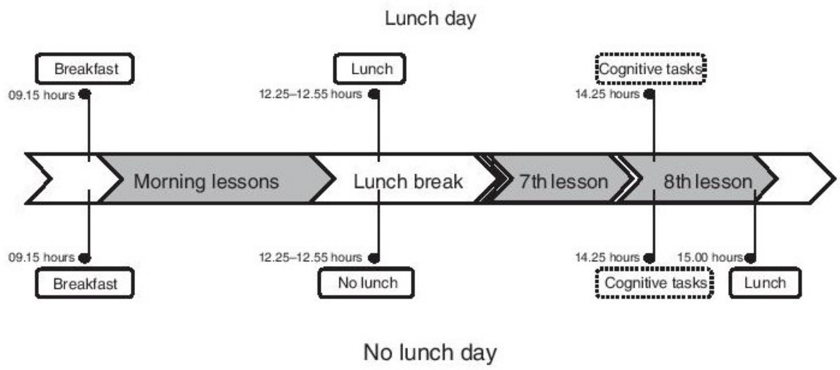

Fig. 1. Schedule of the intervention day and timing for lunch group and no lunch group in the Cognition Intervention Study Dortmund Continued (CoCo) study.

cognitive performance took place in the school's computer room at about 14.25 hours. After completing these tests (about 15.00 hours), participants in the NL condition received their lunch.

Between the morning break and the lunch break (09.35–12.25 hours), all participants were asked to refrain from eating and drinking (except for water and unsweetened tea). The NL group was additionally asked to refrain from eating and drinking until the end of the test day (15.00 hours). In order to assess compliance with the study protocol, the study staff supervised the children in the schoolyard and classrooms during the breaks. In addition, the participants filled out a questionnaire regarding their food and beverage consumption at the end of each intervention day.

Cognitive assessment

For the assessment of CF, a computerised test battery consisting of three tasks (ALA Institute) was used. Before starting, students had to pass a training phase with a task-by-task explanation by the study personnel and a short practise period. After this training and a 5-min break with low physical exercise, the actual cognitive testing began. Subjects were requested to respond as quickly as possible without sacrificing accuracy. The cognition tasks were applied in the following order: task switching, working memory updating and tonic alertness.

After finishing the cognitive testing for two-thirds of the study sample, the school laptops had to be replaced by desktop computers for school intern reasons. The remaining participants were tested on the new computers. Consequently, the participants were either tested on the laptops or on the desktops. Therefore, calibration of the new computer was not necessary as the analyses of the intervention effects considered the individual differences between two tests days.

Task switching. With an alternative version of the trail making task, subjects' ability to switch between two tasks was measured. This task was presented in three sections: the first two sections (section 1: numbers, section 2: letters) in a non-switch condition, and the third section in switch condition (letters and numbers mixed).

First section – numbers: black numbers from 1 to 26 in white squares were presented in an irregular order on a black computer screen (Fig. 2). The children were asked to click the numbers in an ascending order with the mouse curser. The square with the number 1 was marked green as the starting point. The squares turn green after a correct answer and red after a false answer as a form of feedback. Correctly processed squares fade out. The maximum time limit to finish the task was 3 min.

Second section – letters: this section had the same format as the numbers section, but used letters from A to Z instead of numbers (Fig. 3).

Third section – switch: the twenty-six squares contained numbers from 1 to 13 and letters from A to M (Fig. 4). The children were asked to alternately click numbers and letters in ascending order (i.e. 1-A-2-B-3-C…).

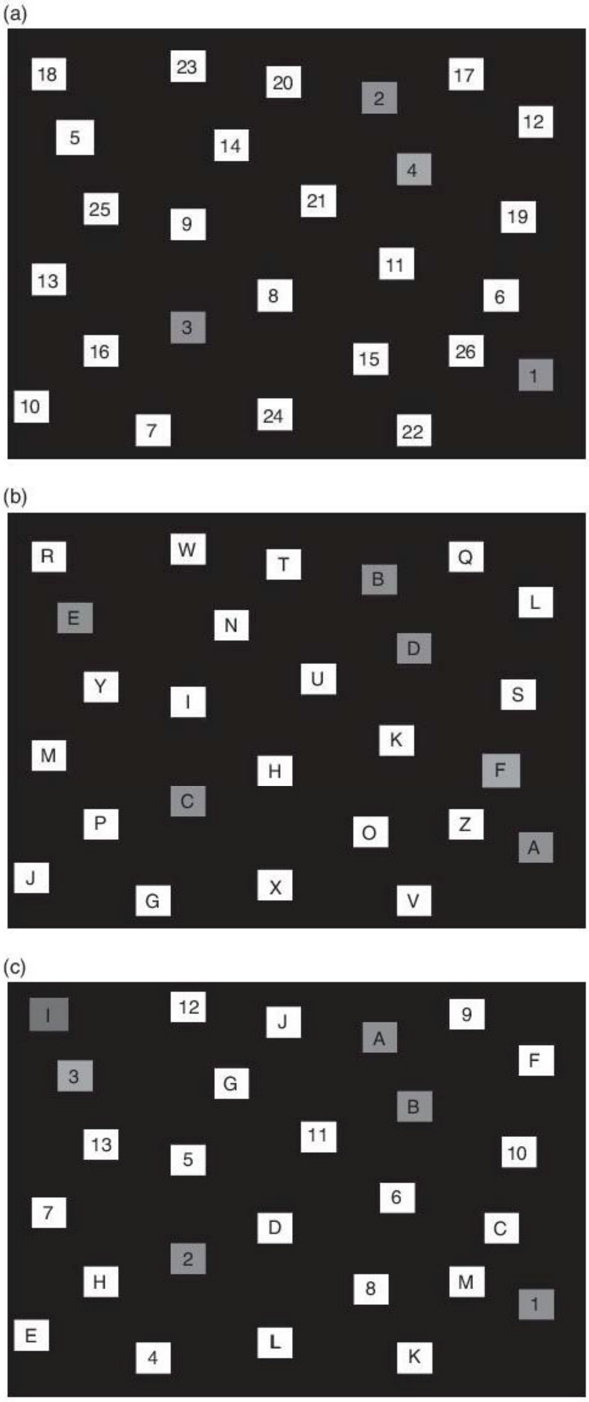

Fig. 2. Cognitive assessment: switch task was presented in three sections – the first two sections (section 1: numbers, section 2: letters) in a non-switch condition and the third section in a switch condition (letters and numbers mixed). Screenshots of the sections. (a) First section, numbers, non-switch: children were asked to click the numbers in ascending order with the mouse curser. (b) Second section, letters, non-switch: same format as the first section, but used letters from A to Z instead of numbers. (c) Third section, switch: children were asked to alternately click numbers and letters in ascending order (i.e. 1-A-2-B-3-C…).

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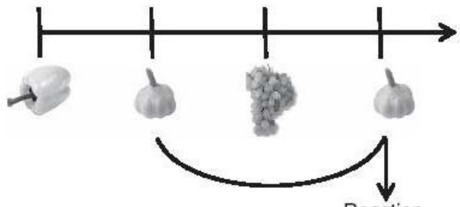

Fig. 3. Cognitive assessment: two-back task to asses working memory updating. Scheme of the task.

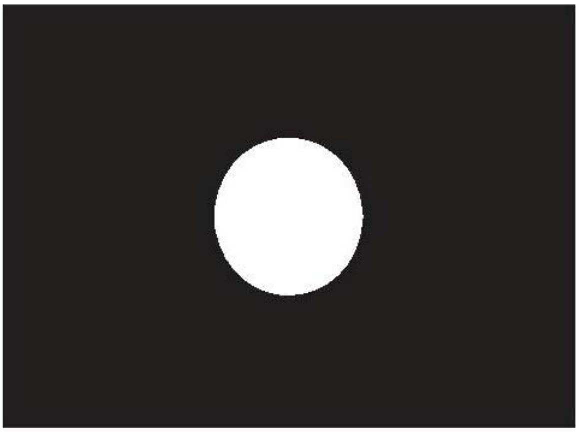

Fig. 4. Cognitive assessment: alertness. Children were instructed to press a predefined key as soon as the white circle appears on the screen.

The outcomes were total reaction time for numbers (for items 2–26), total reaction time for letters (for items 2–13) and switch-costs – that is, the processing time of the third section (switch; items 2–26) minus the first section (numbers; non-switch; items 2–26) minus the difference between the first twelve items of the second section (letter; non-switch; items 2–13) and the first twelve items of the first section (numbers; non-switch; items 2–13). To eliminate implausible data, we excluded all subjects with negative switch costs.

Working memory updating (two-back task). In order to assess the function of constant monitoring and adding or deleting of working memory contents, we used the *n*-back task in a two-back condition. Participants were asked to monitor a sequence of 106 consecutive trials (pictures of fruits and vegetables) presented in a white square in the middle of a black screen. When the current picture matched the picture presented two trials before (n–2), the participant was instructed to press a predefined key on the computer keyboard with the index finger. The stimuli were presented for 500 ms with an inter-stimulus interval of 2100 ms and a maximal reaction time of 1400 ms. No feedback was given. Of the 106 pictures shown, twenty-one were targets (same picture as two trials before).

The outcome variables were the ratio of missings (no reaction while reaction was required), the ratio of false alarms (reaction while no reaction was required) and the mean reaction time while reaction was required. Plausible measurements were defined as ratios of missing ≤50 %, false alarms ≤50 % and reaction times, between quartile 1 minus 1·5 times the inter-quartile range (IQR) and quartile 3 plus 1·5 times the IQR (i.e. ≥196·935 and ≤850·975 ms).

Tonic alertness. To measure the level of tonic alertness, we used a simple reaction task. A white fixation cross was presented in the middle of a black screen. In a response stimulus interval of 3300 ms (±20 %), a circle followed the cross and the subjects were supposed to press a predefined button as soon as the circle appears (maximal reaction time 1500 ms). The test included fifty items.

The outcome variables were the mean reaction time (ms), the deviation of reaction time (ms), the number of omission errors (no reaction after appearance of the circle within 1500 ms) and the number of commission errors (reactions during the presence of the fixation cross). Plausible measurements were defined as reaction times ≥140·9 and ≤492·58 ms.

Statistical analyses

All analyses were performed using the statistical software package SAS 9.2 (SAS Institute). $P < 0{\cdot}05$ was considered as statistically significant.

Before conducting the statistical analyses, the raw data of all cognitive tasks were checked for plausibility. With regard to reaction times (working memory updating and alertness), plausible data were defined as values within the range of quartile 1 minus 1·5 times the IQR and quartile 3 plus 1·5 times the IQR[14]. For the 'ratios of false alarms/missings', a predefined limit of <50 % error rate was defined as plausible as the expected error rate would be 50 % if a subject is choosing his/her reaction completely in random. Therefore, exclusion of error ratios ≥50 % was intended to reduce the risk of a systematic error in observations from single subjects as these values indicate a high risk that the task instructions were misunderstood (e.g. if a subject always pushed a button when no reaction was attended). Only plausible data were included in the analysis.

The parameters of the three cognitive tasks were used as outcome variables. All outcome variables were interval scaled. As recommended by Grizzle[15], the sums of the two individual values of the particular outcomes variables of test days 1 and 2 were compared between groups using an unpaired *t* test for normally distributed data (including normal distribution after transformation) and the Wilcoxon's ranked sum test for non-normally distributed data to examine potential carry-over effects. Non-normally distributed outcomes were transformed (log, square, root or reciprocal transformation) and analysed using unpaired *t* test. If transformation did not result in normally distributed parameters, the non-parametric Wilcoxon's ranked sum test was used to analyse carry-over or intervention effects. As no carry-over effects were observed, results from both days were considered for the calculation of the treatment effect.

In addition, linear regression analyses between the lunch size and the change in cognitive performance parameters

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Table 1. Linear regression of lunch weight on the change of main outcome variables of cognitive performance in schoolchildren (10–12 years) participating in the Cognition Intervention Study Dortmund Continued
(adjusted for sex and age; only valid models presented)

			Lunch weight	
Tasks	Outcome	Intercept	β-value	*P*
Switch	Switch costs	−6754·0	10·966	0·226
	Visual search of letters*	3635·7	−9·344	0·029
Two-back	Ratio of missings	23·3	−0·002	0·835
	Reaction time	4·3	0·004	0·947

* First twelve reactions.

Table 2. Characteristics of the schoolchildren participating in the Cognition Intervention Study Dortmund Continued*
(Numbers and percentages; means and standard deviations; medians and 25th/75th percentiles)

	L-NL (*n* 72)		NL-L (*n* 82)		
	n	%	*n*	%	*P*
Age (years)					0·31†
Mean	11·3		11·4		
SD	0·7		0·6		
Female	36	50	31	37·8	0·13‡
Regular lunch§‖	63	88·7	78	95·1	0·23‡
Meal consumption (g)					0·87‡
Median	360		375		
25th/75th percentiles	275·0/525·0		275·0/505·0		

L, lunch day; NL, no lunch day.
* Group NL-L skipped lunch during the first period, group L-NL skipped lunch during the second period.
† Two sample *t* test.
‡ χ^2 Test/Fisher's exact test.
§ Defined as consuming lunch at the school refectory regularly by subscription.
‖ Missing data from one subject.

(performance on lunch day and performance on no lunch day) were conducted for all parameters, including age and sex as additional covariables. For all parameters of the alertness task, for the parameter visual search of numbers in the task-switching task and the parameter ratio of false alarms (two-back task), the linear regression revealed non-normally distributed residuals (the association with lunch size was not significant for all of these parameters). As the linear regression might not be meaningful for these parameters, we decided not to present these results in (Table 1).

Results

Participants

Of the 204 included participants, nineteen participants (one class) had to be excluded because of a 30-min delay of lunch on their 2nd test day. In addition, thirty-one children were absent on one of the 2 test days, resulting in 154 participants with complete data. Characteristics of the sample stratified by study groups L-NL (*n* 72), that is, having lunch on the 1st test day and skipping it on the 2nd test day, and NL-L (*n* 82) are shown in Table 2. There were no differences in age, sex or consumed amounts of the study meal between both groups. The majority of subjects regularly ate lunch at the school refectory (88·7 % in L-NL and 95·1 % in NL-L) with no significant difference between the groups ($P = 0·23$).

Lunch effects on cognitive functions

Statistical analyses revealed no significant effects of lunch on the examined CF of task switching, working memory updating (two-back task) and alertness (*P* between 0·12 and 0·79, Table 3). The time for visual search of letters on the task-switching task showed a trend to slightly increase after having lunch ($P = 0·07$). After additional exclusion of subjects who did not follow the study protocol (*n* 67, per protocol analysis), this trend disappeared (data not shown).

The linear regression analysis revealed a significant negative association between the lunch size and the change in visual search of letters ($\beta = -9·3$, $P = 0·03$ adjusted) in the task-switching task.

Discussion

The present study revealed no evidence for a lunch-related improvement or decline of cognitive performance in schoolchildren in the early afternoon, about 90 min after finishing lunch. Although our previous studies CogniDo[11] and CogniDo PLUS[12] suggested slight improvements of single cognitive parameters shortly after lunch, the current study did not prove our hypothesis of potentially beneficial cognitive effects of lunch in the afternoon. Interestingly, the linear regression even indicated beneficial effects of lunch size as the individual change between lunch day and no lunch day decreased with

Table 3. Effects of no lunch *v.* lunch on cognitive performance in schoolchildren (10–12 years) participating in the Cognition Intervention Study Dortmund Continued
(Medians and 25th and 75th percentiles)

		No lunch			Lunch			
Tasks	Main outcome	Median	25th	75th	Median	25th	75th	*P*
Switch (*n* 139)	Switch costs (ms)*	23 928	14 990	38 298	21 475	13 367	31 864	0·26†
	Visual search letters (ms)‡	26 255	22 579	31 270	26 958	22 927	33 928	0·07†
	Visual search numbers (ms)	43 967	38 007	50 548	43 854	38 220	50 782	0·36†
Two-back (*n* 87)	Ratio of missings (%)	28·6	19·0	35·0	28·6	19·0	38·1	0·25†
	Ratio of false alarms (%)	7·1	2·4	17·6	7·1	3·5	14·1	0·63†
	RT (ms)	519·0	445·8	584·8	518·5	457·3	615·1	0·36†
Alertness (*n* 148)	Mean RT (ms)	306·0	273·1	343·9	314·9	270·9	357·0	0·12†
	Deviation of RT (ms)	117·7	92·5	173·2	128·6	91·4	179·7	0·53†
	Count of omission errors (*n*)	0	0	1	0	0	1	0·75†
	Count of commission errors (*n*)	3	1	5	2	1	5	0·79†

RT, reaction time.
* Switch costs = (mean RT switch task) – (mean RT number task) – (mean RT first twelve reactions of letter task–mean RT first twelve reactions of number task).
† Wilcoxon's ranked sum test.
‡ First twelve reactions.

larger lunch sizes for the parameter visual search letters. However, more information was needed for interpreting this result – for example, deviation from regular individual lunch size. It was not possible to obtain this information because of organisational reasons, but this question would be interesting for future studies.

In the earlier CogniDo study[11] (*n* 105), the participants made significantly more omission errors in the tonic alertness task on the no lunch day compared with the lunch day ($P=0{\cdot}03$). The CogniDo PLUS study[12] (*n* 195) suggested slightly lower levels of false alarms in the task regarding working memory updating when lunch was eaten compared with the test condition in which lunch was omitted ($P=0{\cdot}01$) – that is, after eating lunch, children responded less frequently to non-targets, to which they should not have responded. Both tasks (tonic alertness and working memory updating) of the previous studies were also conducted in the present study without any hints at beneficial effects. A potential explanation for these divergent study results could be the difference in the time span between lunch and cognitive testing, which was about 45 min after finishing lunch in the former studies, but twice as long with 90 min in the current study. A comparison between the variable values of the present study and the previous CogniDo and CogniDo PLUS studies shows that the values are located in the same data range. Even though the data were not conducted from the same probands, they seem to be comparable. Considering these results, it might be hypothesised that children's cognitive performance may slightly increase immediately after lunch and may not improve when the fasting period is extended into the early afternoon.

Although the reasons for differences in lunch effects depending on the interval until cognitive testing could not be examined in our studies, one plausible explanation may be the course of blood glucose levels. Glucose levels increase in the early postprandial period, but might have been on a decrease at the time when cognitive performance was tested in the present CoCo study. It could be speculated that an increase in blood glucose is beneficial for cognitive performance, whereas a decrease in glucose might attenuate this effect despite higher levels than in the fasting condition. Studies that investigated glucose uptake showed that the resulting increase in blood glucose levels enhances CF such as memory[16,17] or sustained attention[18]. For example, Owen *et al.*[17] demonstrated that a glucose dose of 25 g enhanced working memory performance following a 2-h fast, and Benton *et al.*[18] showed that 25 g glucose as a drink improved sustained attention. However, effects of oral glucose dosage may differ depending on blood glucose resources or the level of depletion of glucose reservoirs, for example, in the liver[17]. Furthermore, tests with pure glucose consumption may not simply mirror the effects of a whole meal as applied in our study. Sugars or other carbohydrates as part of a mixed meal increase blood glucose levels more slowly than pure glucose[19,20]. Therefore, future studies should assess the role of lunches differing in their glycaemic response on cognitive performance in children and adolescents.

Another influencing factor on cognition could be the post-lunch dip phenomenon that may relate to the timing of the meal and the interval until measurements. In adults, a decline in cognitive capabilities was observed in a wide range of about 60–120 min after lunch[1,4,13]. In our previous studies, cognitive parameters were measured 45 min after lunch, without any evidence of a post-lunch dip. It remains an open question whether a meal-enhanced post-lunch dip in children exists as has been suggested for adults. As a post-lunch dip was mostly seen in sustained attention tasks[21,22], it might especially be detectable in the alertness task (which includes testing for sustained attention). As our results did not show a significant difference between the L and the NL condition, it could be assumed that there might not be a post-lunch dip in children. However, to answer this question conclusively, further studies will be needed.

There are several characteristics of the CoCo study design that need to be discussed. The study was not conducted under clinical conditions, but tested the students in real-life conditions in their classroom setting. Accordingly, factors apart from lunch such as environmental stress (e.g. noise, peer group actions) might have influenced individual cognitive performance[23]. Especially auditory distraction can have detrimental effects[24,25] on the cognitive performance in children. Even though the study team tried to keep the children in the testing room as quiet as possible, environmental disturbances could partly have masked acute individual effects of lunch. Clinical studies might be more suitable to clearly identify isolated lunch effects, but do not allow any conclusions on the practical meaning of these results in children's everyday life. In addition, the cross-over design of the CoCo study should have minimised potential effects of individual confounding in the total sample. To counteract a possible learning effect, we used a parallel version of the cognitive task in which the task switching and the two-back sequences differed (task switching task differed in position of the items, two-back task in sequence of fruit and vegetable items). In addition, we conducted a training phase immediately before the actual testing to ensure that every subject understands the tests already before the first assessment. The task-switching task and the two-back task, which were already used in our previous study, were originally designed for adults. Although we adapted these tests for children and tested the entire test battery in a pretest with children of the same age in another school in order to avoid a very sophisticated test, the rate of implausible two-back test results might indicate a floor effect in the same subjects.

In the CoCo study, no pre-lunch performance was measured as opposed to laboratory studies. However, we decided against this practice as we were worried about potential negative effects on the motivation with increasing numbers of tests. If we had included a pre-lunch measurement, the children would have had to complete four tests on 2 test days within a week, and without a pre-lunch testing only two. Negative influences on motivation might not only have impact on the results of the cognitive tasks, but could also result in a high rate of dropouts. Therefore, we decided, for this study, to focus on the after-lunch condition.

Another limitation of the CoCo study is that it was not possible to use a double-blind design with a placebo condition. This leaves the possibility of subject and experimenter bias and is a common problem in food-based trials. However, the randomised, cross-over design eliminates variations between the subjects and reduces bias. In the present study, fasting was the control condition. However, fasting could also be viewed as intervention as the majority of students regularly eat lunch at school. In a recent review, which compared ten studies of adults for the impact of short-term fasting on cognition[26], results were equivocal. Although some studies showed no effects of fasting on cognition, others showed impairments in tasks related to psychomotor speed, mental rotation or executive function. However, these results are not transferable to the current study because of differences in cognitive tasks, fasting periods, time of fasting, time of day and the age of the participants.

Müller *et al.*[11] concluded that lunch effects in studies of adults might have been modified by the fact that the test meal size was larger or smaller than the usual lunch size. Although the test meal size was assessed in the CoCo study, it was not possible to assess probable differences with respect to the individual usual lunch size as well. Hence, future studies on cognitive effects of lunch should assess the usual size of lunch to examine the impact of deviation of the test lunch from habitual eating lunch size (smaller or larger) on cognitive performance.

A considerable number of participants did not completely comply with the study protocol (*n* 67). Reasons for this behaviour were not enquired, but it could be assumed that it was difficult for the children to restrain from eating during this time period, especially when it was explicitly forbidden. Apart from the questionnaire at the end of the test day and supervision in the school yard, children had opportunities for hidden snacking. Thus, it could not be fully ruled out that all participants who did not comply with the protocol were detected. In addition, it has to be mentioned that the sixth grade students of this intervention already participated as fifth grade students in the previous intervention. Therefore, they may have been familiar with two of the three cognitive tasks and were probably less motivated. However, any such effects should have been minimised as the differences between the test days were analysed in the cross-over design.

In conclusion, the present study confirmed the results of our previous studies[11,12] that school lunch does not seem to impair children's CF regarding the conducted tests on task switching, working memory updating and alertness. Although the previous studies[11,12] pointed to slight improvements in single cognitive parameters by lunch shortly after the meal, the current study did not indicate positive effects of lunch on cognition after a prolonged interval until early afternoon.

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The authors' contributions are as follows: L. L., M. K. and M. S. formulated the specific research question and the design of the study. M. F. designed the test battery for cognitive testing and helped with interpreting the results of cognitive testing. The study was carried out and data were analysed by M. S. and L. L. The article was written by M. S. and L. L. with input from K. M., M. F., M. K. and P. S. The final manuscript was read and approved by all the authors.

The authors declare that there are no conflicts of interest.

References

1. Craig A, Baer K & Diekmann A (1981) The effects of lunch on sensory-perceptual functioning in man. *Int Arch Occup Environ Health* **49**, 105–114.

2. Smith AP & Miles C (1986) Effects of lunch on selective and sustained attention. *Neuropsychobiology* **16**, 117–120.
3. Kanarek RB & Swinney D (1990) Effects of food snacks on cognitive performance in male college students. *Appetite* **14**, 15–27.
4. Smith AP & Miles C (1986) The effects of lunch on cognitive vigilance tasks. *Ergonomics* **29**, 1251–1261.
5. Monk TH (2005) The post-lunch dip in performance. *Clin Sports Med* **24**, e15–e23 xi–xii.
6. Pollitt E, Cueto S & Jacoby ER (1998) Fasting and cognition in well- and undernourished schoolchildren: a review of three experimental studies. *Am J Clin Nutr* **67**, 779S–784S.
7. Hoyland A, Dye L & Lawton CL (2009) A systematic review of the effect of breakfast on the cognitive performance of children and adolescents. *Nutr Res Rev* **22**, 220–243.
8. Adolphus K, Lawton CL & Dye L (2013) The effects of breakfast on behavior and academic performance in children and adolescents. *Front Hum Neurosci* **7**, 425.
9. Bellisle F (2004) Effects of diet on behaviour and cognition in children. *Br J Nutr* **92**, Suppl. 2, S227.
10. Müller K, Libuda L, Terschlüsen AM, *et al.* (2013) A review of the effects of lunch: on adults' short-term cognitive functioning. *Can J Diet Pract Res* **74**, 181–188.
11. Müller K, Libuda L, Gawehn N, *et al.* (2013) Effects of lunch on children's short-term cognitive functioning: a randomized crossover study. *Eur J Clin Nutr* **67**, 185–189.
12. Schröder M, Müller K, Falkenstein M, *et al.* (2015) Short-term effects of lunch on children's executive cognitive functioning: the randomized crossover Cognition Intervention Study Dortmund PLUS (CogniDo PLUS). *Physiol Behav* **152**, 307–314.
13. Craig A (1986) Acute effects of meals on perceptual and cognitive efficiency. *Nutr Rev* **44**, Suppl, 163–171.
14. Tukey JW (1977) *Exploratory Data Analysis. Vol. 1: Quantitative Methods, Reading.* Massachusetts, MA: Addison-Wesley.
15. Grizzle JE (1965) The two-period change-over design and its use in clinical trials. *Biometrics* **21**, 467.
16. Donohoe RT & Benton D (1999) Cognitive functioning is susceptible to the level of blood glucose. *Psychopharmacology* **145**, 378–385.
17. Owen L, Scholey AB, Finnegan Y, *et al.* (2012) The effect of glucose dose and fasting interval on cognitive function: a double-blind, placebo-controlled, six-way crossover study. *Psychopharmacology* **220**, 577–589.
18. Benton D, Brett V & Brain PF (1987) Glucose improves attention and reaction to frustration in children. *Biol Psychol* **24**, 95–100.
19. Wang SR, Chase HP, Garg SK, *et al.* (1991) The effect of sugar cereal with and without a mixed meal on glycemic response in children with diabetes. *J Pediatr Gastroenterol Nutr* **13**, 155–160.
20. Buss RW, Kansal PC, Roddam RF, *et al.* (1982) Mixed meal tolerance test and reactive hypoglycemia. *Horm Metab Res* **14**, 281–283.
21. Craig A & Richardson E (1989) Effects of experimental and habitual lunch-size on performance, arousal, hunger and mood. *Int Arch Occup Environ Heath* **61**, 313–319.
22. Smith A, Ralph A & McNeill G (1991) Influences of meal size on post-lunch changes inperformance efficiency, mood, and cardiovascularfunction. *Appetite* **16**, 85–91.
23. Isaacs E & Oates J (2008) Nutrition and cognition: assessing cognitive abilities in children and young people. *Eur J Nutr* **47**, Suppl. 3, 4–24.
24. Beaman CP (2005) Auditory distraction from low-intensity noise: a review of the consequences for learning and workplace environments. *Appl Cogn Psychol* **19**, 1041–1064.
25. Elliott EM (2002) The irrelevant-speech effect and children: theoretical implications of developmental change. *Mem Cognit* **30**, 478–487.
26. Benau EM, Orloff NC, Janke EA, *et al.* (2014) A systematic review of the effects of experimental fasting on cognition. *Appetite* **77**, 52–61.