Acta Tropica xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Acta Tropica

journal homepage: www.elsevier.com/locate/actatropica

Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control

M. Ducrotoy[a], W.J. Bertu[b], G. Matope[c], S. Cadmus[d], R. Conde-Álvarez[e], A.M. Gusi[b], S. Welburn[a], R. Ocholi[b], J.M. Blasco[f], I. Moriyón[e,*]

[a] Division of Infection and Pathway Medicine, Centre for Infectious Diseases, School of Biomedical Sciences, College of Medicine and Veterinary Medicine, The University of Edinburgh, Chancellor's Building, 49 Little France Crescent, Edinburgh EH16 4SB, UK
[b] Brucellosis Research Unit, National Veterinary Research Institute Vom 930001 Plateau State, Nigeria
[c] Department of Paraclinical Veterinary Studies, Faculty of Veterinary Science, University of Zimbabwe, P.O. Box MP 167, Mount Pleasant, Harare, Zimbabwe
[d] Department of Veterinary Public Health & Preventive Medicine, University of Ibadan, Ibadan, Nigeria
[e] Instituto de Salud Tropical y Depto. Microbiología y Parasitología, Universidad de Navarra, Edificio de Investigación, c/Irunlarrea 1, 31008 Pamplona, Spain
[f] CITA—Universidad de Zaragoza—Unidad de Sanidad Animal, Avenida de Montañana 930, 50059 Zaragoza, Spain

ARTICLE INFO

Article history:
Received 12 April 2015
Received in revised form
14 September 2015
Accepted 29 October 2015
Available online xxx

Keywords:
Brucellosis
Sub-Sahara
Africa
Diagnosis
Vaccination
Control

ABSTRACT

Brucellosis is a highly contagious zoonosis caused by bacteria of the genus *Brucella* and affecting domestic and wild mammals. In this paper, the bacteriological and serological evidence of brucellosis in Sub-Saharan Africa (SSA) and its epidemiological characteristics are discussed. The tools available for the diagnosis and treatment of human brucellosis and for the diagnosis and control of animal brucellosis and their applicability in the context of SSA are presented and gaps identified. These gaps concern mostly the need for simpler and more affordable antimicrobial treatments against human brucellosis, the development of a *B. melitensis* vaccine that could circumvent the drawbacks of the currently available Rev 1 vaccine, and the investigation of serological diagnostic tests for camel brucellosis and wildlife. Strategies for the implementation of animal vaccination are also discussed.

1. Introduction

Brucellosis is the collective name given to a group of zoonoses caused by gram-negative bacteria of the genus *Brucella*. The disease has a worldwide distribution and affects economically important domestic livestock as well as a wide range of wild mammals (Godfroid et al., 2011; Zheludkov and Tsirelson, 2010). *Brucella abortus*, *B. melitensis* and *B. suis* are the species that have the highest impact on domestic livestock productivity and human health (Godfroid et al., 2011) and, although they preferentially infect cattle, small ruminants and swine, respectively, cross-infections may be significant in mixed husbandry systems or at the livestock-wildlife interface (Godfroid et al., 2013; Verger et al., 1989; Zheludkov and Tsirelson, 2010). Eradicated from cattle and small ruminants in a handful of industrialized countries, brucellosis remains endemic in most areas of the world (Moreno, 2014).

Brucellosis is a "difficult disease" (Cunningham, 1977). In addition to the wide range of hosts (and subsequent multifaceted epidemiology) and the socioeconomic implications, brucellosis is not readily identified because of its variable picture at both individual and population level. The main clinical signs in bovines and small ruminants are abortions and infertility, but they are neither disease-specific nor present in all infected individuals (Cunningham, 1977). Since abortion usually occurs in the first pregnancy after infection and becomes less likely thereafter because of sustained immunity, the introduction of infected animals into immunologically naïve groups (or of unprotected replacements in infected groups) results in multiple abortions in a short period of time ("abortion storms") (Cunningham, 1977). Whereas exposure to the billions of bacteria released (Corner, 1983) can be controlled to some extent by proper animal management, congenital transmission and the consequent existence of initially asymptomatic and seronegative animals that subsequently become contagious pose a difficult challenge (Catlin and Sheehan, 1986; Plommet, 1977; Ray et al., 1988). When the "acute" phase has passed, individual prevalence stabilizes or even decreases because

* Corresponding author. Fax: +34948425649.
E-mail address: imoriyon@unav.es (I. Moriyón).

http://dx.doi.org/10.1016/j.actatropica.2015.10.023

of the development of herd immunity and a reduced exposure to heavy bacterial challenges, and clinical signs become much less discernible. However, the disease becomes "chronic" in the herd, flock or farm. This "acute"/"chronic" dynamic has been recognized for a long time (Cunningham, 1977) and, as expected, there are fluctuations between these extreme situations caused by management practices such as intensification, sedentarization and the introduction of unprotected exotic breeds (Akakpo, 1987; Akakpo and Bornarel, 1987; Alton, 1981; Roux, 1979). Accordingly, whereas the overall individual prevalence and abortion rates in an endemic area are often moderate, the proportion of infected herds, flocks or farms usually remains high and is the true indicator of the potential of the disease to flare up when conditions promoting transmission occur.

Human brucellosis is a debilitating disease that lacks pathognomonic symptoms (Ariza, 1999; Dalrymple-Champneys, 1960; Spink, 1956), which makes it difficult to distinguish from other febrile conditions, including malaria (Chabasse et al., 1983; El Ansary et al., 2001; Maichomo et al., 1998; Mangen et al., 2002; Mert et al., 2003; Muriuki et al., 1997; Mutanda, 1998). *B. melitensis* is the cause of most of the reported cases of human brucellosis and causes the most severe form of the disease, followed by *B. suis* and *B. abortus*; much less frequent are infections by *B. canis*, a species that is restricted to dogs (Ariza, 1999; Moreno, 2014; Spink, 1956). Since there is no human vaccine and no significant human-to-human transmission, control of animal brucellosis, milk pasteurization and other food hygiene measures are the only options to reduce its occurrence in humans.

Despite underreporting and the scarcity of epidemiologically valid data, the evidence obtained throughout the years shows that brucellosis is a widespread problem in Africa (Akakpo, 1987; Ducrotoy et al., 2014; Grace et al., 2012; Mangen et al., 2002; McDermott et al., 2013; Thimm and Wundt, 1976) a continent where several Sub Saharan countries are estimated to bear a high burden of neglected zoonotic diseases (Grace et al., 2012).

The purpose of this article is twofold. First, we discuss the characteristics of brucellosis in Sub Saharan Africa (SSA), that is, the *Brucella* species involved and epidemiological peculiarities, and the variability of the clinical picture at herd and flock level, and the reasons for this diversity. To this end, we present bacteriological evidence available since *Brucella* was first isolated in SSA, the experience gathered during the course of the European Union funded ICONZ project (http://www.iconzafrica.org), and data from solid sero-epidemiological studies (Sections 2–5). An exhaustive review of the serological evidence including all "grey literature" (Ducrotoy et al., 2014) is out of the scope of this work, and we have used the literature available in PubMed since 2001 (see Supporting Material) to update the evidence presented in the authoritative review of McDermott and Arami (McDermott and Arimi 2002). The second objective of the present work is to discuss the suitability of diagnostic tools, treatments, vaccines and control strategies for the region (Section 6), as well as critical aspects of brucellosis management that have not been discussed previously in the context of SSA.

2. *Brucella* potential hosts and livestock production systems in Africa

McDermott and Arimi (McDermott and Arimi, 2002) have presented in detail the estimated distribution of livestock and productions systems in SSA and here we will only summarize those characteristics that are relevant for the discussions below (Fig. 1A and B). But for the Nile delta and a few thin coastal strips, cattle are mostly reared in the Sahel and Ethiopian highlands and in Kenya, Tanzania, Uganda and other Eastern African countries of the Great Lakes area and in Southern African countries, namely Bostwana, Madagascar, South Africa, South West of Angola, Zambia and Zimbabwe. Small ruminants are found in higher densities than cattle and, although overlapping largely with the latter, penetrate more deeply into semi-arid areas bordering the South and North of Sahara and the horn of Africa. Livestock production systems are shown in Fig. 1C grouped into five broad categories. In SSA, grazing is the predominant production system but most cattle are distributed on mixed rain-fed systems. There are few (mixed) irrigated areas that can support high stocking densities. The broad picture is characterized by a scarcity of intensive livestock production systems and a dominance of extensive pastoralist or agro-pastoralist systems where cattle and small ruminants are co-reared. Camels in the northern and southern borders of Sahara and Ethiopia add another dimension to these mixed breeding systems. In addition, transhumance is practiced according to seasonal variations and grazing availability in many areas.

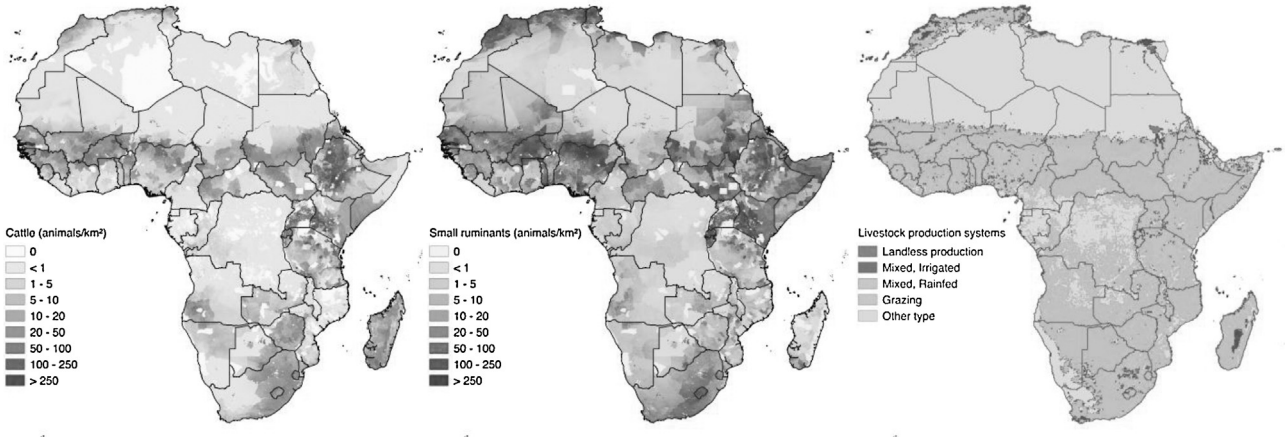

Fig. 1. *Brucella* potential hosts and livestock production systems in Africa.
Livestock density is expressed as the number of cattle (panel A) or small ruminants (B) per square kilometer according to categories of different sizes in order to clearly differentiate between zones of low and high livestock density (data derived from Gridded Livestock of the World; http://livestock.geo-wiki.org). Panel C shows the livestock production systems. Landless production systems are characterized by (i) less than 10% of the income that comes from non-livestock agriculture, (ii) less than 10% of the feed dry matter that is farm produced, (iii) stocking rates of more than 10 livestock units per ha of agriculture land and (iv) a higher ruminant enterprise value than that of pig or poultry. In mixed farming systems the non-livestock agriculture is responsible for a considerable part of the income. A distinction is made between rain-fed crops and land that is irrigated. In the remaining non-mixed farming systems, livestock depends on grazing activities whether extensively mobile as pastoralists or not. Land inaccessible or unsuitable for livestock is mainly categorized under "Other type"

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

3. Brucellosis in domestic animals in SSA

As the clinical signs of this disease are not specific, brucellosis cannot be diagnosed clinically. Hygromas seem a good indirect indicator of brucellosis in African herds (Akakpo, 1987; Ferney and Chantal, 1976; McDermott and Arimi, 2002) probably because infected animals are kept long enough for this arthropathy to develop. Although in African herds hygromas correlate more closely than abortions with a positive brucellosis serology (Akakpo and Bornarel, 1987) and possibly represent one of samples of choice for bacteriological studies (Sanogo et al., 2013), they are not pathognomonic and evidence has to be based on laboratory tests. Of these, only serological tests and bacteriological isolation are presently valid because, despite their high analytical sensitivity under laboratory conditions, the diagnostic sensitivity and specificity of PCR, RT-PCR and other DNA detection tests is undetermined (Yu and Nielsen, 2010).

3.1. Brucella *species*

It is a well-established fact that the dominance and overlapping nature of the C epitope of smooth brucellae (Alonso-Urmeneta et al., 1998) makes it impossible to ascertain the infecting *Brucella* species using serological tests, irrespective of the antigen (*melitensis* or *abortus*) or host species tested (Ariza, 1999; OIE, 2013a,b; Spink, 1956). Thus, bacteriological isolation is strictly necessary to determine the infecting *Brucella* species and to understand the epidemiology when different host species are managed together or share grazing grounds and water sources. However, identification and typing of *Brucella* species by conventional procedures is difficult and molecular methods are preferred for typing strains once these are isolated (OIE, 2013a). This is exemplified by the case of the *B. abortus* biovar 3a strains of Nigeria (Bertu et al., 2015), originally reported as biovar 1 and thought to be an exception in the area [(Sanogo et al., 2013); see also below]. Moreover, these methods can provide insights into the origin of the strains that are not evident from the results of classical phenotyping.

Despite the inconsistencies of reports, the presence of *B. abortus* and *B. melitensis* in African countries of the Mediterranean coast is well known, and both classical biotyping and molecular studies show that they are closely related to other strains in the Mediterranean basin (Lounes et al., 2014; Verger and Grayon, 1984). On the other hand, the number of *B. melitensis* isolates from SSA is very limited (Table 1). Moreover, their identification as a classical "*melitensis*" spp. is not always clear. This is the case of the strains isolated from goats in Nigeria, since their biochemical characteristics are atypical (Falade, 1981a) or their reported rough state precludes clear assignment (Bale et al., 2003). Most isolates have been obtained in Kenya (Table 1). Biovars 1 and 3 were reported in sheep and goats over 40 years ago, in one instance in an outbreak associated with male goats imported from Israel and, interestingly, a more recent isolate of *B. melitensis* from cattle that is closely related to Israel strains has been reported. These data could suggest a Mediterranean origin of at least some the *B. melitensis* strains and their subsequent transmission of *B. melitensis* to cattle, as observed in countries where cattle and small ruminants are kept together (Benkirane, 2006; Refai, 2002). Some biovar 1 strains were also isolated from goats in Natal and Zimbabwe (Table 1). In humans, *B. melitensis* was isolated with frequency higher than *B. abortus* in Kenya (Table 1), which conforms to the greater virulence of the former species in humans, and in a case imported from Somalia (Table 1).

Brucella abortus encompasses the largest number of isolates, presumably because cattle have been sampled more often than small ruminants. For West Africa, almost 90% of the isolates to date were reported over 30 years ago (Table 1). Remarkably, this early work found only *B. abortus* biovar 3 in autochthonous cattle in Senegal, Togo, Rwanda, Guinea Bissau and Niger, and identified significant differences between these isolates and biovar 3 of European strains in oxidase and metabolic oxidative tests (Akakpo, 1987; Verger and Grayon, 1984). Consistent with this, the recent molecular analyses (Table 1) show that strains isolated thus far in SSA represent a genotype (*B. abortus* biovar 3a; Table 1) different from that (*B. abortus* biovar 3b) isolated in Europe and Latin America (Bertu et al., 2015). Very recently, a *B. abortus* strain isolated in Tanzania did not clearly group with 3a, and 11 *B. abortus* strains isolated in Uganda that were found to be monomorphic at molecular analysis may belong to *B. abortus* biovar 7 (Table 1). Biovar 7, although suspended from *Brucella* nomenclature in 1988, has been reintroduced again upon re-examination by molecular methods (Garin-Bastuji et al., 2014), and the original reference strain was of African origin (Meyer and Morgan, 1973). It is noteworthy that *B. abortus* has been isolated several times from milk and abortion products of sheep in Nigeria and also from a goat, and the isolates that have been typed belong to the biovar 3a found in cattle (Table 1). Similarly, the *B. abortus* strains isolated from horses in Nigeria (Table 1) were of the same genotype found in cattle. Finally, although little is known about the zoonotic potential of biovar 3a (not all *Brucella* biovars are highly virulent for humans), the *B. abortus* biovar 3 Tulya reference strain which we know now belongs to biovar 3a (Ocampo-Sosa et al., 2005) was originally isolated from a human case in Uganda (Meyer and Morgan, 1973) and shows characteristics consistent with earlier cattle and human isolates from Zimbabwe (Table 1). Also, the *B. abortus* strain isolated from a human patient in Kenya almost 40 years ago (Table 1) was considered "atypical". *B. suis* biovar 1 has been clearly identified but only in cattle in Zimbawe (Table 1), which suggests its presence in pigs. The only evidence of *B. canis* is limited to two strains isolated in South Africa almost ten years ago (Gous et al., 2005). Bacteriological evidence of *B. ovis* (a non zoonotic species restricted to sheep) exists for several SSA countries (Ate et al., 2011; Cameron et al., 1971; De Wet and Erasmus, 1984; Van Rensburg et al., 1958).

As stressed before (Sanogo et al., 2013), there is a clear need for further bacteriological studies in cattle and small ruminants, particularly with regards to the human disease, supported by molecular typing. Despite this, the above-summarized evidence suggests the existence of a typically SSA *B. abortus* biovar 3a lineage as well as other genetic variants not clearly represented elsewhere. The data also suggest epidemiological situations in which infections of small ruminants by *B. abortus* occur in areas where they are in contact with cattle and *B. melitensis* is absent (co-infections by two different brucellae are rather unlikely because of the development of immunity in an ongoing infection and, in fact, they have never been convincingly proven). It would therefore be important to study to what extent such epidemiological situations are common. In other parts of the world, *B. abortus* has seldom been isolated from sheep, almost always from aborted ewes that had been in contact with infected cattle in *B. melitensis*-free countries (Allsup, 1969; Shaw, 1976). However, it is not known whether this very limited occurrence reflects different patterns of management of cattle and small ruminants or a host preference rooted in the biology of *B. abortus*, or both. Significantly, in one case this *Brucella* species was found to persist in sheep in the absence of a cattle reservoir (Luchsinger and Anderson, 1979) and indirect evidence suggests that *B. abortus* infected sheep may act as reservoirs for cattle brucellosis (Allsup, 1969). Concerning the infection of horses by *B. abortus*, previous evidence shows that it is a spill over disease from cattle and that horses do not act as a reservoir (Cohen et al., 1992).

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

Table 1
Characteristics of the *B. abortus* and *B. melitensis* strains isolated in SSA.

Country	Species	Biovar typing		N° isolates	Host	Period	Reference
		Conventional	Molecular[a]				
Gambia	*B. abortus*	3	3a Lineage	3	Cattle	Not reported	(Bankole et al., 2010)
Guinea Bissau	*B. abortus*	6-Mar	n.d.[b]	7	Cattle	1976–1982	(Verger and Grayon, 1984)[c]
Ivory Coast	*B. abortus*	3	3a Lineage	1	Cattle	2009	(Boukary et al., 2013)
Kenya	*B. melitensis*	1 and 3	n.d.	19	Goats	Not reported	(Philpott and Auko, 1972)
		2	n.d.	1	Sheep		
		1 and 3	n.d.	11	Human	1969–1972	(Oomen, 1976)
		1		1	Cattle	Not reported	(Muendo et al., 2012)
	B. abortus	Ätypical¨	n.d.	1	Human	1969–1972	(Oomen, 1976)
		3	3a Lineage	2	Cattle	Not reported	(Muendo et al., 2012)
Mali	*B. abortus*	1	n.d.	4	Cattle	1991	(Tounkara et al., 1994)
Natal	*B. melitensis*	1	n.d.	7	Goat	1994	(Reichel, 1996)[g]
Niger	*B. abortus*	3 or 3/6	n.d	2	Cattle	1976–1982	(Verger and Grayon, 1984)[c]
		3	3a Lineage	1		2007–2008	(Boukary et al., 2013)
Nigeria	*B. abortus*	1, 2	n.d	13	Cattle	1974–1976	(Eze, 1978)
		1, 3, 4	n.d.	11		Not reported	(Bale and Kumi-Diaka, 1981)
		3	3a Lineage	30		1976–2012	(Bertu et al., 2015)[d]
		3	3a Lineage	2	Sheep	1976–2012	(Bertu et al., 2015)[d]
				5		1977	(Okoh, 1980)
		3	3a Lineage	2	Horses	2004	(Bertu et al., 2015)[d]
		1	n.d	8	Goats	Late 1970s	(Falade, 1981a,b)
	B. melitensis	1	n.d.	23	Goats	Late 1970s	
		n.d.	n.d.	6		Not reported	(Bale et al., 2003)
				4	Sheep		
Rwanda	*B. abortus*	3 or 3/6	n.d	10	Cattle	1982–83	(Verger and Grayon, 1984)[c]
Senegal	*B. abortus*	n.d	n.d	6[e]	Cattle	1960-1962	(Chambron, 1965)
		n.d	n.d	14		1976	(Doutre et al., 1977)
		3 (one 1)	n.d	213		1976–1982	(Verger and Grayon, 1984)[c]
Somalia	*B. melitensis*	n.d	n.d	1	Human	1975	(Wheat et al., 1995)
Tanzania	*B. abortus*	3	? (Not 3a)		Cattle	2012–2013	(Mathew et al., 2015)
Togo	*B. abortus*	3 or 3/6	n.d	30	Cattle	1976–1982	(Verger and Grayon, 1984)[c]
		3 or 3/6	3a Lineage	3		2011–2012	(Dean et al., 2014)
Uganda	*B. abortus*	1, 3, 7 (?)	? (Not 3a)	11	Cattle	2011–2012	(Mugizi et al., 2015a,b)
	B. melitensis	n.d.	n.d	1		2011–2012	Mugizi 2015 (unpublished)
Zimbawe	*B. abortus*	3	n.d	≥4	Human	1921–1930	(Bevan, 1930)[f]
		n.d.	1	1	Cattle	Not reported	(Ledwaba et al. 2014)
	B. suis	n.d.	1	2	Cattle	Not reported	
	B. melitensis	1	n.d	1	Goat	1987	(Madsen, 1989)

[a] Molecular typing, where reported, was done according to (Le Fleche et al., 2006) and is simplified here as 3a lineage because of the greater relatedness to the Tulya strain of biovar 3 [see (Ocampo-Sosa et al., 2005); however, the analyses show internal diversity [see {(Bertu et al., 2015)]. The study by Ledwaba et al. (2014) was done by whole genome sequencing.
[b] n.d., Not done.
[c] This study includes the strains reported by Akapko (Akakpo, 1987) and Akapko and Bornarel (Akakpo and Bornarel, 1987), all of which were sent to the reference laboratory of M. Verger at INRA for a definite typing by oxidative metabolic tests.
[d] This study includes most strains previously reported by Ocholi et al. (Ocholi et al., 2004a,b, 2005) all of which were originally typed as *B. abortus* biovar 1.
[e] One of the strains isolated from cattle in his study showed features partially consistent with *B. melitensis*.
[f] A strain of this set of isolates was denominated as the "Rodesian abortus" (Stableforth and Jones, 1963) and belongs to biovar 3, possibly 3a.
[g] Same strains as those reported by (Emslie and Nel, 2002).

3.2. Serological evidence

Serological evidence of brucellosis is abundant throughout SSA. However, it is scattered in time and space and, in addition, good quality data that can be interpreted in strict epidemiological terms are rather scarce (Ducrotoy et al., 2014; Mangen et al., 2002; McDermott and Arimi, 2002). Over a decade ago, McDermott and Arimi (McDermott and Arimi, 2002) noted that the figures reported had to be interpreted with caution because of uncertainties in test implementation and validation. Tables 2 and 3 summarize serological surveys accessible in PubMed that meet strict scientific criteria (see Supporting Material) published since 2001 and they show that the challenges and uncertainties related to the serological tests remain (see also Section 6.2.1). Despite these diagnostic uncertainties, a number of studies in Africa show that individual brucellosis seroprevalence correlates with the number of abortions (Akakpo, 1987; McDermott and Arimi, 2002; Megersa et al., 2011a; Muma et al., 2012), as expected (see Section 1).

It is a matter of debate whether brucellosis (sero) prevalence is higher under extensive or intensive breeding conditions (Ducrotoy et al., 2014). Concerning individual seroprevalence, McDermott and Arimi (McDermott and Arimi, 2002) summarized data prior to 2001 in cattle as varying from 7.5 to 40% for pastoralists in arid and semiarid areas, 0.3–25.4% for cash/subsistence crops with livestock in sub-humid areas, 1.5–16.2% for crop-livestock in tropical highlands and 2.4–45.0% for crop with small-scale livestock production in humid areas. The wide range in values indicates that, based on early evidence, it is not possible to draw general conclusions on the significance of these production systems on the individual prevalence, as already observed by Mangen et al. (Mangen et al., 2002). This is confirmed by the studies published since 2001 (Table 2) and, indeed, the lack of a uniform trend conforms to the well-known variable clinical picture at individual and herd levels summarized in the Introduction. The same authors also commented that, although highly variable, the data suggested that individual prevalence was usually greater in systems in which large numbers of cattle mix and lowest for small confined herds. Indeed, herd size, movement and congregation of animals for access to pastures, water, or marketing figure among the well-known risk factors of brucellosis and have been identified consistently in previous and more recent stud-

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

Table 2
Seroprevalence studies in SSA cattle since 2001[a].

Country	Population/production system	% Seroprevalence (no.)		Diagnostic test (antigen)	Reference (comments)
		Individual	Herd		
Cameroon	Bos Indicus, (n.s.)	3.1 (1377)	15.9 (146)	cELISA (VLA)[b]	(Scolamacchia et al., 2010)
Chad	Nomadic pastoral	6.6[c] (608)	n.s.	RBT (Sanofi) and iELISA (CHEKIT)[d] in series	(Schelling et al., 2003)
Ethiopia	Pastoral	15 (68)	n.s.	RBT (n.s.)	(Molla and Delil, 2015)
	Pastoral	1.0 (1152)	4.9 (164)	RBT (n.s.) and CFT (n.s.) in series	(Adugna et al., 2013)
	Dairy small holders	1.7 (417)	n.s.	RBT (n.s.) and iELISA (Prionics)[e] in series	(Tschopp et al., 2013)
	Urban & peri-urban dairy, commercial, breeding	1.9 (2334)	10.6 (273)	RBT (Lilidale) and CFT (n.s.) in series	(Asmare et al., 2013a,b) (Herd prevalence 8.6, 16.9 and 20.0% in urban and peri-urban dairy, commercial and breeding farms, respectively)
	Pastoral	8 (575)	51.7 (58)	RBT (Pourquier) and CFT (VLA) in series	(Megersa et al., 2012)
	Dairy farms	1.5 (1202)	n.s.	RBT (n.s.) and CFT (n.s.) in series	(Tesfaye et al., 2011)
	Pastoral	10.6 (283)	n.s.	RBT (n.s.) and CFT (n.s.) in series	(Megersa et al., 2011a,b)
	Mixed crop-livestock system, small-holder dairy	4.9 (1238)	16 (176)	RBT (Pourquier)	(Jergefa et al., 2009)
		2.9 (1238)	13.6 (176)	CFT (VLA)	
	Semi-intensive	7.7 (1120)	63.6 (110)	RBT (Pouquier) and CFT (n.s.) in series	(Mekonnen et al., 2010)
	Extensive	1.2 (848)	3.3 (210)		
	Pastoral & mixed crop-livestock	3.86 (985)	22.9 (105)	RBT (Pourquier) and CFT (n.s.) in series	(Ibrahim et al., 2010)
	Urban semi-intensive	1.97 (610)	8.2 (122)		
Mozambique	Wildlife-livestock interface	9.77 (133)	n.s.	RBT (Onderstepoort)	(Tanner et al., 2014)
Niger	Urban	2 (973)	n.s.	iELISA (n.s.)[e]	(Boukary et al., 2013)
	Peri-urban	1.8 (1473)	n.s.		
	Rural pastoral	4.6 (724)	n.s.		
Nigeria	Pastoral, agropastoral, commercial, zero-grazing	36.6 (4745)	84.9 (271)	RBT (VLA)	(Mai et al., 2012) (Prevalence highest in pastoral system > zero-grazing >agro-past> commercial).
		24 (4745)	77.5 (271)	cELISA (VLA)[b]	
South Africa	Rural	1.45[c] (46025)	n.s.	RBT (n.s.) and CFT (n.s.) in series	(Hesterberg et al., 2008)
Togo	Rural	9.4 (596)	n.s.	RBT (Bio-Rad)	(Dean et al., 2013) (Samples positive to RBT but negative to ELISA were screened with CFT but unclear if this is taken into consideration for seroprevalence interpretation).
		5.7 (464)	n.s.	iELISA (IDEXX)[b]	
	Nomadic pastoral	7.3 (596)	n.s.	RBT (Bio-Rad)	
		4.5 (464)	n.s.	iELISA (IDEXX)[b]	
Uganda	Urban and peri-urban zero-grazing /mixed crop livestock/large scale dairy	5.9[c] (423)	6.5[c] (177)	cELISA (VLA)[b]	(Makita et al., 2011)
	Pastoral	34 (497)	100 (9)	cELISA (Svanova)[b]	(Magona et al., 2009)
	Zero-grazing	3.3 (226)	5.5 (146)		
	Pastoral	15.8 (5987)	78.6 (106)	RBT (Pourquier)	(Bernard et al., 2005)
	Agro-pastoral	12.8 (4542)	46.4 (209)		
Zambia	Commercial & small-scale mixed dairy-beef farms	14.3 (897)	n.s. (55)	RBT (Onderstepoort)	(Chimana et al., 2010)
		8.7 (897)	n.s. (55)	cELISA (Svanova)[b]	
	Wildlife-livestock interface	19[c] (1245)	63[c] (123)	RBT (VLA) and cELISA (Svanova)[b]	(Muma et al., 2007a,b)
Zimbabwe	Wildlife-livestock interface	9.9 (1158)	n.s.	RBT (VLA) and cELISA (Svanova)[b] in series	(Gomo et al., 2012)
	Small-holder dairy farms	5.6[c] (1440)	25.6[c] (203)	RBT (VLA) and cELISA (Svanova)[b]	(Matope et al., 2011)

[a] Abbreviation used in the Table, cELISA- competitive ELISA; iELISA- indirect ELISA; RBT, rose Bengal test; CFT-, complement fixation test; n.s., not specified; VLA, Veterinary Laboratory Agency.
[b] Manufacturer cut-off applied.
[c] Prevalence value adjusted according to sampling method +/− test sensitivity and specificity.
[d] Cut-off determined using ROC plot.
[e] Cut-off not specified.

ies in Africa (Akakpo, 1987; Berhe et al., 2007; Kadohira et al., 1997; Megersa et al., 2011a,b; Mekonnen et al., 2010; Muma et al., 2007a,b; Sanogo et al., 2012). However, although these factors co-exist under extensive breeding conditions, namely pastoralism and nomadism, these management systems are not necessarily associated with a higher individual prevalence. In two excellent reviews, Akapko and Bornarel (Akakpo, 1987; Akakpo and Bornarel, 1987) summarized work carried out in Burkina-Faso, Togo, and Rwanda showing that individual prevalence was higher in settled than in

Table 3
Seroprevalence studies in small ruminants in SSA since 2001[a].

Country	Population/production system	% Seroprevalence (no.)				Diagnostic test (antigen)	Reference (comments)
		Sheep		Goats			
		Individual	Flock	Individual	Herd		
Chad	Nomadic pastoral	0 (367)	NS	0 (374)	NS	RBT (Sanofi) and iELISA (CHEKIT)[b] in series	(Schelling et al., 2003)
Ethiopia	Pastoral	3 (77)	NS	0 (184)	NS	RBT (NS)	(Molla and Delil, 2015)
	Extensive mixed crop-livestock	1.4 (490)	NS	5.5 (495)	NS	mRBT (Lilidale) and CFT (NS) in series	(Teklue et al., 2013) (overall flock prevalence 28.3%)
	Sedentary, agro-pastoral and pastoral			1.9 (3315)	11.2 (448)	mRBT (NS) and CFT (NS) in series	(Asmare et al., 2013a,b)
	Pastoral			1.6 (1248)	13.3 (98)	RBT (Pourquier) and CFT (VLA) in series	(Megersa et al., 2012)
	Pastoral			1.9 (757)	NS	RBT (NS) and CFT (NS) in series	(Megersa et al., 2011a,b)
	Pastoral	4.8 (563)	NS			RBT (NS) and CFT (NS) in series	(Ashenafi et al., 2007)(seroprevalence pooled for sheep and goats)
Niger	Urban	3.6 (216)	NS	0.7 (106)	NS	iELISA (NS)[c]	(Boukary et al., 2013)
	Peri-urban	0.6 (320)	NS	0 (150)	NS		
	Rural pastoral	2.1 (650)	NS	0.4 (583)	NS		
Sudan	NS	2.15 (2005)	NS			RBT (NS)	(Gumaa et al., 2014) (one out of 400 samples mRBT negative found to be cELISA positive)
		3.4 (2005)	NS			mRBT (NS)	
Togo	Rural	0 (465)	NS	0 (221)	NS	mRBT (Bio-Rad) and iELISA (IDEXX)[d]	(Dean et al., 2013) (samples positive to RBT but negative to ELISA were screened with CFT but unclear if this is taken into consideration for seroprevalence interpretation).
	Wildlife-livestock interface	0 (280)	0 (29)			RBT (VLA) and cELISA (Svanova)[c]	(Muma et al., 2006) (pooled sheep and goat data)

[a] Abbreviation used in the Table, cELISA: competitive ELISA; iELISA: indirect ELISA; RBT: rose Bengal test; CFT: complement fixation test; NS: not specified, VLA: veterinary laboratory agency.
[b] Cut-off determined using ROC plot.
[c] Cut-off not specified.
[d] Manufacturer cut-off applied.

transhumant nomadic herds, and this is also observed in some recent studies.

In the course of the European Union funded ICONZ project (http://www.iconzafrica.org), a cross-sectional survey for brucellosis was carried out in the Kachia Grazing Reserve (KGR) of Kaduna (Nigeria). These reserves were established in Nigeria in an attempt to settle nomadic populations to avoid clashes with local farmers. However, KGR is actually a dynamic system where only about half of the population is settled, and even settled households practice seasonal transhumance. Cattle (n, ca. 2000; 40 households) and small ruminants (n, ca. 1500) were serologically examined using the rose Bengal test and, albeit in a limited number of cases, the presence of *B. abortus* (biovar 3a) was confirmed by bacteriological isolation. In cattle, the apparent seroprevalences were ca. 1.0% and 20% at individual and herd level respectively and brucellosis in small ruminants was hardly detected. This picture conforms to a situation of endemicity and moderate rates of transmission that, in addition to the intrinsic dynamics of the disease summarized in the Introduction may have several causes related to animal management and environmental conditions. Fulani have intuitive disease-reducing management practices (e.g. rapid disposal of animals that abort, or those with poor fertility or low milk yields, and removal of foetuses and placenta from the environment), and the low reproductive rates in KGR should also reduce transmission (Racloz et al., 2013). In addition, animals on the move in the sunny, dry and hot weather of the Nigerian Savannah (conditions that, in contrast to those of temperate climates, drastically reduce pathogen viability in the environment [Corbel et al., 2006]) should be less exposed to severe challenges, as proposed by early Nigerian researchers (Esuruoso, 1974) and suggested by others for the lowlands of Ethiopia (Berhe et al., 2007). Observations in Mozambique, Cameroon, Ivory Coast, Burkina Faso and Niger also suggest a correlation between climate and rate of infection, the latter being lower in dry and hot weather (Akakpo, 1987; Akakpo and Bornarel, 1987). Significantly, the same diagnostic methodology used in KGR found individual apparent seroprevalences of ca. 19% and 13% in cattle (n, ca. 350) of nearby intensive dairy farms. This exemplifies how management can drastically alter the picture of brucellosis in the same area and the high risk of brucellosis emergence in intensively managed herds in SSA and elsewhere (Akakpo, 1987; Ducrotoy et al., 2014; McDermott and Arimi, 2002). *B. abortus* biovar 3a was isolated from cattle in both KGR and the intensive farms, which illustrates the potential for extensively managed herds to act as reservoirs even though in these herds the disease is manifested only in a low number of individuals.

Diametric opposite epidemiological pictures to those seen in KGR and the close intensive farms are not uncommon. Concurrent low individual but high herd seroprevalences have been reported before (Akakpo, 1987; Akakpo and Bornarel, 1987; McDermott and Arimi, 2002) and since 2001 they have been documented in Cameroon, Ethiopia and Zimbawe in different production systems (Table 2). Both early and more recent studies have also found high seroprevalences at both individual and herd level in extensively or semi-extensively managed herds (McDermott and Arimi, 2002).

Table 4
Seroprevalence studies in humans in SSA since 2010[a].

Country	Population	Sampling	Diagnostic test (antigen/cut-off)	% Prevalence (no.)	Reference
Uganda	Abattoir workers	NS	MAT & STAT in series (≥1:60)	10 (232)	(Nabukenya et al., 2013; Schelling et al., 2003)
Togo	Fulani and non-Fulani villagers	Random selection villages (GPS ref) and households (along transect)	RBT (Bio-Rad)	0.44 (683)	(Dean et al., 2013; Megersa et al., 2012)
			iELISA IgG (manufacturer [Serion])	0.73 (683)	
			iELISA IgM ((manufacturer [Serion])	0.15 (683)	
Nigeria	Abattoir workers	Stratified random sampling	RBT (NS) & IgG/IgM iELISA (NS) in parallel	24.1 (224)	(Aworh et al., 2013; Megersa et al., 2011b)
Tanzania	Febrile patients	Prospective cohort study	MAT (NVSL)	3.5 (455)	(Ashenafi et al., 2007; Bouley et al., 2012)
Ethiopia	Febrile patients	Prospective cohort study	Rapid slide agglutination (Human GmbH, Wiesbaden))	2.6 (653)	(Animut et al., 2009)
Tanzania	Traditional & small-scale farming community	Cross-sectional survey	RBT (VLA)	5.5 (199)	(Swai and Schoonman, 2008)
Chad	Nomadic pastoralist community	Cross-sectional survey	RBT (Sanofi) & iELISA (CHEKIT/ROC plot) in series	3.9 (860)	(Schelling et al., 2003)
Namibia	Abattoir workers	Random sampling	SAT (Linear/≥1:80) & IgG/IgM ELISA (Panbio) in parallel	2.2 (137)	(Magwedere et al., 2012)
Ethiopia	Farm workers, abattoir workers, veterinarians	Cross-sectional survey	RBT (NS)	10.4 (336)	(Kassahun et al., 2006)
			2-ME (NS)	4.8 (336)	

[a] Abbreviations used in the Table: NS: not specificied; MAT: micro-agglutination test; STAT: serum tube agglutination test; RBT: Rose Bengal Test; iELISA: indirect ELISA; SAT: serum agglutination test; 2-ME: 2-mercaptoethanol test.

Several of the reports in Table 2 studies illustrate the relatively high herd seroprevalences (6.5–10.2%) that can be found in urban or peri-urban dairy or mixed dairy farms (Table 2), as observed by others (Mugizi et al., 2015a,b; Muma et al., 2007a,b; Yohannes et al., 2013). Indeed, these are of great concern as regards to public health. Unsurprisingly, the existence of a number of variables related to climate and geographical parameters, management, human habits, etc., across SSA make it difficult to predict the impact of the disease, emphasizing the necessity to carry out specific investigations.

As compared to cattle, there is limited information on brucellosis in small ruminants in SSA. It is considered that brucellosis in these livestock species is caused mostly by *B. melitensis* but, as indicated above, bacteriological studies are very limited (Table 1) and different epidemiological scenarios are plausible. The pattern of individual/collective prevalence discussed above for cattle is likely to apply to small ruminants, as suggested by the scarce literature available (Ducrotoy et al., 2014; McDermott and Arimi, 2002). The few recent studies generally indicate a low individual prevalence that could reflect a common existence of chronically infected flocks and herds (Table 3). However, flock or herd prevalences have been seldom if ever reported, and these are necessary for a complete picture and an assessment of the problems that the disease may pose if breeding conditions and epidemiological circumstances change.

Information on pig brucellosis is rather scarce and often of difficult interpretation because of the problems associated with the specificity of serological tests in these animals (Ducrotoy et al., 2014; McDermott and Arimi, 2002). Yet, its presence in at least Zimbabwe seems likely (see above). Similarly, camels present in the arid and semi-arid areas of Sahel and East Africa often react in serological tests for brucellosis, suggesting usually low individual seroprevalences (Adugna et al., 2013; Bekele et al., 2013; Ducrotoy et al., 2014; Gwida et al., 2012; McDermott and Arimi, 2002; Megersa et al., 2011a, 2012, 2006; Schelling et al., 2003; Sprague et al., 2012). When reported, herd seroprevalence has been found to be relatively high [15% (Megersa et al., 2012); 25.6% (Matope et al., 2011) and 24% (Bekele et al., 2013)]. However, the tests used in these surveys have not been validated in these animals (Sprague et al., 2012). Similarly, the scarce evidence on *B. canis* is based on tests of unknown specificity (Gous et al., 2005; Muhairwa et al., 2012), a critical problem because of the problems associated with agglutination tests (*B. canis* antigens auto-agglutinate [Carmichael and Shin, 1996]) and the cross reactivity between smooth and rough brucellae in immunoenzymatic methods (Nielsen et al., 2005).

3.3. Control and vaccination

Official data on the existence of control measures and degree of implementation (i.e. figures for outbreaks, number of animals involved, slaughtered and vaccinated, etc.) in SSA can be found on the OIE Handistatus webpage (http://web.oie.int/hs2/) for the 1996–2004 period, and in the WAHID (http://www.oie.int/wahis_2/public/wahid.php/Wahidhome/Home) from 2004 onwards. However, when they are contrasted with the literature and the estimated numbers and distribution of susceptible animals (Fig. 1), it is clear that these official records have very limited or no value. McDermott and Arimi (McDermott and Arimi, 2002) covered the information available up to 2002 and concluded that vaccination was rarely conducted outside of southern Africa and that, if done, it was mostly on an *ad hoc* basis, rather than as part of a systematic campaign. For this region, the disease was reported to be notifiable in several countries, and surveillance, movement control, and stamping out or vaccinations were implemented in Botswana, Namibia, Lesotho and South Africa. In Zimbabwe control programs were targeted to specific cattle production systems. A test-and slaughter program was partially implemented in goats in KwaZulu-Natal, which apparently managed to reduce prevalence but not to eradicate the infection. Problems reported in the implementation of this policy were not different from those observed elsewhere (i.e. time and financial constraints and stockowner disillusionments with the compensations) (Emslie and Nel, 2002).

Where implemented, vaccination of cattle was carried out using *B. abortus* S19 (McDermott and Arimi, 2002) and, since 2002, *B. abortus* RB51 (Davey, 2014; Ekron, 2008). More recently, vaccine RB51 has been introduced in other SSA countries, like Mozambique

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

and Zambia (Muma et al., 2012). South Africa is possibly the only country that has implemented a sustained effort based on a classical control and eradication strategy. However, bovine brucellosis has not been eradicated and it appears that the situation is worsening with more than 250 outbreaks/year reported since 2003 (Davey, 2014).

4. Brucellosis in wildlife

The earliest reports of serological evidence of exposure to *Brucella* spp. in free-ranging wild animals in Southern Africa are attributed to Rollinson (cited in Guilbride et al., 1962) and Guilbride and co-workers (Guilbride et al., 1962) in Tanzania and Uganda, respectively. Later, similar serological surveys in other countries such as Kenya (Waghela and Karstad, 1986), South Africa (De Vos and Van Niekerk, 1969), Tanzania (Sachs and Staak, 1966), Zambia (Bell et al., 1977) and Zimbabwe (Condy and Vickers, 1972) corroborated these reports. It is noteworthy that these and more recent studies consistently reported antibodies to *Brucella* spp. in wild animals like the African buffalo (*Syncerus caffer*), impala (*Aepyceros melampus*), blue wildebeeste (*Connochaetus taurinus*) and zebra (*Equus burchelli*), but were found to be erratic in other species (Alexander et al., 2012; Condy and Vickers, 1972; De Vos and Van Niekerk, 1969; Herr and Marshall, 1981; Waghela and Karstad, 1986). There are indications that the odds of seropositive reactors tend to increase in gregarious wild animal species such as the buffaloes, eland (*Taurotragus oryx*), impalas and wildebeest (Condy and Vickers, 1972; Madsen and Anderson, 1995) and are less of an issue in solitary animals like the white (*Ceratotherium simum*) and black (*Diceros bicornis)* rhinoceros. In fact brucellosis has not been reported in either the black or white rhinoceros (AQUIS, 1999; Motsi et al., 2013), and their susceptibility to *Brucella* spp. and/or ability to seroconvert needs to be explored further.

Although the *Brucella* spp. involved and wild animal host range have not been established, brucellosis has been confirmed by the isolation of *B. abortus* biovar 1 in buffaloes, eland (*Tragelaphus oryx*) and waterbuck (*Kobus ellipsipymnus*) (Condy and Vickers, 1969; Gradwell et al., 1977; Thornton, 1976) and *B. melitensis* from impalas (Schiemann and Staak, 1971). Similar to livestock, clinical signs of brucellosis in wildlife have been related to carpal hygroma, orchitis and abortions (Gradwell et al., 1977; Kaliner and Staak, 1973; Thornton, 1976). The observed inconsistency of reported abortions (Condy and Vickers, 1976) may be related to inadequate disease surveillance in wildlife.

The epidemiological significance of brucellosis in relation to the risk of transmission to livestock and humans has been debated extensively in the literature (Bishop and Bosman, 1994; Motsi et al., 2013). It is widely believed that *Brucella* spp. originate from livestock and spill over into wildlife, where in some species, they appear to perpetuate in the absence of contact with cattle. Although the general argument is that the risk of transmission to domestic animals is low due to infrequent contact (Madsen and Anderson, 1995), the creation of the large landscape mosaics under the transfrontier conservation area (TFCA) initiatives in SSA has allowed sharing of the same ecological systems by wildlife, domestic animals and humans and may thus promote inter-species transmission of *Brucella* spp. Sharing the same ecological space with domestic animals has been shown to be an important risk factor for brucellosis in wildlife (Bell et al., 1977; Muma et al., 2010; Sachs and Staak, 1966) while in the absence of contact with cattle, for instance, the black lechwe (*Kobus leche smithemani*) in Bangweulu plains in Zambia have remained free of brucellosis for many years (Muma et al., 2011). Therefore, the interface created in the TFCAs may bring about an intractable situation where brucellosis is introduced from domestic animals into wildlife where control is hardly practicable. The presence of *Brucella* spp. in wildlife in addition to domestic animals compounds the public health risk of brucellosis, especially for resource-poor communities living at the periphery of the TFCAs since their livelihood is largely dependent on animals and animal products. Therefore, in the quest for One Health in the TFCAs and surrounding areas, it would be important to break the transmission of *Brucella* spp. by re-enforcing control with the view of eradicating brucellosis in domestic animals in these areas (see below).

5. Human brucellosis in SSA

Similar to the disease in ruminants, there is fragmented serological evidence of human brucellosis across the different SSA countries (Ducrotoy et al., 2014; Gomo et al., 2012; McDermott and Arimi, 2002; Pappas et al., 2006; Rubach et al., 2013) but few assess disease frequency using probability sampling methods. Dean et al. (Dean et al., 2012) reviewed the literature published between 1990 and June 2010 and found only one article providing valid data on brucellosis frequency in Africa (Chad). Table 4 covers more recent but similarly scarce data that show the presence of antibodies, particularly in the classical risk groups (concerns about the use of non-validated diagnostic methods also apply here). Regrettably, there is an almost total absence of bacteriological studies (see above) that are strictly necessary to establish whether the relative risks of contact with *B. abortus* and *B. melitensis* observed elsewhere applies to SSA. Indeed, reports on infections by either *B. abortus* or *B. melitensis* based on antibody titres against the respective antigens such as those provided in “febrile antigen” kits (Chipwaza et al., 2015; Ducrotoy et al., 2014) are meaningless as it was established over 70 years ago that identification of infecting species is not possible by serology (Huddleson, 1943; Spink, 1956).

Suspicion of underreporting is accentuated by the fact that a significant proportion of malaria-suspected cases actually show a brucellosis-positive serology (McDermott and Arimi, 2002). Although in general the number of human cases should follow the incidence in animals and be higher in the classical risk groups (Ducrotoy et al., 2014; McDermott and Arimi, 2002), the situation can fluctuate depending on the proximity of contact between humans and livestock, the existence of different *Brucella* spp. and biovars, the alimentary and cultural habits of the population and possibly other factors. In a large study carried out in Niger, Ivory Coast and Burkina Faso (Gidel et al., 1976) it was found that, while the animal disease was more prevalent in the South, the rate of human infections was higher in the Northern Sahelian areas, and authors attributed this to the alimentary habits of the local populace and to the closer animal-human contacts due to environmental conditions (see also (Roux, 1979). For KGR (see above), no seropositives were detected in over 1100 humans tested using the rose bengal test (RBT), serum agglutination and Coombs tests, including butchers. Serologically negative human cases are exceedingly infrequent (Díaz et al., 2011) and, therefore, it seems that human brucellosis in KGR is remarkably rare. This is striking because Fulani in the KGR engage in risky practices such as drinking of unpasteurized milk. Several hypotheses can be proposed to account for this result, including (1) reduced persistence of the pathogen in the environment (see above), (2) shedding of brucellae in milk in numbers lower than those observed in cattle reared under optimal conditions, and (3) the cattle management habits of Fulani (3.1). A comparatively lower virulence of biovar 3a for humans cannot be ruled out, as this is known to occur in some *B.* suis biovars, but this hypothesis is not fully consistent with some bacteriological evidence (see above), and the identification of human disease in outbreaks and risk groups in Nigeria (Ducrotoy et al., 2014).

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

6. Tools for the SSA context

6.1. Human brucellosis

Under most circumstances, the existence of human brucellosis is the best indicator of the animal disease, and emphasis has to be placed on its diagnosis. This is based on the existence of compatible clinical symptoms and, as these overlap with those of some other infectious diseases, laboratory tests. An important consideration that emphasizes the need for laboratory tests is that the overall clinical picture of the human disease is often different in urban areas with easier access to medical facilities, where cases may be detected at an earlier stage, than in rural areas where chronic forms are likely to develop, or depending on the infecting *Brucella* species (Ariza, 1999; Young, 1989). Furthermore, inappropriate therapy caused by misdiagnosis may hide the symptoms and thus lead to the development of permanent sequelae without awareness of their brucellosis aetiology (Roux, 1979). It is obvious that all these circumstances are relevant in many SSA settings.

Bacteriological culture (usually blood culture) is of great value but it requires both skill and adequate facilities that are seldom available in SSA. On the other hand, simple serological tests combined with an evaluation of each case by a clinician should yield a correct diagnosis in the overwhelming majority of cases. In brucellosis, agglutinating antibodies are progressively substituted by non-agglutinating antibodies and, therefore, standard agglutination tests need to be complemented with the Coomb's test for brucellosis (Ariza, 1999; Díaz et al., 2011). Immunoenzymatic tests can detect both types of antibodies but they have not been properly validated for human brucellosis. Simpler and very effective are RBT and the Brucellacapt, two tests that detect both non-agglutinating and agglutinating antibodies because of the acidic conditions used (Díaz et al., 2011). RBT has shown its usefulness in resource-poor settings particularly when adapted to test serum dilutions rather than plain serum (Díaz et al., 2011; Mantur et al., 2014), and its implementation is highly recommended. It is noteworthy that the first recommendations in this regard came from the early work of African physicians (Oomen and Waghela, 1974). The relatively recently developed lateral flow immunochromatography test (Smits et al., 2003) is also a simple and promising test (Díaz et al., 2011), even though the commercial kit presently available shows serious standardization problems (Conde-Álvarez, R. Díaz and I. Moriyón unpublished observations).

Therapy is based on expensive combined antibiotic regimes that need to be modified when complications develop, or in pregnant women and children, and has to be sustained for weeks to minimize relapses (Ariza et al., 2007). However, in many resource-poor settings compliance is difficult and, moreover, the classical treatment (aminoglycosides plus doxycycline) conveys the risk of hepatitis and HIV transmission through syringes. Solera (Solera, 2010) has emphasized that prolonged doxycycline monotherapy (cheap and oral) should be useful in most cases, and this alternative deserves specific clinical trials in SSA.

6.2. Animal brucellosis

6.2.1. Diagnostics

While there is a limited choice of media and protocols for the bacteriological diagnosis of animal brucellosis (Alton et al., 1988; de Miguel et al., 2011), there are a bewildering number of serological tests. Those most often used in blood sera include the RBT (or its equivalent, the card test), serum agglutination in tube (SAT; not recommended by OIE for brucellosis testing for the purpose of international trade), the complement fixation test (CFT), the indirect (iELISA) and competitive (cELISA) enzyme-immunoassays, the fluorescence polarization assay (FPA) and the lateral flow immunochromatography test. These tests can be used for sero-epidemiological surveys but also to monitor the disease once control (and vaccination) is implemented. In the latter case, the RBT screening followed by the assessment of the RBT-positive sera by CFT has been widely used in countries where eradication was achieved and where the relatively sophisticated CFT can be implemented, often in an automatized format.

Although RBT is generally used as a screening test for its simplicity and CFT as a "confirmatory" test, this diagnostic strategy has led to the extended belief that RBT shows low specificity in all circumstances and is only a "presumptive" test (Ducrotoy et al., 2014). Following this misconception, an increasing tendency in recent literature is the reliance on technically appealing and expensive quantitative tests, such as iELISA, cELISA or FPA, to the detriment of the simpler, cheaper and more robust RBT. However, whereas it is well established that iELISA and cELISA do not have better sensitivity and specificity than RBT in the absence of vaccination (Greiner et al., 2009), even under the conditions in SSA (Nizeyimana et al., 2013), they have to be validated in the target populations (OIE, 2013a). Moreover, cut-offs recommended by the makers in Europe, the USA or Canada are unlikely to be adequate in SSA. The main reason for this is the existence of biological factors affecting the serological background that are not equal in different populations (Greiner and Gardner, 2000; Thrusfield, 2013). In brucellosis, such factors include management, male/female ratios and age distribution, animal breed, differences in reproductive periods, repeated exposure to the pathogen in endemic areas, vaccination (by either smooth or rough vaccines; see below) and variable degrees of exposure to cross-reacting bacteria related to hygiene, to autochthonous microbiota or other causes. The trend to use these more fashionable tests seems to uncritically follow the necessity of developed economies to use tests that can be automated for the survey of very large numbers of animals in brucellosis-free areas, and overlooks that diagnosis of brucellosis in successful control and eradication programs was achieved efficiently using simpler tests (Alton, 1981; Moreno, 2014). Yet, the experience of the authors of this review and others (Maichomo et al., 1998) is that even simple tests such as RBT require proper training and standardization, like any other brucellosis test.

For antibodies in milk, the literature reveals an indiscriminate use of the milk ring test (Ducrotoy et al., 2014) overlooking that this test can only be used in cattle and not in small ruminants or camels, and that it is affected by the quality of milk (Alton et al., 1988). A milk antibody test could be useful in some circumstances in SSA but, although it is well known that several ELISA formats can be adapted to detect immunoglobulins in milk, to the best of our knowledge none has been validated for diagnostic sensitivity/specificity in any SSA setting. Similarly, a particularly difficult and largely unsolved problem is an accurate serological diagnosis of brucellosis in wildlife species. Indeed, the diagnostic specificity and sensitivity of serological tests need to be established for each animal species using appropriate diagnostic criteria as references, and suitable studies investigating the performance of serological tests in African wildlife are rare or non-existent. Regrettably, the same problem affects the interpretation of serological results in camels (Sprague et al., 2012).

6.2.2. Vaccines and complementary measures

Three brucellosis vaccines are presently marketed worldwide: *B. abortus* S19 and *B. abortus* RB51 against brucellosis in cattle and *B. melitensis* Rev 1 against brucellosis in small ruminants.[1] It is impor-

[1] Here, we only consider the vaccines accepted by OIE. Among the long series of proposed brucellosis vaccines, only vaccine *B. suis* 2 has been used extensively and only in China. This vaccine was claimed to protect all animal species, even orally (Xin,

tant to stress that they are "host vaccines" and not vaccines against a given *Brucella* species. Accordingly, S19 (or RB51) should be used only in cattle and Rev 1 only in small ruminants no matter whether the animals are exposed to *B. abortus* or *B. melitensis*, as the bacteriological evidence suggests may happen in different SSA contexts (see above). In all cases, emphasis must be placed on the use of vaccines produced under strict quality control to prevent genetic drifts and dissociation. Internationally accepted biological and bacteriological protocols for quality control exist for S19 and Rev 1 but surprisingly not for RB51 (OIE, 2013a). To the best of our knowledge, with the exception of South Africa, no SSA country has the necessary technology, and vaccines produced locally, particularly in liquid form, are of no guarantee. These are freeze-dried live vaccines and, therefore, the cold chain should be maintained to guarantee viability and constant bacterial numbers of the final lyophilized product. The reader is directed to a recent review where the advantages, potential problems and conditions of use of the available vaccines are discussed in depth (Blasco et al., 2015), and here we only summarize their main characteristics: protection, interference in serological tests and abortifacient effect. When these are considered together, it is clear that S19 is the only cattle vaccine that can be recommended for SSA, and that conjunctival vaccination is the route of choice for vaccine administration.

Both controlled studies and field observations with appropriate control groups demonstrate that S19 is superior to RB51 (Moriyón et al., 2004). Contrary to strain 19 that provides long-life useful immunity (Nicoletti, 1990) the "protective" lapse span of RB51 vaccine has not been investigated but RB51 revaccination, even of pregnant animals, has been used without success in attempts to bolster immunity (Herrera et al., 2008; Herrera-Lopez et al., 2010; Leal-Hernandez et al., 2005; Luna-Martinez and Mejia-Teran, 2002; Moriyón et al., 2004). Moreover, no controlled experiments have addressed the advantages of these expensive revaccination strategies. Finally, it has been proven that S19 protects cattle against *B. melitensis* infection, but this has not been proven for the RB51 vaccine. Concerning the serological interference, it is usually argued that while S19 vaccination interferes in serological diagnosis, RB51 does not. However, RB51 vaccination generates antibodies reacting in iELISA, cELISA (Nielsen et al., 2005) and FPA (Blasco and Moriyón, unpublished results). Moreover, animals vaccinated with RB51 develop anti-S-LPS antibodies when exposed to virulent strains (Moriyón et al., 2004), as expected, which shows that they may become positive in S-LPS tests under field conditions creating confusion on the infection or vaccinated status. An additional consideration is that serological interference in standard tests is only important when animals are tagged and a test-and-slaughter strategy can be implemented, which is not the case in the overwhelming majority of situations in SSA. Indeed, if serological interference becomes relevant (when moving from control to eradication) the problem can be minimized by applying S19 by the conjunctival (rather than the subcutaneous) route during calfhood (Nicoletti et al., 1978; Plommet and Fensterbank, 1976; Jiménez de Bagüés et al., 1991).

Both S19 and RB51 can induce abortions in pregnant cattle, but this is reduced to less than 1% when S19 is administered by conjunctival route (see below). Despite early claims (Schurig et al., 2002), a high proportion of animals vaccinated with RB51 in the second half of the gestation abort (Mainar-Jaime et al., 2008; OIE, 2013a). Subcutaneous vaccination of males with S19 is not recommended because of the risk of development of genital infections. It would be worth investigating whether conjunctival S19 vaccination is safe in bulls as dispersion of the vaccine through the body is very limited by this route (Blasco et al., 2015).

Rev 1 has been essential in the eradication of *B. melitensis* infection in sheep and goats in some European Mediterranean countries and the properties and drawbacks of this vaccine are well-known (Blasco, 1997). As for S19, conjunctival vaccination is the procedure of choice. Using this route the post-vaccinal serological response is minimized and Rev 1 is safe in rams and billy goats. However, no matter the route, this vaccine is highly abortifacient and is excreted in milk when applied to pregnant sheep and goats. As indicated above, Rev 1 is markedly virulent for humans and, moreover, it is resistant to streptomycin (a drug that combined with oxytetracycline is of choice for treating human brucellosis [see below]). These are important drawbacks when "mass vaccination" is the only possible strategy (see below).

When using brucellosis vaccines, it is critical to understand that there is a biological limit to their effectiveness. For *B. abortus*, it has been estimated that up to 10^{14} bacteria are released during an abortion (Corner, 1983), which altogether represents about 10^9 and 10^5 times the estimated infectious dose 70 of unvaccinated and S19 vaccinated heifers (Manthei, 1959). Thus, it is not surprising that even the immunity provided by the best vaccines can be overcome upon continuous and direct exposure to recent abortions and abortion fluids. Revaccination does not improve immunity and, indeed, this would be unpractical or unfeasible under most conditions. Therefore, it is clear that for any vaccination program to be successful it is essential to create conditions that minimize exposure. Of utmost importance is the separation of animals during calving and lambing/kidding and in the next weeks that follow, plus proper disposal of aborted foetuses and placentas, the removal of replacements born to infected mothers, and other well-known general hygiene measures (Corbel et al., 2006). Education campaigns to improve awareness of brucellosis and other zoonoses with emphasis on the mechanisms of transmission are of paramount importance.

6.3. Control and eradication strategies

Long experience shows that interventions to control and eradicate brucellosis need to be based on engagement with livestock keepers, education and complementary measures and, where feasible and necessary, vaccination (Corbel et al., 2006). When the resources and capacity for control are limited, interventions could be tailored to the impact of the disease in terms of magnitude of burden in potential animal and human hosts. Evaluating the impact of brucellosis, however, is difficult because of the dual burden of disease in humans and animals, the role of several animal host species in transmission or as reservoirs, the diverse epidemiological picture at herd or flock level, and the variability of the clinical picture in humans. Broadly speaking, where human disease is an occasional occurrence and the individual prevalence remains low in animals, brucellosis possibly ranks as a low priority disease whose effects can be minimised by education, basic hygiene measures in animal management and milk pasteurization. However, it has to be kept in mind that these herds and flocks represent a reservoir that can affect other production systems and develop into a more serious situation if the conditions change. Thus, vaccination should be considered even in these low prevalence situations. On the other hand, if human brucellosis is significant, the problem has a completely different dimension that, in addition, will most often include an important incidence in animals causing significant economic losses by reducing meat and milk outputs. Even though each case requires specific studies in SSA, intensive production systems are more threatened by brucellosis and small holders or backyard livestock keepers in zero grazing systems are particu-

1986). These claims attracted some attention in the 70's, and FAO commissioned several studies to independent reference laboratories. These experiments performed both in laboratory models and under controlled conditions in natural hosts (Blasco et al., 1993; Bosseray and Plommet, 1990; Verger et al., 1995) demonstrated the poor efficacy of this vaccine and its use was thus unsupported by FAO.

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

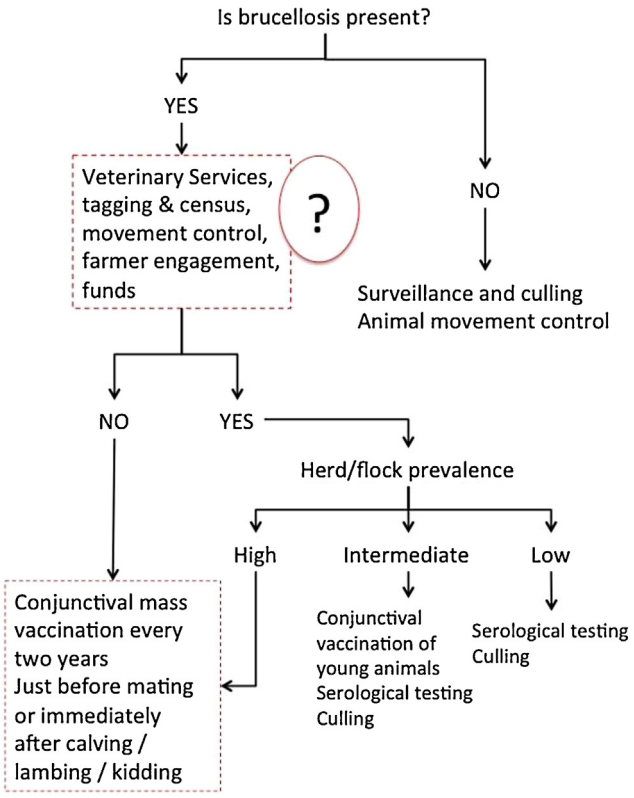

Fig. 2. Decision making tree for controlling/eradicating brucellosis.
Once the presence of brucellosis has been established in humans and/or animals, the critical points are whether the veterinary services can effectively implement vaccination, animals can be individually identified by tagging, a census is available, breeders are actively engaged in the program and the necessary funds available. When these requisites are not met, mass vaccination (*B. abortus* S19 in cattle and *B. melitensis* Rev 1 in small ruminants) is the only option, no matter the prevalence (for details see text). When collective prevalence is high, no matter whether those requisites are met, mass vaccination is also the only realistic option. Usually, after implementation of mass vaccination and even though the collective prevalence remains high, the individual prevalence decreases and then a program based on the combination of vaccination of young replacements with serological testing and culling of seropositives can be considered after careful re-evaluation of the means available. A final step under very favourable conditions would be the removal of vaccination. Yet, experience shows that the most common mistake in brucellosis control/eradication is the premature removal of S19 and Rev 1 (or the substitution of the former by less effective vaccines) a situation that, but for international trade requisites, is unnecessary and of no benefit to most breeders in poor-resource areas.

larly vulnerable (Mangen et al., 2002). In these cases, vaccination is immediately necessary. In addition, attention should be given to the communities in the periphery of the TFCAs because of the clear risk of introduction of brucellosis into wildlife. If this happened, then control in wild species would be exceedingly difficult (if not impossible) with subsequent health implications to the new hosts and the establishment of a permanent reservoir, as the cases of the American bison and elk illustrates (Treanor, 2013).

Fig. 2 summarizes the strategies proposed by several authors for the control and eradication of the disease (Benkirane, 2006; Blasco and Molina-Flores, 2011). Prevalence at herd or flock level (collective prevalence), level of veterinary coverage and organization, appropriate control of animal movements and economic resources are the main requisites necessary to decide on the strategy once the presence of brucellosis (including the human disease) has been established. When the proficiency of veterinary services is not adequate (no matter the prevalence), and when collective prevalence is high, mass vaccination of all animals (with the exception of bovine males) of all species is the only strategy to control the disease. All the above requisites are essential, however, when moving towards eradication programs based on the vaccination of young replacements (3–4 months old) and test and slaughter in adult animals or when the eventual banning of vaccination and implementation of a test-and-slaughter only program is foreseen (Fig. 2).

In addition to economic, political and geographical difficulties, animal health services provided by the public sector are insufficient in SSA and, in general, the gap has not been filled by the private sector (McDermott and Arimi, 2002). A strategy based on vaccination of young replacements (3–4 months old) and test-and-slaughter in adult animals is unrealistic under extensive pastoralist production in SSA and elsewhere (Blasco et al., 2015; Blasco and Molina-Flores, 2011). Test and slaughter is not feasible and, because owners keep replacements throughout the year in these systems, several field visits would be required to attain full vaccine coverage, which increases costs and is seldom feasible. This problem is compounded by the difficulties of including small farms with very few animals and localizing all animals in nomadic pastoralist systems. Accordingly, control by repeated (see below) mass vaccination is the only realistic strategy for most SSA areas. Indeed, this does not exclude actions targeted to control brucellosis in specific production systems or farms or to the more vulnerable settled herds and flocks. For these systems, once prevalence is reduced, vaccination could be restricted to young replacements to solve the problems intrinsic to mass vaccination that are commented upon below.

Mass vaccination (i.e. vaccinating all animals—but for males in the case of S19—with a reduced dose dispensed by conjunctival instillation regardless of age and pregnancy) was proposed in the early seventies for resource poor or remote areas on the basis that, since testing is not routine and test-and-slaughter not applicable in such areas, the serological interference is irrelevant. Moreover, census and identification (ear tagging is not necessarily permanent, it is expensive and may promote myiasis and ear infections in tropical and hot climates) of vaccinated animals are not possible in those areas. However, the undesirable side effects of the vaccines on pregnant and lactating animals should be explained to livestock keepers. Although conjunctival S19 vaccination of lactating cows results in 0.1–1% of mammary infections and milk excretion and in 0.1–1% abortions in pregnant cows, experience shows that adult vaccination is the most practical and economic way to control the disease in cattle (Blasco et al., 2015). Moreover, it has been shown that when mass vaccination is implemented, removal of seropositive animals does not result in a more rapid decrease of prevalence (Enright and Hugh-Jones, 1984), which greatly facilitates implementation of this strategy in difficult contexts.

Regrettably, the safety problems of Rev 1 in pregnant sheep and goats are far greater than those of S19 in cattle and the rates of abortion and milk excretion can be very high. This together with the subsequent risks of human infections by Rev 1 make the vaccination of pregnant sheep and goats unacceptable (Blasco, 1997). Conjunctival vaccination during the late lambing or kidding, lactation and pre-mating periods are indeed the safest times. Thus, it may be possible to find relatively safe “time windows” in areas where parturitions are concentrated across a few weeks because of the seasonal availability of pastures, or where the demand of animals peaks in a short period because of cultural, religious or other reasons. Use of this vaccine in contexts where there is no seasonal breeding, however, is problematic.

To be effective, vaccination has to be maintained over time. Once a first mass vaccination with S19 or Rev 1 has been applied, the ideal procedure would be to vaccinate only the next young replacement generation every year. This would require individual tagging, a measure that, as indicated above, is unrealistic in many situations in SSA. An alternative is to consider the rates of annual replacement and the epidemiological risks according to age, i.e. sexually mature,

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

pregnant animals are more liable to become infected and susceptibility may increase with the length of gestation (Enright, 1990). For example, if the annual replacement were about 20%, as in many extensive management systems, one year after the first mass vaccination only 1/5 of the livestock would be young replacements. Since these replacements are sexually immature they are excluded from the period of maximal risk of infection and spreading and it is acceptable not to vaccinate them. However, in the next year, almost half of the population would be unprotected and a significant proportion would be sexually mature, so that mass vaccination is again in order. This two-year mass vaccination strategy has been applied successfully with few side effects in areas where those safe "time windows" exist (Blasco, 1997). For Rev 1, it is important to stress that failure to adhere to such windows may have dramatic consequences in terms of abortions and milk excretion. There is a clear need for a safer small ruminant brucellosis vaccine for areas where these conditions do not apply.

Monitoring of vaccination programs is essential. A representative sample of animals should be tested with RBT at 15–21 days after vaccination and between 70 and 90% of these vaccinated animals should test positive. To assess the efficacy of the control strategy, the evolution of brucellosis in the exposed human population in the vaccinated area should be followed through screening of people with the RBT.

7. Concluding remarks

Presently, brucellosis in SSA poses a series of challenges that encompass not only the necessary assessment of the prevalence of the disease in humans and animals, including wildlife, and the influence of the various local epidemiological characteristics but also several important methodological gaps that concern the tools to diagnose and combat the disease. As outlined above, although simple and affordable serological tests are available for the diagnosis of human brucellosis, there is a need for simpler and more affordable antimicrobial treatments. Concerning brucellosis in domestic livestock, perhaps the greatest need concerns the development of a *B. melitensis* vaccine that could circumvent the abortifacient effects of Rev 1. These represent a serious drawback for the implementation of mass vaccination in small ruminants, the only realistic strategy in many areas of SSA and elsewhere. The need to investigate the safety of S19 in bulls when administered conjunctively has been stressed above. It is also important to fill the gap in serological diagnostic tests for camel brucellosis, and to investigate the performance of serological tests in wildlife. Brucellosis in pigs remains unexplored and very little is known about *B. canis* and *B. ovis*. Finally, although out of the scope of the present review, it is important to stress that control strategies based on any existing and future tools need delivery methods optimized for the various socio-economic contexts in SSA countries (McDermott and Arimi, 2002).

Acknowledgements

We are grateful to Ward Bryssinckx (Avia-GIS, Belgium) for guidance in preparing the maps included in Fig. 1.

Research at the laboratories of RO, SW and IM has received funding from the European Union's Seventh Framework Program (FP7/2007–2013) under grant agreement n° 221948, ICONZ (Integrated control of Neglected Zoonoses). The contents of this publication are the sole responsibility of the authors and do not necessarily reflect the views of the European Commission. Support from "Ministerio de Economía y Competitividad" of Spain (AGL2014-58795-CA) is also gratefully acknowledged.

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.actatropica.2015.10.023.

References

Adugna, K.E., Agga, G.E., Zewde, G., 2013. Seroepidemiological survey of bovine brucellosis in cattle under a traditional production system in western Ethiopia. Rev. Sci. Technol. 32, 765–773.
Akakpo, A.J., 1987. Brucellose animales en Afrique tropicale. Particularités épidémiologiques: cliniques et bactériologiques. Rev. Elev. Med. Vet. Pays. Trop. 40, 307–320.
Akakpo, A.J., Bornarel, P., 1987. Epidémiologie des brucelloses animales en Afrique tropicale: enquêtes clinique, sérologique et bactériologique. Rev. Sci. Technol. 4, 981–1027.
Alexander, K.A., Blackburn, J.K., Vandewalle, M.E., Pesapane, R., Baipoledi, E.K., Elzer, P.H., 2012. Buffalo, bush meat, and the zoonotic threat of brucellosis in Botswana. PloS One 7, e32842.
Allsup, T.N., 1969. Abortion in sheep associated with *Brucella abortus* infection. Vet. Rec. 84, 104–108.
Alonso-Urmeneta, B., Marin, C.M., Aragon, V., Blasco, J.M., Díaz, R., Moriyón, I., 1998. Evaluation of lipopolysaccharides and polysaccharides of different epitopic structures in the indirect enzyme-linked immunosorbent assay for diagnosis of brucellosis in small ruminants and cattle. Clin. Diagnost. Lab. Immunol. 5, 749–754.
Alton, G.G., 1981. The control of bovine brucellosis. Recent developments. World Anim. Rev., 17–24.
Alton, G.G., Jones, L.M., Angus, R.D., Verger, J.M., 1988. Techniques for the Brucellosis Laboratory. INRA, Paris, France.
Animut, A., Mekonnen, Y., Shimelis, D., Ephraim, E., 2009. Febrile illnesses of different etiology among outpatients in four health centers in Northwestern Ethiopia. Jpn. J. Infect. Dis. 62, 107–110.
AQUIS, 1999. Review of Conditions for the Importation of Rhinoceros from South Africa. Final Report. Australian Quarantine and Inspection Service, Canberra.
Ariza, J., 1999. Brucellosis: an update: the perspective from the Mediterranean basin. Rev. Med. Microbiol. 10, 125–135.
Ariza, J., Bosilkovski, M., Cascio, A., Colmenero, J.D., Corbel, M.J., Falagas, M.E., Memish, Z.A., Roushan, M.R., Rubinstein, E., Sipsas, N.V., Solera, J., Young, E.J., Pappas, G., 2007. Perspectives for the treatment of brucellosis in the 21st century: the Ioannina recommendations. PLoS Med. 4, e317.
Ashenafi, F., Teshale, S., Ejeta, G., Fikru, R., Laikemariam, Y., 2007. Distribution of brucellosis among small ruminants in the pastoral region of Afar, eastern Ethiopia. Rev. Sci. Technol. 26, 731–739.
Asmare, K., Megersa, B., Denbarga, Y., Abebe, G., Taye, A., Bekele, J., Bekele, T., Gelaye, E., Zewdu, E., Agonafir, A., Ayelet, G., Skjerve, E., 2013a. A study on seroprevalence of caprine brucellosis under three livestock production systems in southern and central Ethiopia. Trop. Anim. Health Prod. 45, 555–560, http://dx.doi.org/10.1007/s11250-012-0258-2.
Asmare, K., Sibhat, B., Molla, W., Ayelet, G., Shiferaw, J., Martin, A.D., Skjerve, E., Godfroid, J., 2013b. The status of bovine brucellosis in Ethiopia with special emphasis on exotic and cross bred cattle in dairy and breeding farms. Acta Trop. 126, 186–192, http://dx.doi.org/10.1016/j.actatropica.2013.02.015.
Ate, I.U., Bello, A., Nenshi, P.M., Allam, L., Rashidat, M., 2011. Foetal maceration associated with *Brucella ovis* infection in a Yankassa ewe. REDVET Rev. Electron. Vet. 12, 1–6.
Aworh, M.K., Okolocha, E., Kwaga, J., Fasina, F., Lazarus, D., Suleman, I., Poggensee, G., Nguku, P., Nsubuga, P., 2013. Human brucellosis: seroprevalence and associated exposure factors among abattoir workers in Abuja, Nigeria—2011. Pan. Afr. Med. J. 16, 103.
Bale, O.O.J., Kumi-Diaka, J., 1981. Serological and bacteriological study of bovine brucellae from livestock investigation and breeding centres in Nigeria. Br. Vet. J. 137, 256–261.
Bale, O.O.J., Nuru, S., Addo, P.B., Adeyinka, I.A., 2003. Bacteriological investigation of sheep and goat milk for brucellosis in government farms in Northern Nigeria. Nigerian J. Anim. Prod. 30, 107–116.
Bankole, A.A., Saegerman, C., Berkvens, D., Fretin, D., Geerts, S., Ieven, G., Walravens, K., 2010. Phenotypic and genotypic characterisation of *Brucella strains* isolated from cattle in the Gambia. Vet. Rec. 166, 753–756.
Bekele, W.A., Tessema, T.S., Melaku, S.K., 2013. Camelus dromedarius brucellosis and its public health associated risks in the Afar National Regional State in northeastern Ethiopia. Acta Vet. Scand. 55, 89, http://dx.doi.org/10.1186/1751-0147-55-89.
Bell, L.M., Hayles, L.B., Chanda, A.B., 1977. Serological evidence of *Brucella melitensis* infection in goats and eland in Zambia. Vet. Rec. 101, 305.
Benkirane, A., 2006. Ovine and caprine brucellosis: world distribution and control/eradication strategies in West Asia/North Africa region. Small Rumin. Res. 62, 19–25, http://dx.doi.org/10.1016/j.smallrumres.2005.07.032.
Berhe, G., Belihu, K., Asfaw, Y., 2007. Seroepidemiological investigation of bovine brucellosis in the extensive cattle production system of Tigray region of Ethiopia. Intern. J. Appl. Res. Vet. Med. 5, 65–71.
Bernard, F., Vincent, C., Matthieu, L., David, R., James, D., 2005. Tuberculosis and brucellosis prevalence survey on dairy cattle in Mbarara milk basin (Uganda).

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

Prev. Vet. Med. 67, 267–281, http://dx.doi.org/10.1016/j.prevetmed.2004.11.002.
Bertu, W.J., Ducrotoy, M.J., Muñoz, P.M., Mick, V., Zúñiga-Ripa, A., Bryssinckx, W., Kwaga, J.K.P., Kabir, J., Welburn, S.C., Moriyón, I., Ocholi, R.A., 2015. Phenotypic and genotypic characterization of Brucella strains isolated from autochthonous livestock reveals the dominance of *B. abortus* biovar 3a in Nigeria. Vet. Microbiol. (in press) http://dx.doi.org/10.1016/j.vetmic.2015.08.014.
Bevan, L.E.W., 1930. Blood culture in undulant fever. Br. Med. J. 2, 267.
Bishop, G.C., Bosman, P.P., 1994. Bovine brucellosis BT—infectious diseases of livestock with special reference to Southern Africa. In: Coetzer, J.A., Thompson, R.C. (Eds.), Infectious diseases of livestock with special reference to Southern Africa. Oxford University Press, pp. 1053–1066.
Blasco, J.M., 1997. A review of the use of *B. melitensis* Rev 1 vaccine in adult sheep and goats. Prev. Vet. Med. 31, 275–283.
Blasco, J.M., Marin, C.M., Jimenez de Bagües, M.P., Barberan, M., 1993. Efficacy of *Brucella suis* strain 2 vaccine against *Brucella ovis* in rams. Vaccine 11, 1291–1294.
Blasco, J.M., Molina-Flores, B., 2011. Control and eradication of *Brucella melitensis* infection in sheep and goats. Vet. Clin. North Am. Food Anim. Pract. 27, 95–104.
Blasco, J.M., Moreno, E., Moriyón, I., 2015. Brucellosis vaccines and vaccine candidates—veterinary vaccines for developing countries. In: Metwally, S., Viljoen, G.J., El Idrissi, A. (Eds.), Veterinary Vaccines for Developing Countries. FAO, Rome (in press).
Bosseray, N., Plommet, M., 1990. *Brucella suis* S2, *Brucella melitensis* Rev. 1 and *Brucella abortus* S19 living vaccines: residual virulence and immunity induced against three Brucella species challenge strains in mice. Vaccine 8, 462–468.
Boukary, A.R., Saegerman, C., Abatih, E., Fretin, D., Alambedji Bada, R., De Deken, R., Harouna, H.A., Yenikoye, A., Thys, E., 2013. Seroprevalence and potential risk factors for *Brucella* spp. infection in traditional cattle, sheep and goats reared in urban, periurban and rural areas of Niger. PloS One 8, e83175.
Bouley, A.J., Biggs, H.M., Stoddard, R.A., Morrissey, A.B., Bartlett, J.A., Afwamba, I.A., Maro, V.P., Kinabo, G.D., Saganda, W., Cleaveland, S., 2012. Brucellosis among hospitalized febrile patients in northern Tanzania. Am. J. Trop. Med. Hyg. 87, 1105–1111.
Cameron, R.D., Carles, A.B., Lauerman, L.H.J., 1971. The incidence of *Brucella ovis* in some Kenya flocks and its relationship to clinical lesions and semen quality. Vet. Rec. 89, 552–557.
Carmichael, L.E., Shin, S.J., 1996. Canine brucellosis: a diagnostician's dilemma. Semin. Vet. Med. Surg. Small Anim. 11, 161–165.
Catlin, J.E., Sheehan, E.J., 1986. Transmission of bovine brucellosis from dam to offspring. J. Am. Vet. Med. 188, 867–869.
Chabasse, D., Roure, C., Rhaly, A.A., Maiga, D., Traore, M., Tounkara, A., Dumon, H., Ranque, P., 1983. Evaluation de l'etat sanitaire des populations nomades et semi-nomades du Gourma-Mali: approche epidemiologique II. Resultats globaux et conclusion. Med. Trop. 43, 127–135.
Chambron, J., 1965. La brucellose bovine au Senegal. Rev. Elev. Med. Vet. Pays. Trop. 18, 19–38.
Chimana, H.M., Muma, J.B., Samui, K.L., Hangombe, B.M., Munyeme, M., Matope, G., Phiri, A.M., Godfroid, J., Skjerve, E., Tryland, M., 2010. A comparative study of the seroprevalence of brucellosis in commercial and small-scale mixed dairy-beef cattle enterprises of Lusaka province and Chibombo district, Zambia. Trop. Anim. Health Prod. 42, 1541–1545.
Chipwaza, B., Mhamphi, G.G., Ngatunga, S.D., Selemani, M., Amuri, M., Mugasa, J.P., Gwakisa, P.S., 2015. Prevalence of bacterial febrile illnesses in children in Kilosa district, Tanzania. PLoS Negl. Trop. Dis. 9, e0003750, http://dx.doi.org/10.1371/journal.pntd.0003750.
Cohen, N.D., Carter, G.K., McMullan, W.C., 1992. Fistulous withers in horses: 24 cases (1984–1990). J. Am. Vet. Med. Assoc. 201, 121–124.
Condy, J.B., Vickers, D.B., 1969. Isolation of *Brucella abortus* from a waterbuck (*Kobus ellipsiprymnus*). Vet. Rec. 85, 318.
Condy, J.B., Vickers, D.B., 1972. Brucellosis in rhodesian wildlife. J. S. Afr. Vet. 43, 175–179.
Condy, J.B., Vickers, D.B., 1976. Brucellosis in buffalo in Wankie National Park. Rhod. Vet. J. 7, 58–60.
Corbel, M.J., Alton, G.G., Banai, M., Díaz, R., Dranovskaia, B.A., Elberg, S.S., Garin-Bastuji, B., Kolar, J., Mantovani, A., Mousa, A.M., Moriyón, I., Nicoletti, P., Seimenis, A., Young, E.J., 2006. Brucellosis in Humans and Animals. WHO Press, Geneva.
Corner, L.A.A., 1983. Three aspects of bovine brucellosis: epidemiology: the role of bulls and vaccines. N. S. W. Vet. Proc. 19, 47–48.
Cunningham, B., 1977. A difficult disease called brucellosis BT. In: Crawford, R.M., Hidalgo, R.J. (Eds.), Bovine Brucellosis. An International Symposium. Texas A&M University Press, College Station, pp. 11–20.
Dalrymple-Champneys, W., 1960. Clinical features. In: Brucella Infection and Undulant Fever in Man. Oxford University Press, London, pp. 61–94.
Davey, S., 2014. Bovine brucellosis in South Africa, In: Proceedings of the annual congress of the Southern African Society For Veterinary Epidemiology and Preventive Medicine. August. Port Elizabeth, Republic of South Africa.
de Miguel, M.J., Marin, C.M., Muñooz, P.M., Dieste-Pérez, L.I.A., Grillo, M.J., Blasco, J.M., 2011. Development of a selective culture medium for primary isolation of the main *Brucella* species. J. Clin. Microbiol. 49, 1458–1463.
De Vos, V., Van Niekerk, C., 1969. Brucellosis in the Kruger national park. J. S. Afr. Vet. 40, 331–334.
De Wet, J.A., Erasmus, J.A., 1984. Epididymitis of rams in the central and southern districts of the Orange Free State. J. S. Afr. Vet. Assoc. 55, 173–179.
Dean, A.S., Crump, L., Greter, H., Schelling, E., Zinsstag, J., 2012. Global burden of human brucellosis: a systematic review of disease frequency. PLoS Negl. Trop. Dis. 6, http://dx.doi.org/10.1371/journal.pntd.0001865.
Dean, A.S., Bonfoh, B., Kulo, A.E., Boukaya, G.A., Amidou, M., Hattendorf, J., Pilo, P., Schelling, E., 2013. Epidemiology of Brucellosis and Q fever in linked human and animal populations in Northern Togo. PloS One 8, e71501, http://dx.doi.org/10.1371/journal.pone.0071501t003.
Dean, A.S., Schelling, E., Bonfoh, B., Kulo, A.E., 2014. Deletion in the gene BruAb2 _0168 of *Brucella abortus* strains: diagnostic challenges. Clin. Microbiol. Infect. 20, 550–553.
Díaz, R., Casanova, A., Ariza, J., Moriyón, I., 2011. The rose Bengal test in human brucellosis: a neglected test for the diagnosis of a neglected disease. PLoS Negl. Trop. Dis. 5, 1–7, http://dx.doi.org/10.1371/journal.pntd.0000950.
Doutre, M.P., Fensterbank, R., Sagna, F., 1977. Étude de la brucellose bovine dans un village de Basse-Casamance (Senegal): I. Diagnostic serologique et bacteriologique. Rev. Elev. Med. Vet. Pays Trop. 30, 345–351.
Ducrotoy, M.J., Bertu, W.J., Ocholi, R.A., Gusi, A.M., Bryssinckx, W., Welburn, S.C., Moriyón, I., 2014. Brucellosis as an emerging threat in developing economies: lessons from Nigeria. PLoS Negl. Trop. Dis. 8, http://dx.doi.org/10.1371/journal.pntd.0003008.
Ekron, M., 2008. Experiences using RB51 vaccine as an aid to brucellosis control in Mpumalanga Province I South Africa. In: Proceedings of the Brucellosis 2008 International Conference, Royal Holloway College, University of London, UK, 10–13 September 2008, p. p. 41.
El Ansary, E.H., Mohammed, B.A., Hamad, A.R., Karom, A.G., 2001. Brucellosis among animals and human contacts in eastern Sudan. Saudi Med. J. 22, 577–579.
Emslie, F.R., Nel, J.R., 2002. An overview of the eradication of *Brucella melitensis* from KwaZulu-Natal. Onderstepoort J. Vet. Res. 69, 123–127.
Enright, F.M., 1990. The pathogenesis and pathobiology of *Brucella* infection in domestic animals. In: Nielsen, K.H., Duncan, J.R. (Eds.), Animal Brucellosis. CRC Press, Boca Raton, pp. 301–320.
Enright, F.M., Hugh-Jones, M.E., 1984. Effects of reactor retention on the spread of brucellosis in strain 19 adult vaccinated herds. Prev. Vet. Med. 2, 505–514.
Esuruoso, G.O., 1974. Bovine brucellosis in Nigeria. Vet. Rec. 95, 54–58.
Eze, E.N., 1978. Isolation of *Brucellae* from the Nigerian livestock and the typing of such isolates. Bull. Anim. Health Prod. Afr. 26, 29–36.
Falade, S., 1981a. Caprine brucellosis: serological studies in Nigeria. Bull. Anim. Health Prod. Afr. 29, 157–161.
Falade, S., 1981b. Brucellae isolated from goats. Zentralbl. Veterinarmed. B. 28, 205–209.
Ferney, J., Chantal, J., 1976. Aspects cliniques et epidemiologiques de la brucellose bovine en Afrique tropicale. Dev. Biol. 31, 274–278.
Garin-Bastuji, B., Mick, V., Le Carrou, G., Allix, S., Perrett, L.L., Dawson, C.E., Groussaud, P., Stubberfield, E.J., Koylass, M., Whatmore, A.M., 2014. Examination of taxonomic uncertainties surrounding *Brucella abortus* bv. 7 by phenotypic and molecular approaches. Appl. Environ. Microbiol. 80, 1570–1579.
Gidel, R., Albert, J.P., Le, M.G., Retif, M., 1976. Epidèmiologie de la brucellose humain et animale en Afrique de louest. Results de dix enquetes efectuées en Côte D'Ivoire, Haute-Volta et Niger. Dev. Biol. Stand. 31, 187–200.
Godfroid, J., Bastuji, B.G., Saegerman, C., 2013. Brucellosis in terrestrial wildlife. Rev. Sci. Technol. 32, 27–42.
Godfroid, J., Scholz, H.C., Barbier, T., Nicolas, C., Wattiau, P., Fretin, D., Whatmore, A.M., Cloeckaert, A., Blasco, J.M., Moriyón, I., Saegerman, C., Muma, J.B., Al Dahouk, S., Neubauer, H., Letesson, J.-J., 2011. Brucellosis at the animal/ecosystem/human interface at the beginning of the 21st century. Prev. Vet. Med. 102, 118–131, http://dx.doi.org/10.1016/j.prevetmed.2011.04.007.
Gomo, C., de Garine-Wichatitsky, M., Caron, A., Pfukenyi, D.M., 2012. Survey of brucellosis at the wildlife-livestock interface on the Zimbabwean side of the Great Limpopo Transfrontier Conservation Area. Trop. Anim. Health Prod. 44, 77–85.
Gous, T.A., van Rensburg, W.J., Gray, M., Perrett, L.L., Brew, S.D., Young, E.J., Whatmore, A.M., Gers, S., Picard, J., 2005. *Brucella canis* in South Africa. Vet. Rec. 157, 668.
Grace, D., Mutua, F., Ocungo, P., Kruska, R., Jones, K., Brierley, L., Lapar, L., Said, M., Herrero, M., Duc Phuc, P., Bich Tao, N., Akuku, I., Ogutu, F., 2012. Mapping of poverty and likely zoonoses hotspots. Zoonoses Project 4, 1–119.
Gradwell, D.V., Schutte, A.P., van Niekerk, C.A., Roux, D.J., 1977. The isolation of *Brucella abortus* biotype I from African buffalo in the Kruger National Park. J. S. Afr. Vet. Assoc. 48, 41–43.
Greiner, M., Gardner, I.A., 2000. Epidemiologic issues in the validation of veterinary diagnostic tests. Prev. Vet. Med. 45, 3–22.
Greiner, M., Verloo, D., De Massis, F., 2009. Meta-analytical equivalence studies on diagnostic tests for bovine brucellosis allowing assessment of a test against a group of comparative tests. Prev. Vet. Med. 92, 373–381.
Guilbride, P.D.L., Coyle, T.J., McAnulty, E.G., Barber, L., Lomax, G.D., 1962. Some pathogenic agents found in hipopotamus in Uganda. J. Comp. Path 72, 137–141.
Gumaa, M.M., Osman, H.M., Omer, M.M., Sanousi, E.M.E., 2014. Seroprevalence of brucellosis in sheep and isolation of *Brucella abortus* biovar 6 in Kassala State, Eastern Sudan. Rev. Sci. Technol. 33, 957–965.
Gwida, M., El-Gohary, A., Melzer, F., Khan, I., Rosler, U., Neubauer, H., 2012. Brucellosis in camels. Res. Vet. Sci. 92, 351–355.
Herr, S., Marshall, C., 1981. Brucellosis in free-living African buffalo (*Syncerus caffer*): a serological survey. Onderstepoort J. Vet. Res. 48, 133–134.

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

Herrera-Lopez, E., Suarez-Guemes, F., Hernandez-Andrade, L., Cordova-Lopez, D., Diaz-Aparicio, E., 2010. Epidemiological study of brucellosis in cattle, immunized with *Brucella abortus* RB51 vaccine in endemic zones. Vaccine 28 (Suppl 5), F59–F63.
Herrera, E., Palomares, G., Diaz-Aparicio, E., 2008. Milk production increase in a dairy farm under a six-year brucellosis control program. Ann. N. Y. Acad. Sci. 1149, 296–299.
Hesterberg, U.W., Bagnall, R., Perrett, K., Bosch, B., Horner, R., Gummow, B., 2008. A serological prevalence survey of *Brucella abortus* in cattle of rural communities in the province of KwaZulu-Natal, South Africa. J. S. Afr. Vet. Assoc. 79, 15–18.
Huddleson, I.F., 1943. Laboratory diagnosis of brucellosis, in: Brucellosis in Man and Animals. New York, 214–284.
Ibrahim, N., Belihu, K., Lobago, F., Bekana, M., 2010. Sero-prevalence of bovine brucellosis and its risk factors in Jimma zone of Oromia Region, South-western Ethiopia. Trop. Anim. Health Prod. 42, 35–40.
Jergefa, T., Kelay, B., Bekana, M., Teshale, S., Gustafson, H., Kindahl, H., 2009. Epidemiological study of bovine brucellosis in three agro-ecological areas of central Oromiya, Ethiopia. Rev. Sci. Technol. 28, 933–943.
Jiménez de Bagüés, M.P., Marín, C.M., et al., 1991. Effect of antibiotic therapy and strain-19 vaccination on the spread of *Brucella melitensis* within an infected dairy herd. Prev. Vet. Med. 11, 17–24.
Kadohira, M., McDermott, J.J., Shoukri, M.M., Kyule, M.N., 1997. Variations in the prevalence of antibody to brucella infection in cattle by farm, area and district in Kenya. Epidemiol. Infect. 118, 35–41, http://dx.doi.org/10.1017/s0950268896007005.
Kaliner, G., Staak, C., 1973. A case of orchitis caused by *Brucella abortus* in the African Buffalo. J. Wildl. Dis. 9, 251–253.
Kassahun, J., Yimer, E., Geyid, A., Abebe, P., Newayeselassie, B., Zewdie, B., Beyene, M., Bekele, A., 2006. Sero-prevalence of brucellosis in occupationally exposed people in Addis Ababa, Ethiopia. Ethiopian Med. J. 44, 245–252.
Le Fleche, P., Jacques, I., Grayon, M., Al-Dahouk, S., Bouchon, P., Denoeud, F., Nockler, K., Neubauer, H., Guilloteau, L.A., Vergnaud, G., 2006. Evaluation and selection of tandem repeat loci for a *Brucella* MLVA typing assay. BMC Microbiol. 6, 1–14.
Leal-Hernandez, M., Diaz-Aparicio, E., e rez, P.R., Hern a ndez-Andrade, L., Arellano-Reynoso, B., Alfonseca, E., Suarez-Guemes, F., 2005. Protection of *Brucella abortus* RB51 revaccinated cows, introduced in a herd with active brucellosis, with presence of atypical humoral response. Comp. Immunol. Microbiol. Infect. Dis. 28, 63–70.
Ledwaba, B., Mafofo, J., van Heerden, H., 2014. Genome Sequences of *Brucella abortus* and *Brucella suis* strains isolated from bovine in Zimbabwe. Genome Announcements 2, http://dx.doi.org/10.1128/genomeA.01063-14.
Lounes, N., Cherfa, M.-A., Le Carrou, G., Bouyoucef, A., Jay, M., Garin-Bastuji, B., Mick, V., 2014. Human Brucellosis in Maghreb: existence of a lineage related to socio-historical connections with Europe. PloS One 9, e115319, http://dx.doi.org/10.1371/journal.pone.0115319.
Luchsinger, D.W., Anderson, R.K., 1979. Longitudinal studies of naturally acquired *Brucella abortus* infection in sheep. Am. J. Vet. Res. 40, 1307–1312.
Luna-Martinez, J.E., Mejia-Teran, C., 2002. Brucellosis in Mexico: current status and trends. Vet. Microbiol. 90, 19–30.
Madsen, M., 1989. The current status of bovine brucelosis. Zimb. Vet. J. 20, 133–145.
Madsen, M., Anderson, E.C., 1995. Serologic survey of Zimbabwean wildlife for brucellosis. J. Zoo Wildl. Med., 240–245.
Magona, J.W., Walubengo, J., Galiwango, T., Etoori, A., 2009. Seroprevalence and potential risk of bovine brucellosis in zerograzing and pastoral dairy systems in Uganda. Trop. Anim. Health Prod. 41, 1765–1771, http://dx.doi.org/10.1007/s11250-009-9375-y.
Magwedere, K., Bishi, A., Tjipura-Zaire, G., Eberle, G., Hemberger, Y., Hoffman, L.C., Dziva, F., 2012. Brucellae through the food chain: the role of sheep, goats and springbok (*Antidorcus marsupialis*) as sources of human infections in Namibia. J. S. Afr. Vet. Assoc. 82, 205–212, http://dx.doi.org/10.4102/jsava.v82i4.75.
Mai, H.M., Irons, P.C., Kabir, J., Thompson, P.N., 2012. A large seroprevalence survey of brucellosis in cattle herds under diverse production systems in northern Nigeria. BMC Vet. Res. 8, 144.
Maichomo, M.W., McDermott, J.J., Arimi, S.M., Gathura, P.B., 1998. Assessment of the Rose-Bengal plate test for the diagnosis of human brucellosis in health facilities in Narok district, Kenya. East Afr. Med. J. 75, 219–222.
Mainar-Jaime, R.C., Marin, C.M., de Miguel, M.I.A.J., Mu n oz, P.M., Blasco, J.M., 2008. Experiences on the use of RB51 vaccine in Spain. In: Proceedings of the Brucellosis 2008, International Conference, Royal Holloway College, University of London, UK, pp. 10–13.
Makita, K., F e vre, E.M., Waiswa, C., Eisler, M.C., Thrusfield, M., Welburn, S.C., 2011. Herd prevalence of bovine brucellosis and analysis of risk factors in cattle in urban and peri-urban areas of the Kampala economic zone, Uganda. BMC Vet. Res. 7, 60, http://dx.doi.org/10.1186/1746-6148-7-60.
Mangen, M.J., Otte, J., Pfeiffer, D., Chilonda, P., 2002. Bovine Brucellosis in Sub-Saharan Africa: Estimation of Sero-Prevalence and Impact on Meat and Milk Offtake Potential. FAO, Livestock Information and Policy Branch, AGA.
Manthei, C.A., 1959. Summary of controlled research with strain 19. Proc. 63rd. Annu. Meet. U.S. Livestock Sanit. Assoc., 128.
Mantur, B.G., Amarnath, S.K., Patil, G.A., Desai, A.S., 2014. Clinical utility of a quantitative Rose Bengal slide agglutination test in the diagnosis of human brucellosis in an endemic region. Clin. Lab. 60, 533–541.
Mathew, C., Stokstad, M., Johansen, T.B., Klevar, S., Mdegela, R.H., Mwamengele, G., Michel, P., Escobar, L., Fretin, D., Godfroid, J., 2015. First isolation, identification, phenotypic and genotypic characterization of *Brucella abortus* biovar 3 from dairy cattle in Tanzania. BMC Vet. Res. 11 (1), http://dx.doi.org/10.1186/s12917-015-0476-8.
Matope, G., Bhebhe, E., Muma, J.B., Oloya, J., Madekurozwa, R.L., Lund, A., Skjerve, E., 2011. Seroprevalence of brucellosis and its associated risk factors in cattle from smallholder dairy farms in Zimbabwe. Trop. Anim. Health Prod. 43, 975–982.
McDermott, J.J., Arimi, S.M., 2002. Brucellosis in sub-Saharan Africa: epidemiology, control and impact. Vet. Microbiol. 90, 111–134, http://dx.doi.org/10.1016/s0378-1135(02) 00249-3.
McDermott, J.J., Grace, D., Zinsstag, J., 2013. Economics of brucellosis impact and control in low-income countries. Rev. Sci. Technol. 32, 249–261.
Megersa, B., Biffa, D., Abunna, F., Regassa, A., Godfroid, J., Skjerve, E., 2012. Seroepidemiological study of livestock brucellosis in a pastoral region. Epid. Infect. Infect. 140, 887–896, http://dx.doi.org/10.1017/s0950268811001178.
Megersa, B., Biffa, D., Abunna, F., Regassa, A., Godfroid, J., Skjerve, E., 2011a. Seroprevalence of brucellosis and its contribution to abortion in cattle, camel, and goat kept under pastoral management in Borana, Ethiopia. Trop. Anim. Health Prod. 43, 651–656, http://dx.doi.org/10.1007/s11250-010-9748-2.
Megersa, B., Biffa, D., Niguse, F., Rufael, T., Asmare, K., Skjerve, E., 2011b. Cattle brucellosis in traditional livestock husbandry practice in Southern and Eastern Ethiopia, and its zoonotic implication. Acta Vet. Scand. 53, 24, http://dx.doi.org/10.1186/1751-0147-53-24.
Megersa, B., Molla, B., Yigezu, L., 2006. Seroprevalence of brucellosis in camels (*Camelus dromedarius*) in Borena Lowland: Southern Ethiopia. Bull. Anim. Health Prod. Afr. 53, 252–257.
Mekonnen, H., Shewit, K., Kyule, M., 2010. Serological survey of bovine brucellosis in barka and arado breeds (*Bos indicus*) of western Tigray, Ethiopia. Prev. Vet. Med. 94, 28–35, http://dx.doi.org/10.1016/j.prevetmed.2009.12.001.
Mert, A., Ozaras, R., Tabak, F., Bilir, M., Yilmaz, M., Kurt, C., Ongoren, S., Tanriverdi, M., Ozturk, R., 2003. The sensitivity and specificity of *Brucella* agglutination tests. Diag. Microbiol. Infect. Dis. 46, 241–243, http://dx.doi.org/10.1016/s0732-8893(03) 00081-6.
Meyer, M.E., Morgan, W.J.B., 1973. Designation of neotype strains and of biotype reference strains for species of the genus *Brucella* Meyer and Shaw. Int. J. Syst. Bacteriol. 23, 135–141, http://dx.doi.org/10.1099/00207713-23-2-135.
Molla, B., Delil, F., 2015. Mapping of major diseases and devising prevention and control regimen to common diseases in cattle and shoats in Dassenech district of South Omo Zone, South-Western Ethiopia. Trop. Anim. Health Prod. 47, 45–51, http://dx.doi.org/10.1007/s11250-014-0681-7.
Moreno, E., 2014. Retrospective and prospective perspectives on zoonotic brucellosis. Front. Microbiol. 5 (213).
Moriyón, I., Grillo, M.J., Monreal, D., Gonzalez, D., Marin, C.M., L o pez-Go n i, I., Mainar-Jaime, R.U.L.C., Moreno, E., Blasco, J.M., 2004. Rough vaccines in animal brucellosis: structural and genetic basis and present status. Vet. Res. 35, 1–38, http://dx.doi.org/10.1051/vetres:2003037.
Motsi, T.R., Tichiwangana, S.C., Matope, G., Mukarati, N.L., 2013. A serological survey of brucellosis in wild ungulate species from five game parks in Zimbabwe. Onderstepoort J. Vet. Res. 80, 586, http://dx.doi.org/10.4102/ojvr.v80i1.586.
Muendo, E.N., Mbatha, P.M., Macharia, J., Abdoel, T.H., Janszen, P.V., Pastoor, R., Smits, H.L., 2012. Infection of cattle in Kenya with *Brucella abortus* biovar 3 and *Brucella melitensis* biovar 1 genotypes. Trop. Anim. Health Prod. 44, 17–20, http://dx.doi.org/10.1007/s11250-011-9899-9.
Mugizi, D.R., Boqvist, S., Nasinyama, G.W., Waiswa, C., Ikwap, K., Rock, K., Lindahl, E., Magnusson, U., Erume, J., 2015a. Prevalence of and factors associated with *Brucella* sero-positivity in cattle in urban and peri-urban Gulu and Soroti towns of Uganda. J. Vet. Med. Sci., http://dx.doi.org/10.1292/jvm14-0452.
Mugizi, D.R., Muradrasoli, S., Boqvist, S., Erume, J., Nasinyama, G.W., Waiswa, C., Mboowa, G., Klint, M., Magnusson, U., 2015b. Isolation and molecular characterization of *Brucella* isolates in cattle milk in Uganda. BioMed. Res. Int., 720413, http://dx.doi.org/10.1155/2015/720413.
Muma, J.B., Godfroid, J., Samui, K.L., Skjerve, E., 2007a. The role of *Brucella* infection in abortions among traditional cattle reared in proximity to wildlife on the Kafue flats of Zambia. Rev. Sci. Tech. 26, 721–730.
Muma, J.B., Samui, K.L., Oloya, J., Munyeme, M., Skjerve, E., 2007b. Risk factors for brucellosis in indigenous cattle reared in livestock-wildlife interface areas of Zambia. Prev. Vet. Med. 80, 306–317, http://dx.doi.org/10.1016/j.prevetmed.2007.03.003.
Muma, J.B., Lund, A., Siamudaala, V.M., Munang'andu, H.M., Munyeme, M., Matope, G., Nielsen, K.H., Djonne, B., Godfroid, J., Tryland, M., Skjerve, E., 2010. Serosurvey of *Brucella* spp. infection in the Kafue lechwe (*Kobus leche kafuensis*) of the Kafue flats in Zambia. J. Wildl. Dis. 46, 1063–1069.
Muma, J.B., Pandey, G.S., Munyeme, M., Mumba, C., Mkandawire, E., Chimana, H.M., 2012. Brucellosis among smallholder cattle farmers in Zambia: public health significance. Trop. Anim. Health Prod. 44, 915–920.
Muma, J.B., Samui, K.L., Siamudaala, V.M., Oloya, J., Matope, G., Omer, M.K., Munyeme, M., Mubita, C., Skjerve, E., 2006. Prevalence of antibodies to *Brucella* spp. and individual risk factors of infection in traditional cattle, goats and sheep reared in livestock-wildlife interface areas of Zambia. Trop. Anim. Health Prod. 38, 195–206.
Muriuki, S.M., McDermott, J.J., Arimi, S.M., Mugambi, J.T., Wamola, I.A., 1997. Criteria for better detection of brucellosis in the Narok District of Kenya. East Afr. Med. J. 74, 317–320.
Mutanda, L.N., 1998. Selected laboratory tests in febrile patients in Kampala, Uganda. East Afr. Med. J. 75, 68–72.

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023

Muhairwa, A.P., Mwakijungu, E.O., Msoffe, P., Mtambo, M., 2012. Seroprevalence and factors affecting canine monocytic ehrlichiosis and canine brucellosis in Tanzania. Res. Opin. Anim. Vet. Sci. 2, 181–188.
Nabukenya, I., Kaddu-Mulindwa, D., Nasinyama, G.W., 2013. Survey of *Brucella* infection and malaria among abattoir workers in Kampala and Mbarara Districts, Uganda. BMC Public Health 13, 901, http://dx.doi.org/10.1186/1471-2458-13-901.
Nicoletti, P., 1990. Vaccination. In: Nielsen, K.H., Duncan, J.R. (Eds.), Animal Brucellosis. CRC Press, Boca Raton, pp. 283–299.
Nicoletti, P., Jones, L.M., Berman, D.T., 1978. Comparison of the subcutaneous and conjunctival route of vaccination with *Brucella abortus* strain 19 vaccine in adult cattle. J. Am. Vet. Med. Assoc. 173, 1450–1456.
Nielsen, K.H., Smith, P., Yu, W., Nicoletti, P., Elzer, P.H., Robles, C., Bermudez, R., Renteria, T., Moreno, F.S., Ruiz, A., Massengill, C., Muenks, Q., Jurgersen, G., Tollersrud, T., Samartino, L.E., Conde, S., Forbes, L., Gall, D., Perez, B., Rojas, X., Minas, A., 2005. Towards single screening tests for brucellosis. Rev. Sci. Technol. 24, 1027–1037.
Nizeyimana, G., Mwiine, F.N., Ayebazibwe, C., 2013. Comparative *Brucella abortus* antibody prevalence in cattle under contrasting husbandry practices in Uganda. J. S. Afr. Vet. Assoc. 84, E1–5.
Ocampo-Sosa, A.A., Agüero, J., Garcia-Lobo, J.M., 2005. Development of a new PCR assay to identify *Brucella abortus* biovars 5, 6 and 9 and the new subgroup 3b of biovar 3. Vet. Microbiol. 110, 41–51, http://dx.doi.org/10.1016/j.vetmic.2005.06.007.
Ocholi, R.A., Bertu, W.J., Kwaga, J.K., Ajogi, I., Bale, O.O.J., Okpara, J., 2004a. Carpal bursitis associated with *Brucella abortus* in a horse in Nigeria. Vet. Rec. 155, 566–567.
Ocholi, R.A., Kwaga, J.K., Ajogi, I., Bale, O.O.J., 2004b. Phenotypic characterization of *Brucella* strains isolated from livestock in Nigeria. Vet. Microbiol. 103, 47–53.
Ocholi, R.A., Kwaga, J.K., Ajogi, I., Bale, O.O.J., 2005. Abortion due to *Brucella abortus* in sheep in Nigeria. Rev. Sci. Tech. 24, 973–979.
Oie, 2013a. Bovine brucellosis, In: Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. pp. 1–35.
Oie, 2013b. Caprine and ovine brucellosis (excluding *Brucella ovis*), In: Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. pp. 1–10.
Okoh, A.E., 1980. An investigation of abortion in sheep in Rano L.I.B.C. near Kano, Nigeria. Bull. Anim. Health Prod. Afr. 28, 135–138.
Oomen, L.J., 1976. Human brucellosis in Kenya. Trop. Geogr. Med. 28, 33–45.
Oomen, L.J., Waghela, S., 1974. The rose bengal plate test in human brucellosis. Trop. Geogr. Med. 26, 300–302.
Pappas, G., Papadimitriou, P., Akritidis, N., Christou, L., Tsianos, E.V., 2006. The new global map of human brucellosis. Lancet Infect. Dis. 6, 91–99.
Philpott, M., Auko, O., 1972. Caprine brucellosis in Kenya. Br. Vet. J. 128, 642–651.
Plommet, M., 1977. Studies on experimental brucellosis in cows in France. In: Crawford, R.P., Hidalgo, R.J. (Eds.), Bovine brucellosis. An International Symposium. Texas A\&M University Press, College Station and London, pp. 116–134.
Plommet, M., Fensterbank, R., 1976. Vaccination against bovine brucellosis with a low dose of strain 19 administered by the conjunctival route. III: Serological response and immunity in the pregnant cow. Ann. Rech. Vet. 7, 9–23.
Racloz, V., Schelling, E., Chitnis, N., Roth, F., 2013. Persistence of brucellosis in pastoral systems. Rev. Sci. Technol. 32, 61–70.
Ray, W.C., Brown, R.R., Stringfellow, D.A., Schnurrenberger, P.R., Scanlan, C.M., Swann, A.I., 1988. Bovine brucellosis: an investigation of latency in progeny of culture-positive cows. J. Am. Vet. Med. Assoc. 192, 182–186.
Refai, M., 2002. Incidence and control of brucellosis in the Near East region. Vet. Microbiol. 90, 81–110.
Reichel, R., 1996. *Brucella melitensis* biotype 1 outbreak in goats in northern KwaZulu-Natal. J. S. Afr. Vet. Assoc. 67, 2.
Roux, J., 1979. Epidémiologie et prévention de la brucellose. Bull. World Health Org. 57, 179–194.
Rubach, M.P., Halliday, J.E.B., Cleaveland, S., Crump, J.A., 2013. Brucellosis in low-income and middle-income countries. Curr. Opin. Infect. Dis. 26, 404–412.
Sachs, R., Staak, C., 1966. Evidence of brucellosis in antelopes of the Serengeti. Vet. Rec. 79, 857–858.
Sanogo, M., Abatih, E., Thys, E., Fretin, D., Berkvens, D., Saegerman, C., 2013. Importance of identification and typing of *Brucellae* from West African cattle: a review. Vet. Microbiol. 164, 202–211, http://dx.doi.org/10.1016/j.vetmic.2013.02.009.
Sanogo, M., Abatih, E., Thys, E., Fretin, D., Berkvens, D., Saegerman, C., 2012. Risk factors associated with brucellosis seropositivity among cattle in the central savannah-forest area of Ivory Coast. Prev. Vet. Med. 107, 51–56, http://dx.doi.org/10.1016/j.prevetmed.2012.05.010.
Schelling, E., Diguimbaye, C., Daoud, S., Nicolet, J., Boerlin, P., Tanner, M., Zinsstag, J., 2003. Brucellosis and Q-fever seroprevalences of nomadic pastoralists and their livestock in Chad. Prev. Vet. Med. 61, 279–293.
Schiemann, B., Staak, C., 1971. *Brucella melitensis* in impala (*Aepyceros melampus*). Vet. Rec. 88, 344.
Schurig, G., Sriranganathan, N., Corbel, M.J., 2002. Brucellosis vaccines: past, present and future. Vet. Microbiol. 90, 479–496.
Scolamacchia, F., Handel, I.G., Fevre, E.M., Morgan, K.L., Tanya, V.N., Bronsvoort, B.M., 2010. Serological patterns of brucellosis, leptospirosis and Q fever in *Bos indicus* cattle in Cameroon. PloS One 5, e8623.
Shaw, W.B., 1976. *Brucella abortus* infection in sheep. I. Field case. Br. Vet. J. 132, 18–27.
Smits, H.L., Abdoel, T.H., Solera, J., Clavijo, E., Diaz, R., 2003. Immunochromatographic *Brucella*–specific immunoglobulin M and G lateral flow assays for rapid serodiagnosis of human brucellosis. Clin. Diagn. Lab. Immunol. 10, 1141–1146.
Solera, J., 2010. Update on brucellosis: therapeutic challenges. Int. J. Antimicrob. Agents 36S, S18–S20, http://dx.doi.org/10.1016/j.ijantimicag.2010.06.015.
Spink, W.W., 1956. The diagnosis of human brucellosis. In: The Nature of Brucellosis. The University of Minnesota Press, Minneapolis, pp. 191–215.
Sprague, L.D., Al-Dahouk, S., Neubauer, H., 2012. A review on camel brucellosis: a zoonosis sustained by ignorance and indifference. Pathog. Glob. Health 106, 144–149.
Stableforth, A.W., Jones, L.M., 1963. Report of the Subcommittee on Taxonomy of the genus *Brucella*: speciation in the genus *Brucella*. Int. J. Syst. Bacteriol. 13, 145–158.
Swai, E.S., Schoonman, L., 2008. Human brucellosis: seroprevalence and risk factors related to high risk occupational groups in Tanga municipality, Tanzania. Zoonoses Public Health 59, 193–201.
Tanner, M., Inlameia, O., Michel, A., Maxlhuza, G., Pondja, A., Fafetine, J., Macucule, B., Zacarias, M., Manguele, J., Moiane, I.C., Marranangumbe, A.S., Mulandane, F., Schönfeld, C., Moser, I., Van Helden, P., Machado, A., 2014. Bovine tuberculosis and brucellosis in cattle and African buffalo in the Limpopo National Park, Mozambique. Transbound. Emerg. Dis., http://dx.doi.org/10.1111/tbed.12210.
Teklue, T., Tolosa, T., Tuli, G., Beyene, B., Hailu, B., 2013. Sero-prevalence and risk factors study of brucellosis in small ruminants in Southern Zone of Tigray Region, Northern Ethiopia. Trop. Anim. Health Prod. 45, 1809–1815, http://dx.doi.org/10.1007/s11250-013-0439-7.
Tesfaye, G., Tsegaye, W., Chanie, M., Abinet, F., 2011. Seroprevalence and associated risk factors of bovine brucellosis in Addis Ababa dairy farms. Trop. Anim. Health Prod. 43, 1001–1005, http://dx.doi.org/10.1007/s11250-011-9798-0.
Thimm, B., Wundt, W., 1976. The epidemiological situation of brucellosis in Africa. Dev. Biol. Stand. 31, 201–217.
Thornton, H., 1976. Unusual pathological conditions: Brucella hygromas in an eland (*Tragelapus oryx*). Rhod. Vet. J. 7, 64.
Thrusfield, M., 2013. Veterinary Epidemiology, 3rd ed. Blackwell Science, Oxford.
Tounkara, K., Maiga, S., Traore, A., Seck, B.M., Akakpo, A.J., 1994. Epidémiologie de la bnrucellose bovine au Mali: enquête sérologique et isolement des premières souches de *Brucella abortus*. Rev. Sci. Technol. 13, 777–786.
Treanor, J.J., 2013. Integrating ecology with management to control wildlife brucellosis. Rev. Sci. Technol. 32, 239–247.
Tschopp, R., Abera, B., Sourou, S.Y., Guerne-Bleich, E., Aseffa, A., Wubete, A., Zinsstag, J., Young, D., 2013. Bovine tuberculosis and brucellosis prevalence in cattle from selected milk cooperatives in Arsi zone, Oromia region, Ethiopia. BMC Vet. Res. 9, 163, http://dx.doi.org/10.1186/1746-6148-9-163.
Van Rensburg, S.W., Van Heerden, K.M., Le Roux, D.J., Snyders, A.J., van Heerden, K.M., 1958. Infectious infertility in sheep. J. S. Afr. Vet. Assoc. 29, 223–233.
Verger, J.M., Garin-Bastuji, B., Grayon, M., Mahe, A.M., 1989. La brucellose bovine à *Brucella melitensis* en France. Ann. Rech. Vet. 20, 93–102.
Verger, J.M., Grayon, M., 1984. Charactéristiques de 273 souches de *Brucella abortus* d'origin Africaine. Dev. Biol. Stand. 56, 63–71.
Verger, J.M., Grayon, M., Zundel, E., Lechopier, P., Olivier-Bernardin, V., 1995. Comparison of the efficacy of *Brucella suis* strain 2 and *Brucella melitensis* Rev. 1 live vaccines against a *Brucella melitensis* experimental infection in pregnant ewes. Vaccine 13, 191–196.
Waghela, S., Karstad, L., 1986. Antibodies to *Brucella* spp. among blue wildebeest and African buffalo in Kenya. J. Wildl. Dis. 22, 189–192.
Wheat, P.F., Dabbs, D.J., Thickett, K.J., 1995. *Brucella melitensis*: an unexpected isolate from cerebrospinal fluid. Commun. Dis. Rep. 5, R56–R57.
Xin, X., 1986. Orally administrable brucellosis vaccine: *Brucella suis* strain 2 vaccine. Vaccine 4, 212–216.
Yohannes, M., Degefu, H., Tolosa, T., Belihu, K., Cutler, R., Cutler, S., 2013. Brucellosis in Ethiopia. Afr. J. Microbiol. Res. 7, 1150–1157.
Young, E.J., 1989. Clinical manifestations of human brucellosis. In: Young, E.J., Corbel, M.J. (Eds.), Brucellosis: Clinical and Laboratory Aspects. CRC Press Inc., Boca Raton, Florida, pp. 98–126.
Yu, W.L., Nielsen, K., 2010. Review of detection of *Brucella* spp. by polymerase chain reaction. Croat. Med. J. 51, 306–313, http://dx.doi.org/10.3325/cmj.2010.51.306.
Zheludkov, M.M., Tsirelson, L.E., 2010. Reservoirs of *Brucella* infection in nature. Biol. Bull. 37, 709–715.

Please cite this article in press as: Ducrotoy, M., et al., Brucellosis in Sub-Saharan Africa: Current challenges for management, diagnosis and control. Acta Trop. (2015), http://dx.doi.org/10.1016/j.actatropica.2015.10.023