Immunogenicity and protection from a single dose of internationally available killed oral cholera vaccine: a systematic review and meta-analysis

Anna Lena Lopez[1,2], Jacqueline Deen[1,2], Andrew S. Azman[3], Francisco J Luquero[4], Suman Kanungo[5], Shanta Dutta[5], Lorenz von Seidlein[2,6], David A Sack[2]

1 Institute of Child Health and Human Development, National Institutes of Health, University of the Philippines – Manila, Philippines

2 Delivering Oral Vaccine Effectively (DOVE), Department of International Health, Johns Hopkins University, Baltimore, MD, USA

3 Department of Epidemiology, Johns Hopkins University, Baltimore, MD, USA

4 Epicentre, Paris, France

5 National Institute of Cholera and Enteric Diseases, Beliaghata, Kolkata, West Bengal, India

6 Mahidol-Oxford Tropical Medicine Research Unit, Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand

Corresponding author:	**Alternate corresponding author:**
Jacqueline Deen, MD, MSc	Anna Lena Lopez
University of the Philippines	University of the Philippines
Manila, Philippines	Manila, Philippines
E-mail: deen.jacqueline@gmail.com	E-mail: annalenalopez@gmail.com
Telephone +66 81-110-3809	Telephone: +63 917-861-4496

Main point: Current immunologic and clinical data suggest that protection conferred by a single dose of killed OCV may be sufficient to reduce short-term risk in outbreaks or other high-risk settings, which may be especially useful when vaccine supply is limited.

Abstract

In addition to improved water supply and sanitation, the two-dose killed oral cholera vaccine (OCV) is an important tool for the prevention and control of cholera. We aimed to document the immunogenicity and protection (efficacy and effectiveness) conferred by a single OCV dose against cholera. The meta-analysis showed an estimated 73% and 77% of individuals seroconverted to the Ogawa and Inaba serotypes, respectively, after an OCV first dose. The estimates of single-dose vaccine protection from available studies are 87% at 2 months decreasing to 33% at 2 years. Current immunologic and clinical data suggest that protection conferred by a single dose of killed OCV may be sufficient to reduce short-term risk in outbreaks or other high-risk settings, which may be especially useful when vaccine supply is limited. However, until more data suggests otherwise, a second dose should be given as soon as circumstances allow to ensure robust protection.

Key words: cholera, oral cholera vaccine, cholera vaccine, *Vibrio cholerae*

Introduction

Cholera is an acute watery diarrheal disease that can spread rapidly and lead to widespread outbreaks. An estimated 2.9 million cholera cases occur annually in endemic countries [1]. Improved water, sanitation and hygiene (WASH) is the cornerstone for cholera prevention and control but the world is falling short of meeting these targets [2]. Displacements due to natural disasters or conflicts and population growth will result in continuing cholera outbreaks in the future unless preventive measures are applied.

In parallel with WASH, timely treatment and community engagement, the World Health Organization (WHO) recommends that oral cholera vaccination be considered in areas where the disease is endemic, as part of the response to outbreaks or in a humanitarian crisis where there is a high risk of cholera [3]. There are three internationally-available, killed oral cholera vaccines (OCV). The first is a monovalent (*Vibrio cholerae* O1) whole cell OCV containing recombinant B-subunit, marketed as Dukoral (Valneva, Lyon, France). Randomized, placebo-controlled trials of earlier versions of Dukoral in Bangladesh showed a two-dose protective efficacy at one and three years of follow-up of 74% [4] and 64% [5], respectively. Dukoral was the first OCV that was internationally-licensed in 1991 and WHO-prequalified in 2001, but it is relatively expensive and requires a buffer for administration. Dukoral is primarily used by travellers. The second vaccine is Shanchol (Shantha Biotechnics Ltd, Hyderabad, India), a bivalent (*V. cholerae* O1 and O139) whole-cell OCV. A randomized, placebo-controlled trial in India showed that a 2-dose regimen confers 67% protective efficacy against cholera within two years of vaccination [6], 66% at three years [7], and 65% at five years [8]. Continued protection up to five years in this endemic setting may have been due to boosting from natural exposure. Shanchol was licensed in India in 2009 and received prequalification from the WHO in 2011. WHO and its partners established an OCV stockpile [9] through which about 4 million doses of Shanchol have been deployed to date, in mass vaccination campaigns in 11 countries but demand exceeds supply [10, 11]. A third

vaccine is Euvichol (Eubiologics, Gangwon-do, South Korea), another bivalent-OCV based on the same formulation as Shanchol. At the time of writing, there is no published clinical efficacy data for Euvichol, but following a Phase I trial in Korea [12] and a bridging non-inferiority immunogenicity study in the Philippines [13], Euvichol was licensed and WHO-prequalified in 2016. WHO-prequalification is necessary for the purchase of vaccines by UN agencies, including UNICEF.

Two-dose regimens are recommended for Dukoral, Shanchol and Euvichol but delivering two doses can be challenging during emergency situations. The difficulties include accessing the same population twice, maintaining vaccine storage and retaining vaccination staff during the inter-dose period. Also, the response lag combined with the shortened duration of outbreaks after a first dose is given may render the additional protection conferred by a second dose less important. Previous modeling suggests that reactive vaccination campaigns using a single dose of OCV may prevent more cases and deaths than a two-dose campaign when vaccine supplies are limited, while at the same time reducing logistical complexity [14]. Our primary question of interest is how well a single-dose regimen of killed OCV protects against cholera. As there are relatively few studies that document OCV efficacy and effectiveness in conferring protection against disease and since vaccine-induced increase in vibriocidal antibody titer has been linked with protection [15], we included both a systematic review of the efficacy and effectiveness data and a systematic review and meta-analysis of a larger body of immunologic response data.

Methods

This systematic review and meta-analysis was conducted according to Preferred Reporting Items for Systematic Review and Meta-Analyses guidelines [16]. We searched PubMed, the Cochrane Central Register of Controlled Trials and Scopus from 1 January 2005 to 13 November 2016 combining MeSH and free-text terms for the following: "killed cholera vaccine", "oral cholera

vaccine", OCV, immun*, serolog*, "immune response", "serologic response", vibriocidal, protect*, efficac* and effective*, without any language or age restrictions. The search was limited to the past decade, from 2005 onwards, to only include studies on the currently available vaccine formulations (Dukoral underwent formulation changes before international licensure and WHO-prequalification). We also contacted public health personnel and experts in the field to identify unpublished documents such as meeting presentations to ensure completeness. The detailed search strategy is shown in Supplementary Figure 1 (see Supplementary data, page 6).

Titles and abstracts were compiled in Endnote X6 (Thomson Reuters, Philadelphia, PA, USA). Two authors (ALL and JD) screened the list of titles and abstracts independently to ensure that they included information on immunogenicity as measured by vibriocidal antibodies or protection following one dose of current formulations of internationally-available killed OCV (Dukoral, Shanchol and Euvichol). Case reports, animal studies, socio-behavioral studies, economic evaluations, and articles that did not report original research (e.g. comparison of previously reported data, reviews, modeling studies, correspondence, and editorials) were excluded. The full-text of eligible articles were downloaded and reviewed in detail. Immunogenicity and vaccine protection (efficacy or effectiveness) data were extracted and analyzed. Basic analysis and data summarization was done in Microsoft Excel 2011 (Seattle, WA, USA). We used the GRADE guidelines to assess the risk of bias [17] and the quality of evidence [18] of the included articles, as shown in the Supplementary Table 1 (see Supplementary data, pages 1-3).

Immunogenicity

From the reports on vaccine immunogenicity, we tabulated the number and age group of participants, study location, vibriocidal baseline geometric titers, the geometric mean fold rises (GMF-rise) after a first and subsequent vaccine dose, and the number and percentage of participants who seroconverted after a first and subsequent dose. Following convention,

seroconversion was defined as a ≥4-fold rise from baseline. Vibriocidal responses to Inaba and Ogawa were tabulated separately. Vibriocidal responses to O139 were not included in this analysis because of the variability in the laboratory testing among different groups and because its utility as an immunologic correlate of protection remains unknown [19]. Furthermore, outbreaks of cholera due to *V. cholerae* O139 have not been reported during the past decade, although sporadic cases have been identified [20]. We performed a meta-analysis of the proportion of individuals seroconverting after one- and two-doses of OCV, using binomial-normal random effects regression models [21], including baseline geometric mean titer as a covariate. The meta-analysis focused on the bivalent vaccines (Shanchol and Euvichol) since they are currently the most commonly used OCVs and because of the sparse data available on Dukoral. Geometric mean titers were centered at the mean and scaled (by the standard deviation) and the pooled seroconversion estimates presented assumed the mean GMT value. Three age groups were modeled separately for each serotype, in addition to models combining all age groups. Studies varied in the reporting of results by age groups; for the purpose of the meta-analysis we classified results from participants 15 years of age and older (including those with only 18 years and older) as from "adults", those from participants under 18 years as from "children" and those from participants 5 years of age or younger as from "young children." We estimated the I^2 statistic as a measure of unexplained heterogeneity between studies. Meta-analyses were performed with the *metafor* package in R (version 3.2.3) and data used are available at https://github.com/scottyaz/singledose-immuno-review.

Vaccine protection

We included studies on killed OCV efficacy and effectiveness against cholera. We defined vaccine efficacy as the protection conferred under ideal conditions of a randomized controlled trial, whereas vaccine effectiveness is the protection when the vaccine is given under actual public health situations, assessed by observational studies. From the reports on vaccine

protection, we tabulated data on study design, site and year, intervention, study population and vaccine coverage, primary assessment of protection (clinical endpoints and definitions), total number of cholera cases, main infection serotypes and biotypes, estimated vaccine efficacy or effectiveness after a single dose with sub-analysis by diseases severity or age-group, and duration of follow-up. We plotted the estimated vaccine efficacy or effectiveness by duration of follow-up.

Results

We identified 422 records on killed oral cholera vaccine immunogenicity or protection, 421 through the database search and one meeting presentation (Figure 1). We removed 228 duplicates and screened the titles and abstracts of 198 articles, of which 145 (75%) were excluded and 49 (25%) full text articles were downloaded and reviewed. Of these, 23 studies fulfilled the inclusion criteria and were included in the systematic review: 17 articles and one presentation on single-dose vaccine immunogenicity and 6 articles on single-dose vaccine protection (see references listed in Supplementary data, pages 4-5). General descriptions, risk of bias within the study [17], main biases and quality grading scores [18] of the 23 included studies are shown in the Supplementary Table 1 (see Supplementary data, pages 1-3).

Immunogenicity

Four articles reported on the immunogenicity of the monovalent OCV containing recombinant B-subunit (Dukoral), while 13 reported on the bivalent OCV, Shanchol and Euvichol (see references listed in Supplementary data, pages 4-5). Two articles reported on the persistence of antibodies up to one year following two doses of the monovalent vaccine (see references listed in Supplementary data, pages 4-5). The baseline titers and vibriocidal immune response to Ogawa and Inaba serotypes following killed oral cholera vaccination are shown in Tables 1 and 2, respectively. Baseline titers varied considerably by age group and across the study sites, with the highest in Kolkata, India and the lowest in Chungnam, Korea. We found that 43% to 95% of

participants seroconverted to the Ogawa serotype and 52% to 90% to the Inaba serotype following the first dose.

The results of the meta-analysis are shown in Figures 2, 3 and 4. Overall, the median proportion of individuals seroconverting after the first dose of a bivalent OCV was 73% (95%CI 67-78%, I^2=85.4%) to the Ogawa serotype and 77% (95%CI 73-81%, I^2=64.3%) to the Inaba serotype (Figure 2). In the sub-analysis of study populations with young children, children and adults, the median proportion of individuals seroconverting to Ogawa after the first dose was 67% (95% CI 61-72%), 80% (95% CI 74-85%), 71% (95% CI 64-77%), respectively (Figure 3 - A, B and C). The responses by age group to the Inaba serotype were consistent with that to the Ogawa serotype (Supplementary Figure 2 –A, B and C).

Overall, in adults and in children, a second dose of OCV did not increase the median seroconversion compared to the response to the first dose (Figures 2, 3 - C and – B). However, in the analysis limited to young children (Figure 3 – A), the proportion seroconverting to Ogawa increased from 67% (95% CI 61-72%, I^2=0) after dose 1 to 85% (95% CI 79-89%, I^2=0) after dose 2 (p-value = 0.002 for difference after adjusting for baseline GMT) with a smaller and statistically insignificant increase for Inaba (p-value= 0.12) Supplementary Figure 2-A).

Only one article reported on the immunogenicity among infants (using the monovalent OCV containing recombinant B-subunit, Dukoral) [22], which showed a significantly higher GMF-rise after the second dose only among the 6 to 9-month-old age group and not in the 10-18-month-old age group. One report on the immune response five years after a 2-dose vaccination schedule [23] showed no significant differences in baseline titers among those previously vaccinated compared to those who had not received the vaccine in the past.

Alam, et al, compared the vibriocidal responses of one- and two-dose recipients with 70 adult cholera patients [24]. Adult cholera patients had a 96 GMF-rise from baseline (considered as the

second day of presentation) in vibriocidal response to the homologous serotype as the infecting organism and 93% of patients had a ≥4-fold rise from baseline 30 days later. Vibriocidal titers gradually declined until at 1 year of follow-up when 43% still had ≥4-fold titer from baseline. In comparison, 43 to 50% of vaccinated adults seroconverted (≥4-fold titer from baseline) after the first dose with a 5-fold GMF-rise by day 3 (Table 1). Leung, et al, reported on the vibriocidal responses of comparably aged pediatric patients [25]. Cholera patients who were aged 2-5 years had the highest GMF-rise from baseline on the seventh day of presentation. Although increases from baseline were comparable in all age groups, vibriocidal antibodies waned earlier and were at baseline levels on the 42nd or 90th day after the first dose [26].

Vaccine protection

We identified one randomized study and five observational studies that reported on single dose protection (see references listed in Supplementary data, pages 4-5). The study design, site and year, intervention, study population and vaccine coverage, primary assessment of protection (clinical endpoints and definitions), total number of cholera cases, main infection serotypes and biotypes, estimated vaccine efficacy or effectiveness after a single dose with sub-analysis by diseases severity or age-group (when available) and duration of follow-up are shown in Table 3. There was one study on the monovalent OCV containing recombinant B-subunit (Dukoral) [27], while five reported on bivalent OCV (Shanchol) (see references listed in Supplementary data, pages 4-5). A single-dose of killed OCV conferred 87% protection at 2 months, declining to 33% at 2 years of follow-up (Table 3 and Figure 4). There was only one study that assessed vaccine protection by age group; the single-dose protection in young children was considerably lower than in older children and adults [28].

Discussion

We found a significant immunologic response to a single dose of killed OCV. Following a titer rise post-first dose, there was little change after a subsequent dose, when given within 14 to 28 weeks after the first. The exception is in young children where a second dose substantially increased the proportion seroconverting to the Ogawa serotype (p-value=0.002). The OCV immune response correlated with the evidence of single-dose protection, which is lowest in young children according to the single randomized controlled trial that has so far been conducted [28]. Overall, single-dose protection was highest soon after vaccination with waning over time. These data indicate that although a single dose of killed OCV may confer a lower protection of shorter duration compared to two doses, it may be adequate for situations when immediate protection from cholera is needed.

This aggregated review is warranted since the killed OCVs are highly related in terms of composition. However, this study has several limitations. First, the vibriocidal assay procedures varied across the studies raising concerns about combining and comparing results. Due to differences in methodology, the GMF rise (and related measures) may be the most practical parameter for comparison since it is a relative measure less affected by inter-laboratory variability. Aside from differences in assay procedures, age and baseline titers in endemic and non-endemic locations may influence the immune response to OCV, as has been noted previously [29]. The relatively high proportion of variance in first-dose seroconversion explained by heterogeneity between studies as opposed to within study sampling variance, or I^2, is likely a result of these differences between settings and laboratory methods. The blurring of the assessment of vaccine immunogenicity by pre-vaccination titers has also been reported for other vaccines [30]. But we could not detect any clear patterns in the responses to the first and second dose by study location. Age group and setting may similarly affect vaccine protection estimates due to immune status and the presence or absence of on-going natural exposure. Second, the immunogenicity data was analyzed in overlapping age distributions. Ideally, discrete age groups

should have been used but this was not possible due to data presented by varying age groups in the publications. Third, although seroepidemiological and human challenge studies have shown the association of serum antibody levels with protection,[15], there is no established immunologic correlate of protection (a "protection threshold") for OCVs. As indicated by our review, the relationship of vibriocidal antibodies to protection may be particularly uncertain in young children. The vibriocidal antibody response may be an imperfect indicator of protection but it is currently the most commonly used marker of an immune response to OCV [15, 31]. Detection of antibody secreting cells (ASCs) against *V. cholerae* lipopolysaccharide and O-specific polysaccharide in blood has also been done [32, 33] but requires more technically challenging procedures. Fourth, except for the Bangladesh trial [28], data on protection conferred by a single dose are from observational studies. The Bangladesh single-dose efficacy trial is therefore quite important as it confirms findings from observational studies. Fifth, protection data are based on only six clinical studies, with the highest estimate coming from a study in Juba, South Sudan measuring protection within the first two months after vaccination [34]. In this study, the apparent protection from a single dose may have in part reflected boosting of immunity from natural exposure that occurred during a large epidemic the year of the study and/or the previous year. However, the immunogenicity data summarized here do suggest that protection is likely to begin within 2-weeks of the first dose thus lending support to these short-term protection estimates. Sixth, and most importantly, this review is unable to assess exactly when protection from a single-dose starts, the additional boosting that a second dose provides or the interval between vaccine doses that maximizes the duration of protection. Aside from one study that looked into a 28-day interval between dosing[19], no other studies that used vibriocidal antibodies assessed longer dosing intervals. In a study using ASC to measure immunogenicity, robust responses were induced after a first but not after a second dose of the bivalent OCV given 14 days later, suggesting that the current dosing schedule may not be optimal for inducing an anamnestic response [32]. To maximize the benefits of the second dose, it is critical to establish the interval

between vaccine doses that confers the best and longest protection. Future studies comparing post-first and post-second dose immune kinetics may shed more light on these questions.

Protection, even short-term, from a single dose of OCV provides a way forward in considering alternative vaccine strategies to contain cholera outbreaks. For example, during cholera outbreaks with logistic challenges or insufficient OCV doses, single-dose coverage of a population at high-risk for cholera using all immediately-available vaccine could be implemented rapidly, followed by administration of a second dose when feasible [34, 35]. A second possibility, if the population at risk in an endemic area is too large to be covered, is the rapid door-to-door administration of a single-dose using a ring vaccination strategy during localized outbreaks to provide protection of contacts of index cases [36]. Although it is not known how quickly vaccinees are protected, this period may be short in endemic areas with on-going exposure to *V. cholerae*. Vaccination of secondary and tertiary contacts may prevent spread of the disease, even if the vaccine does not protect the primary contacts of the index case who have already been exposed and infected, but this assumption would need to be assessed for feasibility and effectiveness. The second dose could potentially be self-administered or deployed through vaccination posts at a later date [37].

When considering the use of single-dose OCV, it should be kept in mind that the meta-analysis of the immunogenicity data and the subgroup analysis of the Bangladesh study showed lower immune response and inadequate protection among children less than 5 years of age. Based on principles of cocooning [38], oral cholera vaccination of older children and adults around those too young to be vaccinated or to mount an adequate response could be beneficial. While both one and two doses of killed OCV appears to be less protective for young children, there is evidence for substantial indirect protection for these children when a large proportion of older persons in the community are vaccinated [39].

Notes section:

Contributors

ALL and JD conducted the literature search, compiled and screened the list of articles, selected the relevant documents, reviewed the eligible articles and extracted the data. All authors provided additional data sources. ALL, JD and ASA analyzed the information and interpreted the data. ALL, JD and ASA wrote the initial draft. JD and ASA prepared the figures. All authors contributed to the writing and revision of the text and finalization of the manuscript.

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Conflicts of interest

We declare that we have no conflicts of interest.

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Table 1: Vibriocidal immune response to Ogawa serotype following killed oral cholera vaccination (see references listed in Supplementary data, pages 4-5)

Study	Number and age group of participants	Study site	Baseline geometric mean titres	Interval between doses	No [%] that seroconverted after:		GMF rise after:	
					1st dose[a]	2nd dose[a]	1st dose[a]	2nd dose[a]
Monovalent OCV containing recombinant B-subunit								
Ahmed, 2009	49 [10-18 months] 47 [6-9 months]	Dhaka, Bangladesh	NA[b]	14 days 14 days	NA[b] NA	[56] [57]	4 2	5 5
Alam, 2011	30 Adult 1- dose recipients 30 Adult 2-dose recipients	Dhaka, Bangladesh	27 26	NA[b] 14 days	[50][c] [43][c]	NA[b,d] [36][e]	5-fold by day 3[c] 5-fold by day 3[c]	5-fold by day 17[e] 5-fold by day 17[e]
Leung, 2012	20 Children aged 2- 5 years 20 Children aged 6-17 years	Dhaka, Bangladesh	37 32	14 days 14 days	0 [35]	[65] [90]	2-fold by day 3[c] 3-fold by day 3[c]	23-fold by day 7[f] 42-fold by day 7[f]
Bivalent OCV								
Saha, 2011	53 [18-45 years] 55 [2-4 years] 54 [12-24 months]	Dhaka, Bangladesh	61 39 9	14 days	[70][f] [75][f] [78][f]	[59][f] [75][f] [74][f]	7[f] 9[f] 6[f]	NA[b] NA[b] NA[b]
Charles, 2014	23 Adults 45 [6 to 17 years] 42 [1-4 years old]	Haiti	14 21 14	14 days	[77] [f] [69] [f] [64] [f]	[91][f] [74][f] [73][f]	19[f] 11[f] 9[f]	19 [f] 10 [f] 10 [f]
Kanungo, 2015	96 [15+ years] 90 [6-14 years]	Kolkata, India	329 102	14 days 14 days	57 [59] 79 [88]	51 [53] 65 [66]	6 22	5 13

Study	Number and age group of participants	Study site	Baseline geometric mean titres	Interval between doses	No [%] that seroconverted after:		GMF rise after:	
					1st dose[a]	2nd dose[a]	1st dose[a]	2nd dose[a]
	91 [15+ years][g]		475	14 days	50 [55]	37 [41]	4	3
	93 [6-14 years][g]		236	14 days	65 [70]	67 [72]	16	9
Kanungo, 2015	86 Adults [≥18 years]	Kolkata, India	364	14 days	48 [56]	39 [45]	6	4
			359	28 days	52 [62]	41 [49]	6	4
	83 Children [1-17 years]		125	14 days	63 [75]	61 [73]	19	11
			131	28 days	65 [79]	59 [72]	16	8
Aloysia, 2015	112 [15+ years]	Philippines	69	14 days	[78]	[69]	14	11
	112 [5- 14 years]		18		[86]	[88]	48	43
	112 [1-4 years]		3		[72]	[96]	61	82
Desai, 2015	37 [18+ years]	Ethiopia	24	14 days	24 [65]	26 [70]	13	13
	45 [1-17 years]		4		36 [80]	38 [84]	35	35
Ivers, 2015	25 [HIV +ve adults]	Haiti	11	14 days	[52][f]	[65][f]	6[f]	7[f]
	25 [HIV –ve adults		14		[77][f]	[91][f]	10[f]	13[f]
Baik, 2014	25 Adults	Korea	4	14 days	19 [95]	19 [95]	115	108
Baik, 2015	**Shanchol**	Philippines		14 days				
	376 Adults		74		295 [79]	278 [74]	17	13
	235 [1-17 years]		13		197 [84]	207 [88]	49	57
	Euvichol							
	377 Adults		77		322 [85]	302 [80]	22	16
	231 [1-17 years]		13		200 [87]	209 [90]	61	66
Saha, 2016	143 Adults[h]	Bangladesh	71	14 days	106 [74][f]	103 [72][f]	NA[b]	7[f]

Study	Number and age group of participants	Study site	Baseline geometric mean titres	Interval between doses	No [%] that seroconverted after:		GMF rise after:	
					1st dose[a]	2nd dose[a]	1st dose[a]	2nd dose[a]
Iyer, 2016	37 1-5 years[i]	South Sudan	15	~21 days	9 [82]	11 [79]	11	14
	67 6-17 years [i]		28		8 [53]	18 [55]	4	4
	101 18-59 years [i]		36		20 [43]	28 [52]	7	5
Matias, 2016	22 adults	Haiti	35	14 days	14 [64]	16 [76]	7	5

[a] Blood for vibriocidal tests were obtained at 14 days after said dose, unless otherwise specified.
[b] Not available
[c] Blood for vibriocidal tests were obtained at 3 days after said dose
[d] Result of vibriocidal tests on 30th day after the single dose: 44%
[e] Blood for vibriocidal tests were obtained on 16th day after the said dose or 30 days after the first dose
[f] Blood for vibriocidal tests were obtained on 7th day after said dose, i.e. 7 or 21 days after the first dose
[g] These individuals previously received vaccine, 5 years earlier.
[h] Results included are only for those who received vaccine at the current storage recommendation of 2-8°C
[i]Not all individuals were sampled at both time points to denominators for seroconversion changes.

Table 2: Vibriocidal immune responses to Inaba serotype following killed oral cholera vaccination (see references listed in Supplementary data, pages 4-5)

Study	Number of participants [age group]	Study site	Baseline geometric mean titres	Interval between doses	No [%] that seroconverted after:		GMF rise after:	
					1st dose[a]	2nd dose[a]	1st dose[a]	2nd dose[a]
Bivalent OCV								
Kanungo, 2009	37 [18+ years]	India	186	14 days	24 [65]	17 [46]	9	5
	39 [1-17 years]		37	14 days	34 [87]	32 [82]	47	24
Saha, 2011	53 [18-45 years]	Bangladesh	55	14 days	[60][f]	[57][c]	9[c]	NA[b]
	55 [2-4 years]		55		[78][f]	[76][c]	12[c]	NA[b]
	54 [12-24 months]		8		[52][f]	[72][c]	7[c]	NA[b]
Charles, 2014	23 Adults	Haiti	11	14 days	[77][f]	[91][c]	19[c]	19[c]
	45 [6 to 17 years]		27		[69][f]	[74][c]	11[c]	10[c]
	42 [1-4 years]		16		[64][f]	[73][c]	9[c]	10[c]
Kanungo, 2015	96 [15+ years]	India	171	14 days	67 [70]	58 [60]	7	5
	90 [6-14 years]		50	14 days	[88]	[79]	26	14
	91 [15+ years]		238	14 days	[57]	[51]	5	4
	93 [6-14 years]		81	14 days	[85]	[82]	33	16
Kanungo, 2015	86 Adults [≥18 years]	India	191	14 days	59 [69]	47 [55]	7	5
			144	28 days	55 [66]	49 [58]	9	5
	84 Children [1-17 years]		47	14 days	72 [86]	67 [80]	30	18
			89	28 days	73 [89]	63 [77]	21	11
Aloysia, 2015	112 [15+ years]	Philippines	36	14 days	[83]	[78]	25	18
	112 [5-14 years]		3		[88]	[87]	58	49
	112 [1-4 years]		1		[88]	[89]	67	67

Study	Number of participants [age group]	Study site	Baseline geometric mean titres	Interval between doses	No [%] that seroconverted after:		GMF rise after:	
					1st dose[a]	2nd dose[a]	1st dose[a]	2nd dose[a]
Desai, 2015	54 [18+ years]	Ethiopia	16	14 days	37 [70]	43 [81]	11	15
	53 [1-17 years]		6		39 [74]	41 [77]	13	13
Ivers, 2015	25 HIV +ve adults	Haiti	11	14 days	[65][f]	[74] [c]	7 [c]	7 [c]
	25 HIV –ve adults		11		[82] [f]	[91] [c]	17 [c]	20[c]
Baik, 2014	20 Adults	Korea	4	14 days	18 [90]	19 [95]	74	94
Baik, 2015	**Shanchol**	Philippines		14 days				
	376 Adults		36		315 [84]	287 [76]	30	21
	235 [1-17 years]		12		198 [84]	209 [89]	51	52
	Euvichol							
	366 Adults		36		317[84]	308 [82]	32	22
	236 1-17 years		12		198 [86]	202 [87]	55	51
Saha, 2016	143 Adults[d]	Bangladesh	99	14 days	109 [76][c]	105 [73][c]	11[c]	9[c]
Iyer, 2016	37 1-5 years	South Sudan	11	~21 days	9 [75]	12 [80]	11	9
	67 6-17 years		30		8 [53]	12 [38]	2	3
	101 adults [18-59 years]		22		28 [61]	31 [57]	8	7
Matias, 2016	22 adults	Haiti	29	14 days	16 [73][c]	17 [81][c]	9[c]	9[c]

[a] Blood for vibriocidal tests were obtained at 14 days after said dose, unless otherwise specified.
[b] Not available
[c] Blood for vibriocidal tests were obtained on 7th day after said dose, i.e. 7 and 21 days after the first dose.
[d] Results included are only for those who received vaccine at the current storage recommendation of 2-8°C

Table 3: Estimated protection conferred by a single dose of killed oral cholera vaccine (see references listed in Supplementary data, pages 4-5)

Study	Site [study year] study design	Vaccine	Study population and number of participants	Primary assessment of protection [clinical endpoints and definitions]	Total number of cholera cases and serotypes and biotypes	Vaccine protection after 1 dose [95% confidence interval]	Duration of follow-up [months]
Wierzba, 2015	Odisha, India [2011] Test-negative design	Bivalent OCV [Shanchol]	Of 51,488 eligible residents of the study area, 31,552 [61%] received at least one dose and 23,751 [46%] received two doses	Compare odds of having been vaccinated between cholera cases and test-negative controls • Cases were diarrhoea patients found positive for *V. cholerae* • Controls were diarrhoea patients negative for *V. cholerae* infection	44 patients included in the analysis • 44 [100%] O1 Ogawa, of which 34 [77%] were El Tor Variant and 10 [23%] were Hybrid [El Tor / Classical] biotypes	• 33% [−318 to 89]	24
Ivers, 2015	Haiti [2012] Case-control design with bias-indicator study	Bivalent OCV [Shanchol]	45,417 people were vaccinated in the campaign, 91% of whom received both doses	Compare odds of having been vaccinated between cholera cases and matched controls • Cases were diarrhoea patients with a stool sample positive for *V cholerae* O1 • Controls were individuals who did not seek treatment for diarrhoea between the first day of	48 patients [one excluded from the analysis due to lost data] • 36 [37%] O1 Ogawa • 11 [23%] O1 Inaba	• 67% [–62 to 93]	23

Study	Site [study year] study design	Vaccine	Study population and number of participants	Primary assessment of protection [clinical endpoints and definitions]	Total number of cholera cases and serotypes and biotypes	Vaccine protection after 1 dose [95% confidence interval]	Duration of follow-up [months]
				study enrolment and the date of onset of symptoms in their corresponding case, matched to each case by location of residence, enrolment time [within 2 weeks of the case], and age group [1–4 years, 5–15 years, and >15 years].			
Khatib, 2012	Zanzibar [2008] Cohort design with bias indicator study	Monovalent OCV containing recombinant B-subunit [Dukoral]	Of 48,178 eligible residents of the study area, 23,921 [50%] received two complete doses of vaccine	Compare incidence of cholera in recipients the vaccine and non-recipients	42 patients included in the primary analysis • 42 [100%] O1 El Tor Ogawa	• 46% [–80 to 83]	14
Luquero, 2014	Boffa and Forecariah, Guinea [2012] Case-control design with bias-indicator study	Bivalent OCV [Shanchol]	Target population was163,000 people in Boffa district, and 46,000 people in parts of Forecariah. Coverage with at least one dose was 92% in Boffa and 71% in Forecariah, [40]	Compare odds of having been vaccinated between cholera cases and matched controls • Cases were diarrhoea patients with a stool sample positive for *V cholerae* O1 • Controls were neighbours of the case who did not seek treatment for diarrhoea between the first day of study enrolment and the date of onset of symptoms in their	40 patients included in the primary analysis. Of the 36 for whom a specimen was sent for culture and PCR analysis: • 18 [50%] O1 El Tor Ogawa; • 13 had positive results of	• 66% [−53 to 93] - based on culture or PCR positive cholera • 43% [−84 to 82] - based on RDT positive cholera	6

Study	Site [study year] study design	Vaccine	Study population and number of participants	Primary assessment of protection [clinical endpoints and definitions]	Total number of cholera cases and serotypes and biotypes	Vaccine protection after 1 dose [95% confidence interval]	Duration of follow-up [months]
				corresponding case, matched to each case by age group [[1 to 4, 5 to 9, 10 to 19, 20 to 29, 30 to 39, or 40 years of age or older].	culture and PCR • 5 had positive PCR results but negative culture results.		
Qadri, 2016	Bangladesh [2013] Phase III randomised control trial	Bivalent OCV [Shanchol]	204,700 persons underwent randomization, received one dose, and were included in the analysis [102,552 received vaccine and 102,148 received placebo]	Compare incidence of cholera in randomly assigned recipients the vaccine and placebo.	101 cholera cases included in the analysis • 100 [99%] O1 El Tor Ogawa • 1 [1%] O1 El Tor Inaba	• 40% [11 to 60] against all cholera episodes • 63% [24-82] against severely dehydrating cholera • 56% [16 to 77], 63% [−39 to 90], and 16% [49 to 53] against all cholera episode among persons vaccinated at the age of 15 or more years, 5 to 14 years and 1 to 4 years, respectively	6
Azman, 2016	Juba, South Sudan [2015] Case-cohort study	Bivalent OCV [Shanchol]	Juba was estimated to have between 500 000 and 1 million inhabitants, with massive population movements because of civil strife. 140,249 doses were administered in targeted areas of Juba. [11]	Compare hazard ratios of cholera between unvaccinated and vaccinated persons.	34 cholera cases included in the analysis	• 87% [70 to 100]	2

Figure legends

Figure 1: Selection of reports included in the analysis

Figure 2: Seroconversion after the first and second dose of a bivalent oral cholera vaccine, all age groups

Figure 3 - A, B, and C: Seroconversion to the Ogawa serotype after the first and second dose of a bivalent oral cholera vaccine in young children, children, and adults

Figure 4: Estimated protection [95% confidence intervals] conferred by a single dose of killed oral cholera vaccine, by study site and duration

Figure 1.

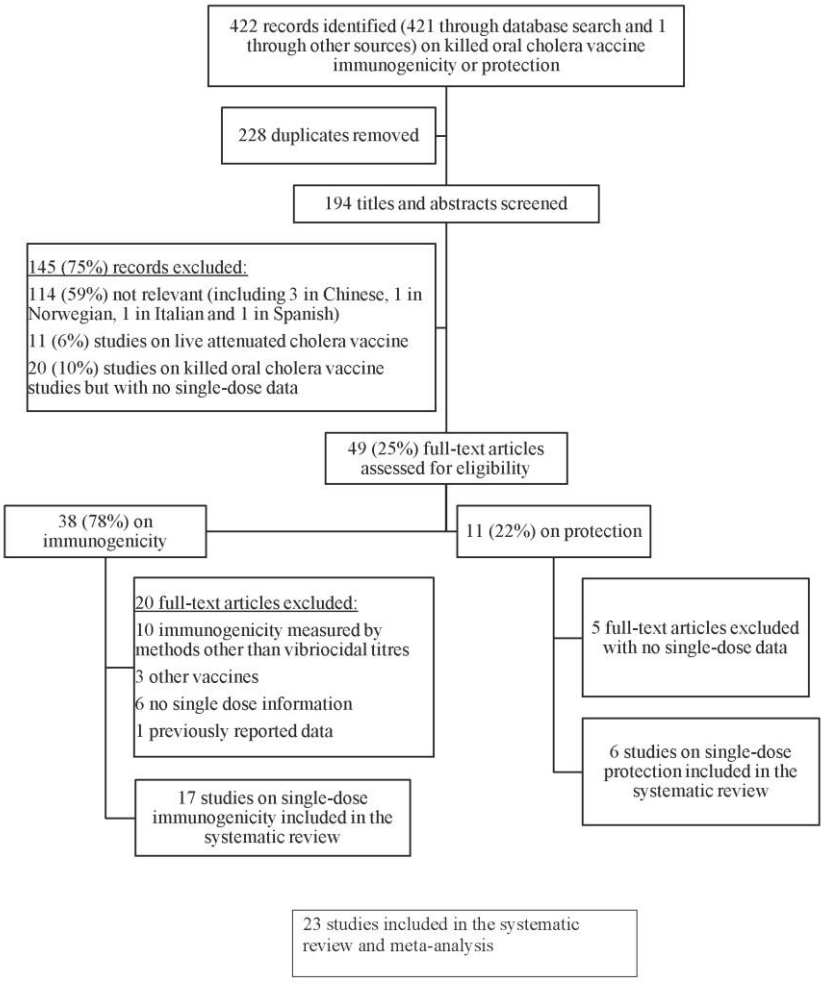

Figure 2.

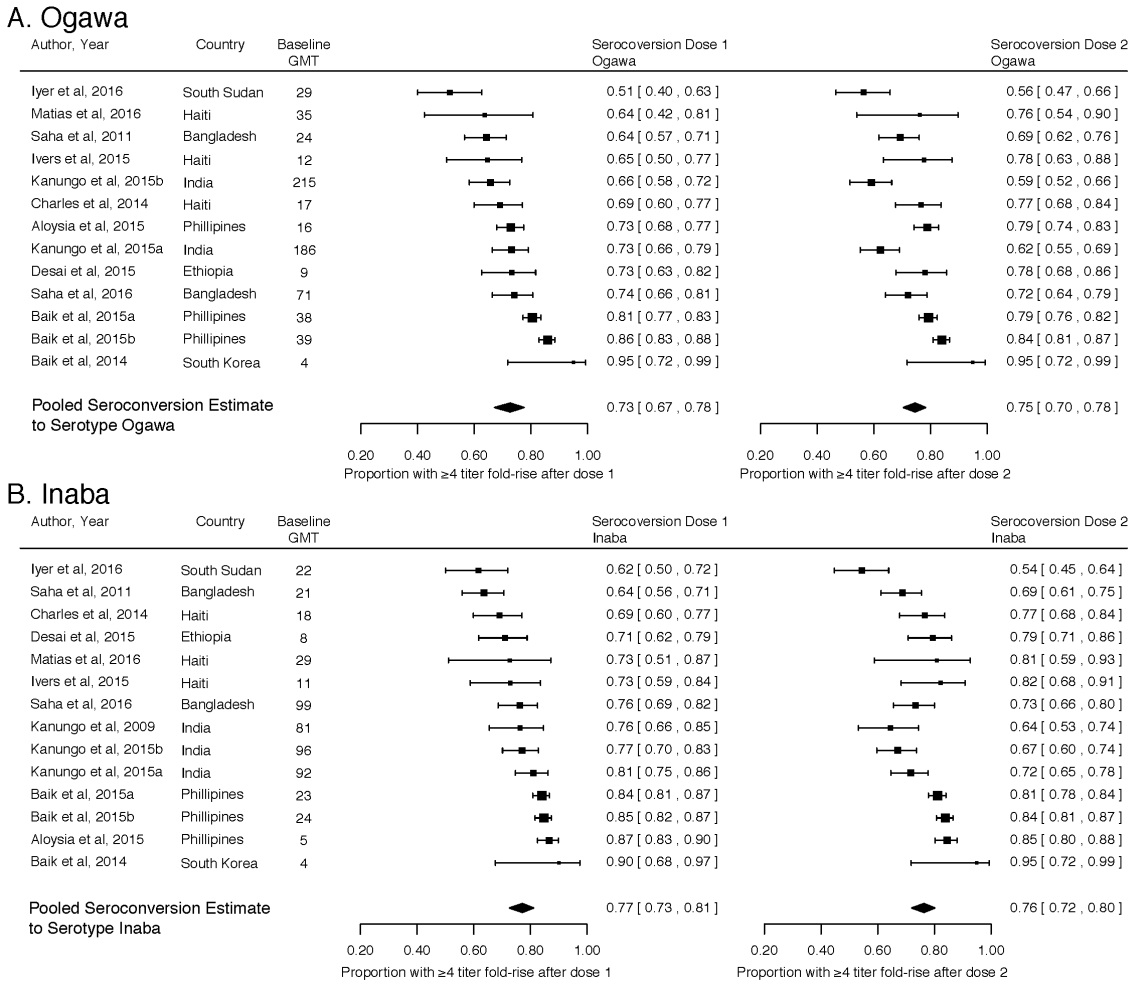

Figure 3.

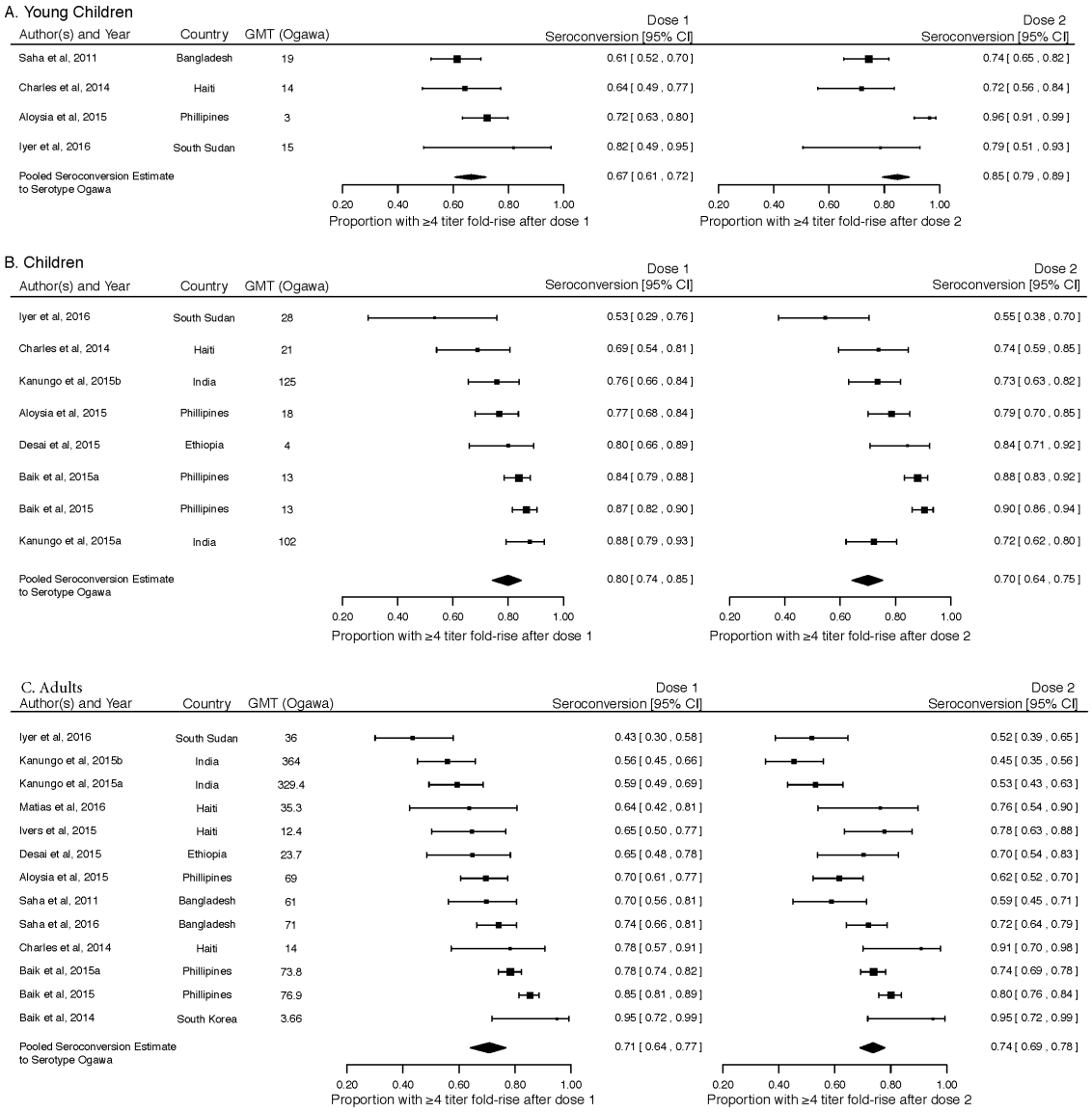

Figure 4.