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Adverse reactions to the Bacillus Calmette–Guérin (BCG) vaccine in new-born infants—an evaluation of the Danish strain 1331 SSI in a randomized clinical trial

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ABSTRACT

Objective: To evaluate adverse reactions of the Bacillus Calmette–Guérin (BCG) Statens Serum Institut (SSI) (Danish strain 1331) used as intervention in a randomized clinical trial.
Design: A randomized clinical multicenter trial, The Danish Calmette Study, randomizing newborns to BCG or no intervention. Follow-up until 13 months of age.
Setting: Pediatric and maternity wards at three Danish university hospitals.
Participants: All women planning to give birth at the three study sites (n = 16,521) during the recruitment period were invited to participate in the study. Four thousand one hundred and eighty four families consented to participate and 4262 children, gestational age 32 weeks and above, were randomized: 2129 to BCG vaccine and 2133 to no vaccine. None of the participants withdrew because of adverse reactions.
Main outcome and measure: Trial-registered adverse reactions after BCG vaccination at birth. Follow-up at 3 and 13 months by telephone interviews and clinical examinations.
Results: Among the 2118 BCG-vaccinated children we registered no cases of severe unexpected adverse reaction related to BCG vaccination and no cases of disseminated BCG disease. Two cases of regional lymphadenitis were hospitalized and thus classified as serious adverse reactions related to BCG. The most severe adverse reactions were 10 cases of suppurative lymphadenitis. This was nearly a fivefold increase compared to what was expected based on the summary of product characteristics of the vaccine. All cases were treated conservatively and recovered. Six of 10 (60%) families of children experiencing suppurative lymphadenitis compared to 117/2071 (6%) of those with no lymphadenitis indicated that the vaccine had more adverse effects than expected (p-value <0.001).
Conclusions and relevance: BCG vaccination was associated with only mild morbidity and no mortality. A higher incidence of suppurative lymphadenitis than expected was observed. All children were treated conservatively without sequelae or complications.
Trial registration: Trial registration number NCT01694108 at www.clinicaltrials.gov

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1. Introduction

The Bacillus Calmette–Guérin (BCG) vaccine is the most widely administered vaccine in the world, and it remains the primary prophylaxis against tuberculosis (TB). The protective effect of BCG vaccination is well studied, with highest efficacy against active disease, especially TB meningitis and disseminated TB in children, whereas there is great variation in the effect of preventing pulmonary TB [1,2]. BCG is part of the immunization program in most low-income countries with a high prevalence of TB and a recent study confirm that it is effective in preventing TB [3]. In most high-income countries BCG vaccination was discontinued as a consequence of reduced TB prevalence.

The adverse reactions (ARs) of BCG have been described thoroughly. The normal local reaction following intradermal BCG vaccination is swelling and redness which appears at the site of injection after a few weeks. This develops into a small pustule or an ulcer that heals and leaves a small scar after weeks to months. Local lymphadenopathy < 1 cm is also part of a normal reaction [4]. BCG is considered a safe vaccine and serious ARs are rarely seen [5].

Despite the fact that normal reactions and ARs after BCG vaccination are well described, studies report very different rates [6–8]. This could be due to different vaccination procedures and BCG strains. Early studies have shown that vaccination technique, dose, and preparation of the vaccine are important risk factors for adverse reactions [9–11], and that BCG strain, change of BCG vaccine, and HIV infection are also significant risk factors [12–14]. Differences in interpretation of a normal reaction and an AR could also explain the disparities; the normal local reaction and the 'adverse' local reactions represent a continuum of the same pathological process. The shift from a normal reaction to an AR is often defined by size which is difficult to assess accurately in a clinical setting.

Recent studies from West Africa have found that BCG, apart from providing some protection against TB, may also have beneficial non-specific effects on childhood mortality and morbidity [15,16]. Immunological studies have supported this by showing that BCG induces epigenetic changes at the monocyte level which lead to "trained innate immunity" [17]. To investigate these possible non-specific effects of BCG vaccination in a high-income country, we conducted a randomized clinical trial ('The Danish Calmette Study') from October 2012 to January 2015 in which 4262 Danish children at birth were randomized to BCG or no intervention [18]. Within this trial, we evaluated the ARs to BCG when administered in a setting with no routine BCG vaccination and low prevalence of HIV [19].

2. Methods

2.1. Study design

The Danish Calmette Study is a randomized clinical multicenter trial investigating the effect of BCG vaccination at birth on childhood morbidity in Denmark, where BCG was withdrawn in the early 1980s, due to low prevalence of TB. The study was carried out at three Danish university hospitals, Rigshospitalet, Hvidovre Hospital, and Kolding Hospital.

At the three study sites, 4262 children were randomized after birth to BCG vaccination or no intervention. The primary outcome was child morbidity assessed as all-cause hospitalization. Secondary outcomes were obtained by telephone interviews and clinical examinations at the age of 3 and 13 months. The study is described in detail elsewhere [18].

The three study sites were organized differently with respect to the randomization and vaccination procedure. One study site had predominately one midwife vaccinating; at this site most children were vaccinated more than 24 h after birth. At the other two sites the task was split between 11 and 15 midwives, respectively; most children here were vaccinated within the first 24 h of life. All study staff were trained specifically to administer BCG vaccination correctly.

2.2. Vaccination procedure

The BCG SSI (Danish strain 1331) was used in the study. A dose of 0.05 mL of the vaccine suspension was applied intradermally on the left upper arm. A sterile 1 mL syringe with a short (25-27 G) needle was used for the procedure.

2.3. Data collection

Before giving oral and written consent and again after the vaccination, the families were informed about the normal reaction following a BCG vaccination and were advised to seek more information at the Calmette Study homepage if needed [20]. The families were encouraged to contact the study facility in case of unexpected reactions or if they felt uneasy about a reaction. To collect information regarding study outcomes the families were contacted at 3- and 13-months by telephone and were subsequently scheduled for a clinical examination at the study site. During these contacts, information on ARs was collected only when mentioned by the families and never asked for actively by the study staff.

2.4. Classification of adverse events

All potential adverse events were classified into five categories according to standard Good Clinical Practice (GCP) guidelines: adverse event (AE), adverse reaction (AR), severe adverse event (SAE), severe adverse reaction (SAR), and suspected unexpected severe adverse reaction (SUSAR). For the present study, severity was defined by hospitalization also if the hospitalization was only briefly and due to anxiousness about an AR, and assessed according to international guidelines [21]. An AE was defined as a non-severe event not causally related to BCG vaccination. An AR was defined as a non-severe event that was expected after BCG vaccination. This includes suppurative lymphadenitis when the child was not hospitalized. An SAE was defined as a severe event not causally related to BCG vaccination. An SAR was defined as a severe event that was expected after BCG vaccination and led to hospitalization. An SUSAR was defined as a severe unexpected event with suspected relation to BCG [22]. This includes disseminated BCG disease. All cases of SAR and SUSAR were discussed with the data and safety monitoring board (DSMB) and reported to the national health authorities according to Danish law.

All BCG-vaccine related events were categorized into four categories: (1) local reactions including abscess at the injection site and prolonged pustule healing (AR), (2) regional lymphadenitis (AR; SAR if leading to hospitalization), (3) suppurative lymphadenitis (AR; SAR if leading to hospitalization), and (4) others. The distinction between regional lymphadenitis and suppurative lymphadenitis was based on the clinical finding of fluctuant swelling and/or secretion from a fistula. Most cases of suppurative lymphadenitis were confirmed by ultrasound examination [23]. All cases of suppurative lymphadenitis had preceding regional lymphadenitis, but were registered exclusively in the suppurative lymphadenitis category.

2.5. Deaths

All fatalities during follow-up were evaluated for potential relation to BCG.

Table 1
Risks of adverse reactions after BCG vaccination according to The Danish Calmette Study and the summary of product characteristics of BCG SSI.

Adverse reaction	The Danish Calmette Study		SPC of BCG SSI
	n/*N*	Risk(95%CI)	Risk
Regional lymphadenitis	13/2118	6.1/1000 (3.3/1000–1/100)	≥1/1000–<1/100
Suppurative lymphadenitis	10/2118	4.7/1000 (2.3/1000–8.7/1000)	≥1/10,000–1/1000

SPC: Summary of product characteristics; *n* = number of cases; *N* = number of vaccinated children.

2.6. Recording of adverse events

The study had a standard operating procedure (SOP) for the recording of adverse events. Evaluation during the study period revealed some major differences in the recording of adverse events. Since morbidity is an outcome in itself and registered elsewhere in the study, some study assistants did not simultaneously register AEs and SAEs in the adverse event log. Furthermore, according to the SOP, the ARs reported at the families' initiative during telephone interviews/clinical examinations should be reported in the e-crf, even though classified as a normal reaction after BCG vaccine; we experienced some inconsistency in this procedure. Distinction of an adverse reaction from a normal reaction according to size of a local abscess or lymphadenitis >1 cm proved difficult to assess on the telephone and the distinction were inconsistent during data collection. These site and interpersonal differences in the interpretation of the SOP imply that we were able to report solid data on the more severe ARs only, namely regional lymphadenitis, suppurative lymphadenitis, SARs, and SUSARs, which all study staff registered in a similar way.

2.7. Treatment of adverse reactions

An SOP for treatment of ARs, SARs, and SUSARs was distributed by the study to the pediatric departments at the three study sites to ensure that the three study sites would give a uniform and high standard of diagnosis and treatment of possible ARs.

2.8. Parents experience with adverse reactions

At the end of the 13-month follow-up interview the parents of the children in the BCG group were asked if the BCG vaccine had more adverse effects than they had anticipated and if they were satisfied with their decision of having their child vaccinated. Level of agreement was compared between families with and without regional and suppurative lymphadenitis.

2.9. Statistical analysis

Statistical analyses were performed using STATA 13.1 (Statacorp LP, College Station, TX, USA). All risk estimates were assessed relative to the summary of product characteristics (SPC) of the BCG SSI and previously reported studies.

3. Results

A total of 4262 children were randomized to BCG or control and 2118 children were BCG vaccinated within 7 days of life. Hospitalization and morbidity were study outcomes and will be reported elsewhere (Stensballe et al., submitted; Kjærgaard et al., submitted). Due to the limitations of the non-severe AR data, we are not able to report solid data on all events. Crude numbers of all registered events are shown as supplementary data, but comparison with data from other BCG studies should be done with caution **(S 1 + 2)**.

This report focuses on severe adverse reactions categorized as causally related to BCG vaccination. Two children, one with regional lymphadenitis and one with suppurative lymphadenitis, were briefly hospitalized for ultrasonic examination and were therefore categorized as SARs. They were discharged with conservative management. The severity and time course did not differ from the other children with regional and suppurative lymphadenitis registered as ARs. No child had disseminated BCG disease. No cases of SUSARs were seen. The most severe adverse reaction seen was suppurative lymphadenitis.

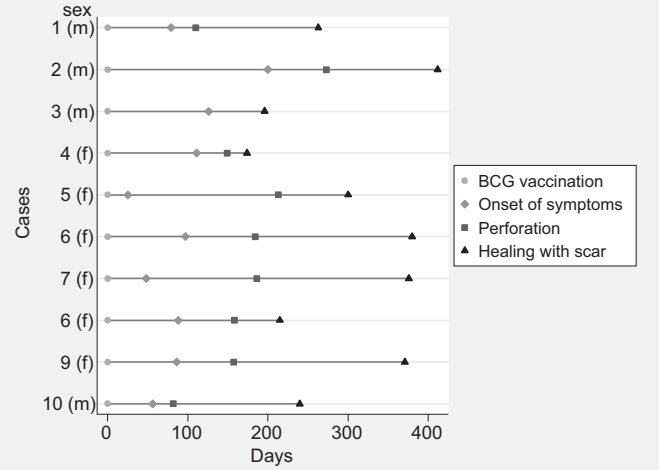

Fig. 1. Time course of 10 cases of suppurative lymphadenitis after BCG vaccination within The Danish Calmette Study. In case 3 it was not possible to determine the exact time of perforation

3.1. Suppurative lymphadenitis

Suppurative lymphadenitis was registered for 10 of the 2118 vaccinated children in the study (4.7/1000, [95% CI 2.3/1000–8.7/1000]) (Table 1). All children had lymphadenitis in the left axilla as their primary symptom. This occurred at a median age of 87 days, ranging from 25 to 200 days. All cases of suppurative lymphadenitis perforated spontaneously and healed with a scar in the left axilla. The median time from onset of symptoms to development of a scar was 198 days, ranging from 63 to 328 days (Fig. 1, Table 2). All 10 children were regularly examined and treated conservatively. Apart from the time of perforation, none of the children showed signs of pain and were in general unaffected by their suppurative lymphadenitis.

Table 2
Characteristics of children with suppurative lymphadenitis after BCG vaccination within The Danish Calmette Study.

	N = 10, median (range)
Sex (m)	4
Age at BCG vaccination (days)	0 (0–6)
Age at onset of symptoms (days)	87 (25–200)
Time from onset of symptoms to perforation (days)	71 (26–188)
Time from perforation to healing (days)	153 (25–214)
Time from onset of symptoms to healing (days)	198 (63–328)

Table 3a
Parental experience of side effects from BCG when asked 13 months after vaccination. BCG vaccinated children within The Danish Calmette Study.

BCG had more side effects than we expected?	All	Participants with suppurative lymphadenitis		Participants with regional lymphadenitis	
	N = 2071(%)[a]	N = 10(%)	p-value[b]	N = 12(%)[c]	p-value[b]
Agree or strongly agree	117(6)	6(60)	<0.001	3(25)	0.004
Neither nor, disagree or strongly disagree	1942(94)	4(40)		9(75)	

[a] Only participants not experiencing suppurative or regional lymphadenitis.
[b] Chi-2 test for comparison with BCG vaccinated participants not experiencing suppurative or regional lymphadenitis.
[c] One family with regional lymphadenitis did not answer the questions.

Two different batches were used in the study, with a suppurative lymphadenitis risk of 3/923 (3.3/1000) and 7/1195 (5.9/1000) vaccinated, respectively (*p*-value for same risk = 0.40). The mother had been BCG-vaccinated in 20% (2/10) of the children with suppurative lymphadenitis. This was comparable to 17.6% for all BCG-vaccinated children.

Non-suppurative regional lymphadenitis was experienced by 13 of the 2118 vaccinated in the study (6.1/1000, [95% CI 3.3/1000–1/100]) (Table 1).

3.2. Death during follow-up

Four children died during the 13 months follow-up period of The Danish Calmette Study (0.1%). Three were vaccinated and one was in the control group. None of the cases were considered to be related to the BCG vaccination when evaluated by the local health authorities and the DSMB of the Calmette Study **(S 3).**

3.2.1. Case 1: Girl vaccinated on the day of birth

The girl was admitted to the pediatric ward the same day with hypoglycemia and a heart murmur. Diagnosed with persistent truncus arteriosus and died 19 days old after surgery for truncus arteriosus.

3.2.2. Case 2: Girl vaccinated two days old

The girl was admitted 27 days old with apathy and a distended abdomen. Diagnosed with congenital hepatic mesenchymal hamartoma and died after cardiovascular collapse 29 days old.

3.2.3. Case 3: Boy vaccinated one day old

The boy was admitted 78 days old after the child was found lifeless in bed. Autopsy did not reveal the cause of death and the child was diagnosed with sudden infant death syndrome. The DSMB and the responsible doctors did not consider BCG vaccination to be related to the death.

3.2.4. Case 4: Boy randomized to the control group

The boy was hospitalized two days old with apathy and hypotonia. Genetic investigation diagnosed the child with Zellweger syndrome. The child died 320 days old.

3.3. Parents experience with adverse reactions

When asked whether they agreed with the statement: "The BCG vaccine had more adverse effects than we had anticipated", 6% of the parents agreed or strongly agreed. There was no significant difference between parents of boys and girls (5.7% vs. 6.5%, *p*-value 0.40). In children with suppurative lymphadenitis or non-suppurative regional lymphadenitis, 6/10 (60%) and 3/12 (25%), respectively, indicated to agree or strongly agree. These experiences differed significantly from those of parents whose did not experience suppurative or regional lymphadenitis (*p*-values <0.001 and 0.004, respectively) (Table 3a).

When asked whether they agreed with the statement: "We are satisfied with our decision of having our child BCG vaccinated", 94% of the parents of the overall BCG-vaccinated study population agreed or strongly agreed. There was no significant difference between parents of boys and girls (94% vs. 93%, *p*-value 0.50). In children with suppurative lymphadenitis, or regional lymphadenitis, 70% and 83%, respectively, agreed or strongly agreed (*p*-values were 0.003 and 0.20 compared with those of parents who did not experience suppurative or regional lymphadenitis) (Table 3b).

4. Discussion

In The Danish Calmette Study, we found no SUSARS, and no disseminated BCG disease cases among more than 2000 vaccinated children. We found a nearly fivefold higher risk of suppurative lymphadenitis than anticipated based on the risk of ≥1/10,000 to 1/1000 reported in the SPC for the BCG SSI vaccine whereas the risk of non-suppurative lymphadenitis was within the range of expected events compared to the risk of ≥1/1000 to 1/100 reported by the SPC [4]. The children with suppurative lymphadenitis were treated conservatively and all healed without sequelae. Four children died during follow-up without a causal relation to the BCG vaccination. Six percent of families stated that the BCG had more adverse effects than they had expected.

4.1. Strengths

The Danish Calmette Study had a followup rate of 98% (Kjærgaard et al., submitted) and all the study participants lived in the catchment area for the hospitals hosting the study sites.

Table 3b
Parental satisfaction with having their child vaccinated with BCG when asked 13 months after vaccination. BCG vaccinated children within The Danish Calmette Study.

We are satisfied with our decision of having our child BCG vaccinated?	All	Participants with suppurative lymphadenitis		Participants with regional lymphadenitis	
	N = 2055(%)[a]	N = 10(%)	p-value[b]	N = 12(%)[c]	p-value[b]
Agree or strongly agree	1922(94)	7(70)	0.003	10(83)	0.2
Neither nor, disagree or strongly disagree	133(6)	3(30)		2(17)	

[a] Only participants not experiencing suppurative or regional lymphadenitis.
[b] Chi-square test for comparison with BCG vaccinated participants not experiencing suppurative or regional lymphadenitis.
[c] One family with regional lymphadenitis did not answer the questions.

Thus, we are fairly confident that the reported risk of suppurative lymphadenitis is accurate.

4.2. Limitations

The limitations of this study are the diverse reporting and recording of less severe ARs leaving no robust estimates for overall risk of ARs. As this study only relies on parental reporting of adverse effects there is a risk of underreporting of adverse reactions that a focused clinical examination would have revealed. There is a potential risk that some of the participants withdrew from the study before the occurrence of an adverse reaction. As only 2% were lost to follow-up and all the study participants lived in the catchment area for the hospitals hosting the study sites we believe that the risk of bias to the outcome is low.

4.3. Comparison with other studies

We found the risk of suppurative lymphadenitis to be almost five times higher than expected. Previous studies have reported an increase in the incidence of ARs when changing BCG vaccine strain and several studies report such changes when shifting to the BCG SSI strain 1331 [6,24–26]. In many cases, change of vaccine strain also leads to change in dose or techniques, which are both important factors in the development of ARs [13,27]. Other studies report batch variation within the same strain of BCG. This was described in Singapore. A batch-related issue led to a 3–4 fold rise in the incidence of suppurative lymphadenitis but then returned to usual incidences. The spike of suppurative lymphadenitis was most likely due to manufacturing issues [28]. We found no significant difference between the two batches used in our study.

It could be speculated that the increased risk of suppurative lymphadenitis observed in the present study was because most mothers were not BCG vaccinated themselves [29–32]. However, the prevalence of maternal BCG vaccination in children with suppurative lymphadenitis (2/10 children had BCG vaccinated mothers) was comparable with that in the rest of the study population (17%).

All children with suppurative lymphadenitis were examined at the pediatric department and followed as outpatients in collaboration with the principle investigator of The Danish Calmette Study. None of the children received medical or surgical treatment and all lesions healed with no other sequelae than a scar in the left axilla. This illustrates that though BCG-induced suppurative lymphadenitis often is prolonged, it is benign and well-responsive to conservative treatment [33–35].

No disseminated BCG-infections occurred. In the pre-HIV-era, incidences of 1.9/mil (1955–1974) and 4.29/mil (1979–1981) among infants <1 year old were reported [36]. Many studies have shown that BCG vaccination can result in serious complications in children with HIV infection, and other immunosuppressive diseases [8,14,37]. A report of increasing incidence of suppurative lymphadenitis in Saudi Arabia [38] lead to concern about the use of BCG in a setting with a 20 times higher incidence of severe combined immunodeficiency (SCID) than most European countries[39]. However, in a Danish setting with low incidence of HIV and of primary immunodeficiency disorders, the risk of severe complication seems negligible, when care is taken to avoid vaccinating immunocompromised patients by awaiting HIV status in children of HIV infected mothers.

In this study we were unable to report an overall AR rate; we did however find that only 6% of the parents experienced the adverse effects to exceed what they had expected. Prior studies of the BCG-vaccine revealed varying rates of ARs: A South African study, evaluating BCG SSI strain 1331, reported an overall AR rate of 3.1/100, whereas an Australian study found an overall AR rate of 5/100 [40,41]. In France, an overall incidence of 18/100 in a study using active case finding at 4 and 12 months after vaccination were reported [7]. Active case finding may explain the high incidence with an overreporting of what would have been considered normal reactions in other studies with passive case finding like ours. Clear definitions of how to report and register ARs are important, especially when comparing studies.

A previous study reported different risk rates between boys and girls with a higher incidence in girls. This could represent a sex differential adverse effect of the BCG vaccine [41]. It could on the other hand also represent an over-reporting of local reactions by parents of girls, as other studies among schoolchildren have shown that girls are more worried about scar development than boys, which most likely would be reflected in the parents' threshold for reporting a local reaction [42]. In our study, parents of girls did not experience more adverse effects than they expected compared to parents of boys.

4.4. Perspectives

Clear definitions of how to report and register ARs are important. Developing standard information for recipients of BCG, describing the normal reaction to BCG, and classifying ARs as reactions that lead to a health care contact could be a possibility. With the emerging focus on NSEs by WHO, more clinical trials on BCG could be expected. If BCG is reintroduced in a high-income setting with low prevalence of TB and HIV this paper adds to the field that: (1) BCG is safe with only well described adverse effects, (2) suppurative lymphadenitis should be expected to be more common than stated in the SPC, and (3) treatment of suppurative lymphadenitis can be handled conservatively.

5. Conclusion

We found higher incidence rate of suppurative lymphadenitis than reported elsewhere. All children were treated conservatively and healed without sequelae. This suggests that long-time treatment with antibiotics and/or surgery can be avoided.

Authors' contributions

LGS, GG, PEK, OP, DJ, TH, PA, and CSB supervised the collection of data and NMB, JK, TNN, GTP, and LMT participated in the data collection. TNN carried out data management, analyzed the data, and drafted the manuscript. All authors revised the manuscript and approved the final version.

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Conflict of interest

All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and all authors declare to have no potential conflict of interest.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.vaccine.2016.03.100.

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