

1 **Full title:** Contribution of livestock marketing chains and role played by stakeholders'  
2 knowledge, attitude and practice in spreading cystic hydatidosis to Busia Town, Kenya, 2018.

3 **Short title:** Cystic hydatidosis in cattle

4

5 **Authors;** Henry Joash Ogutu<sup>1, \*</sup>, Maurice Owiny<sup>1</sup>, Bernard Bett<sup>3</sup>, Christina Otieno<sup>2</sup>

6 **Affiliations;**

7 <sup>1</sup>Kenya Field Epidemiology and Laboratory Training Program, Nairobi, Kenya

8

9 <sup>2</sup>School of Public Health, Moi University, Eldoret, Kenya

10

11 <sup>3</sup>International Livestock Research Institute, Nairobi, Kenya

12

13 **\*Corresponding author:**

14 Email: [ojoash2015@gmail.com](mailto:ojoash2015@gmail.com) (HJO)

15

16

17

18

19

20

21

22

23

24

## 25 **Abstract**

### 26 **Background:**

27 Cystic hydatidosis (CH), a neglected parasitic zoonosis, is endemic in many parts of Kenya  
28 and could be spread along livestock marketing chains. Poor knowledge, attitude and practices  
29 (KAP) enables this spread in remote areas with inadequate public health services. We  
30 estimated prevalence, identified possible origin of CH to Busia, Kenya and assessed KAP  
31 among cattle owners and abattoir workers.

### 32 **Methods and Principal Findings:**

33 We conducted a cross-sectional study on slaughtered livestock and interviewed their owners and  
34 abattoir workers in Busia in May–June 2018. We used visual observation, palpation and incision  
35 to identify cysts. Polymerase chain reaction (PCR) was used for confirmatory diagnosis. Epi  
36 Info 7 was used to calculate descriptive and associative statistics. Of 302 carcasses inspected,  
37 cysts were visualized in nine (2.98%, 95% Confidence Interval (CI): 1.46–5.78). Fourteen  
38 samples were collected and 13 (92.86%) were positive on PCR (sensitivity=92%,  
39 specificity=95%). All carcasses with cysts were from West Pokot County, which borders Busia  
40 to the north. We interviewed 310 participants: 260 were males (83.87%, 95% CI: 79.19 – 87.69);  
41 median age was 41 years (range=21-69). Dogs were kept by 221 (71.99%, 95% CI: 66.55 –  
42 76.87), of which 83 (37.56%, 95% CI: 28.33 – 48.52) improperly disposed of dog faeces. Home  
43 slaughtering was practiced by 196 (63.23%, 95% CI: 58.78-69.80), of which 115 (58.67%, 95%  
44 CI: 51.44-65.64) were not inspected and 85 (43.37%, 95% CI: 36.32-50.62) fed raw organs to  
45 dogs. Adequate knowledge was associated with butcher ownership (P-value = 0.002), age  $\geq$ 35  
46 years (P-value = 0.002) and higher literacy level (P-value <0.001).

### 47 **Conclusions and Significance:**

48 There is non-negligible risk of CH in Busia communities which might worsen with time given  
49 that the county is connected to endemic areas through livestock trade. Poor KAP by the people  
50 on the disease calls for need to implement information, education and communication campaigns  
51 to improve KAP on CH in the area.

52

53 **Key words:** Cystic hydatidosis, cattle marketing chains, KAP, Busia-Kenya.

54

**55 Author summary**

56 Cystic hydatidosis is a globally neglected parasitic zoonosis which is endemic in many parts of  
57 the world including Kenya. It is majorly a problem among pastoral communities where there is  
58 close contact between human, livestock and dogs. Busia County, in Western Kenya is part of a  
59 livestock marketing chain between Kenya and Uganda. Animals from high endemic regions in  
60 Uganda and Kenya can easily spread the parasite to Busia through improper disposal of their  
61 infested organs. Non-pastoral communities like Busia may not have much cumulative experience  
62 about the disease though their practices may contribute to the perpetuation of the parasite in their  
63 environment. The parasite is gradually spreading to new areas and it is very important to the  
64 public health players in Kenya to take action so as to prevent further spread of this disease.  
65 Findings from this study show that the disease is no longer limited to pastoral communities only.  
66 There is need for the implementation of information, education and communication campaigns to  
67 improve the knowledge, attitude and practices of Busia community and other non-endemic  
68 regions on the disease.

69  
70  
71  
72  
73  
74  
75  
76  
77  
78  
79  
80  
81  
82  
83  
84  
85  
86  
87  
88  
89  
90  
91  
92  
93  
94  
95  
96

## 97 **Introduction**

98 Hydatidosis is a neglected parasitic zoonotic disease caused by larval stage of *Echinococcus* and  
99 affects mostly dogs, livestock and humans [1,2]. The parasite has four species, but only two are  
100 of public health importance; *E. granulosus* which causes cystic hydatidosis (CH), commonly  
101 occurs in tropical regions and infests ungulates as its prime intermediate host hence its  
102 significance in livestock and *E. multilocularis*, which causes alveolar hydatidosis (AH) and  
103 occurs in the temperate regions [3]. Others are *E. vogeli*, and *E. oligarthrus* [3,4]. Hydatidosis  
104 has been reported in Europe and south eastern Australia. It is endemic in China, Indian  
105 Subcontinent and Middle East and re-emerging in the former Soviet Republics. In Africa *E.*  
106 *granulosus* is a particular problem in Northern and Eastern Africa countries including Kenya [2].

107         Hydatidosis affects 2–3 million people worldwide in extensive livestock farming areas. In  
108 2014 the WHO estimated that it caused more than 3,000 human and animal deaths. These deaths  
109 contributed to economic losses estimated at three billion United States Dollars (USD) covering  
110 costs on interventions, livestock organ condemnation and reduced livestock productivity.  
111 Livestock related losses in Kenya are estimated at more than 240,000 USD annually [5].

112         Busia, a border town in Kenya, is a major livestock market for traders in Kenya and  
113 Uganda. There are fears that the parasite is being introduced into the county via livestock  
114 marketing chains by improper disposal of infested organs [5]. Previous studies on the disease  
115 have put less focus on the risks of spread via livestock trade that connect high and low endemic  
116 areas. Findings from such studies could aid public health actors in formulating evidence-based  
117 prevention and control policies for this disease. This study estimated prevalence of CH in cattle  
118 slaughtered at Busia abattoirs, identified possible origin of CH to Busia and assessed knowledge,  
119 attitude and practices (KAP) on hydatidosis among livestock owners and people working in  
120 slaughter houses in Busia, Kenya.

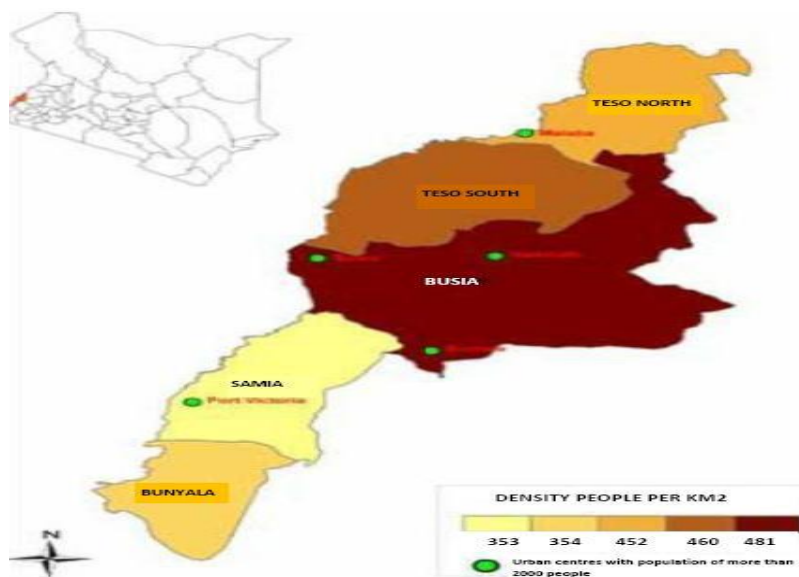
## 121 **Methods**

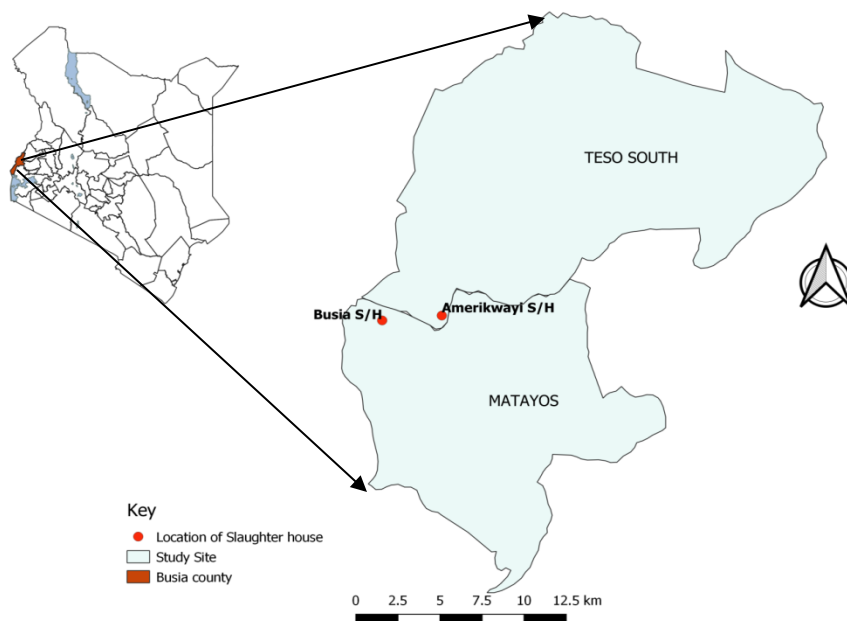
### 122 **Study design**

123 We conducted a cross-sectional study on cattle carcasses and interviewed abattoir workers as  
124 well as people who presented their cattle for slaughter on their knowledge attitude and practices  
125 on hydatidosis. The study was conducted in two busy abattoirs in Busia Town; Busia Municipal  
126 and Amerikwai abattoirs form May to June 2018. Busia Town is at the border of Kenya and  
127 Uganda in Busia County (Figure 1). It has a total human population of 111,345 and livestock  
128 population of 215,871 out of which cattle are 132,804 [6]. The main economic activity in Busia  
129 town is trade with neighboring Uganda, but crop and livestock farming is also done in small  
130 scale in the outskirts of the town. We included male and female cattle of all age categories  
131 destined for slaughter at the two abattoirs. All carcasses, whose owners consented, were eligible  
132 for inspection. Consenting adult cattle owners and abattoir workers were assessed on their KAP  
133 on hydatidosis. Carcasses condemned for other infections like emphysema, liver flukes and  
134 tuberculosis were not eligible for consideration into the study. Cattle whose owners could not be  
135 reached or traced for interviewing were not eligible.

136

137





138

139 **Fig 1: Map of study area. Brown rectangle in Kenyan map represent the location of Busia**  
 140 **in relation to neighboring Uganda, Red circle represent location of Busia Municipal and**  
 141 **Amerikwai abattoirs. This figure was created for this manuscript in QGIS using open source**  
 142 *data from ESRI and GPS points collected during data collection in the field.*

### 143 **Sample size estimation and assumptions**

144 A minimum sample size of 294 was calculated using the Cochran formula with the following  
 145 assumptions; *a priori* prevalence of 25.8 % (1), 95 % as the level of confidence, 5% desired  
 146 absolute precision and 10% anticipated non-response rate. The sample size was proportionately  
 147 divided between the two abattoirs based on average monthly slaughter figures as per the monthly  
 148 meat hygiene reports over a period of three years.

### 149 **Variables and measurements**

150 Among the variables collected at animal level included sex, estimated age, breed and origin of  
 151 each animal. We used questionnaires to collect data on whether or not the respondent had gone  
 152 through training on CH, knowledge on the mode of transmission and control measures for the

153 disease, importance of deworming dogs and livestock to control hydatidosis, slaughtering  
154 animals at home, keeping dogs at home, feeding dogs on raw meat, disposal of infected organs  
155 and dog feces at home.

#### 156 **Data collection**

157 We conducted ante-mortem examinations of all study cattle to estimate age by dentition [7,8],  
158 identify breed, determine sex and record the origin of each animal. The origin of each animal  
159 was confirmed from the animal movement permits and 'no objection' forms filed in the Busia  
160 veterinary offices and also from interviews held with livestock traders and farmers who brought  
161 their animals for slaughter. Post-mortem inspections were conducted immediately after  
162 slaughtering, skinning and evisceration. Standard morphological meat inspection procedures  
163 including visual observation, palpation and systematic incision by making deep longitudinal cuts  
164 in organs and muscles. according to Kenya Meat Control Act , CAP 356, were used to determine  
165 infestation status of each carcass. Number of cysts per organ were counted and recorded and  
166 cysts were removed whole and put in zipped polythene bags in cool boxes with icepacks for  
167 conventional polymerase chain reaction (PCR) tests at the International Livestock Research  
168 Institute (ILRI) field laboratory in Busia. Each sample was appropriately labelled for ease of  
169 traceability in the laboratory.

#### 170 **Laboratory methods**

171 The samples were gradually frozen to -20°C, kept for one month then processed and stored in  
172 70% ethanol. For DNA extraction, up to 20 mg of tissue samples was excised and placed in a  
173 nuclease-free microfuge tube. We added 300 microliters ( $\mu\text{L}$ ) of digestion buffer A to the tissue  
174 and 12  $\mu\text{L}$  of proteinase K and left to incubate at 55°C for 1.5 hours. We then added 300  $\mu\text{L}$  of  
175 buffer SK to the lysate and mixed by vortexing and then added 300  $\mu\text{L}$  of 100% ethanol. A micro

176 spin column with a provided collection tube was assembled and up to 600  $\mu$ L of the mixture was  
177 applied to the spin column assembly. The unit was capped and centrifuged for three minutes at  
178 8,000 rotations per minute (RPM). After centrifugation, we discarded the flow-through and  
179 reassembled the spin column with its collection tube. This was repeated until all the lysate had  
180 passed through the column. To wash the bound DNA, we applied 500  $\mu$ L of wash solution A to  
181 the column and centrifuged the unit for one minute at 14000 RPM. After centrifugation, we  
182 discarded the flow-through and reassembled the spin column with its collection tube. We applied  
183 500  $\mu$ L of wash solution A to the column and centrifuged the unit for two minutes at 14000RPM.  
184 The spin column was detached from the collection tube and discarded the collection tube and  
185 flow-through. We assembled the spin column with DNA bound to the resin with a provided 1.7  
186 mL elution tube. Two hundred microliters of Elution Buffer B was added to the center of the  
187 resin bed then allowed to stand for 10 minutes. It was then centrifuged for one minute at  
188 6000RPM. A portion of Elution Buffer B passed through the column which allowed for the  
189 hydration of the DNA to occur. We again centrifuged at 14000RPM for an additional two  
190 minutes to collect the total elution volume. The purified genomic DNA was stored at -20°C for  
191 one day to await PCR process.

#### 192 Identification of *nad5* gene

193 Conventional PCR was carried out on all the 14 hydatid cysts DNA isolated. The PCR primers  
194 Mit-F/Mit-R were used to amplify a 562 bp fragment of the mitochondrion the NADH-  
195 Ubiquinone oxidoreductase (complex I), chain 5 N-terminus (*nad5* gene) of *E. granulosus* (Gen  
196 Bank accession No. ARO49807). The PCR amplification reactions containing 3  $\mu$ L mtDNA, 0.5  
197  $\mu$ L each of the forward and reverse primers (this study), and 12.5  $\mu$ L of *Taq* PCR Master Mix  
198 (Qiagen) in a final reaction volume of 28  $\mu$ L. After denaturation at 95°C for 10 min,

199 amplification cycles were performed for four-stage, 25 cycles of 95°C for 30 seconds (s), 58°C  
200 for 30 s, and 72°C for 30 s for seven cycles in stage one, 95°C for 30 s, 56°C for 30 s, and 72°C  
201 for 30 s for seven cycles in stage two, 95°C for 30 s, 55°C for 30 s, and 72°C for 30 s for seven  
202 cycles in stage three, 95°C for 30 s, 54°C for 30 s, and 72°C for 30 s for four cycles in stage four,  
203 followed by 72°C for 10 min and cooling to 10°C. PCR products were loaded on 1.2% (w/v) Hi-  
204 Standard Agarose gel (AGTC Bio-products Limited, Hessle, UK) in 1X Tris-Boric-EDTA and  
205 stained with 0.5 µg/ml Safe White Nucleic Acid Stain (NBS Biologicals, Cambridge-shire, UK).  
206 Electrophoresis was carried out for 40 min at 190 V. The bands were visualized in UV trans-  
207 illuminator and digitally photographed.

#### 208 **Knowledge, attitude and practices**

209 We used a pre-tested questionnaire with open ended and closed-questions for interviews. The  
210 interviews were conducted in a separate room within the abattoir compounds to maintain  
211 confidentiality.

#### 212 **Statistical methods**

213 The collected data were entered, cleaned and analyzed using Epi Info™ 7.1.4.0 (CDC, Atlanta,  
214 GA, USA). Measures of central tendency and dispersion for continuous variables and  
215 frequencies, proportions and 95% CI for categorical variables were calculated. We assigned  
216 scores to knowledge questions in the questionnaire. A correct response earned a score of one (1)  
217 while an incorrect or “*I don't know*” response scored a zero (0). Adequate knowledge was  
218 considered as a total score above or equal to half ( $\geq 5$ ) of the overall score (10). We used  
219 bivariate and logistic regression to examine the factors associated with adequate knowledge  
220 among the study participants. From the bivariate analysis, variables that had P-values of  $\leq 0.1$   
221 were entered into a multivariate regression model. The final model was arrived at using

222 backward stepwise elimination method where variables with P-values  $\leq 0.05$  were considered to  
223 have statistical associations with adequate knowledge, as dependent coefficient. The participants  
224 who considered keeping dogs and livestock in same homestead to increase the risk of hydatidosis  
225 were considered to have a good attitude. Regular deworming of livestock and dogs, good meat  
226 hygiene and proper disposal of infected organs and dog faeces were considered as good  
227 practices.

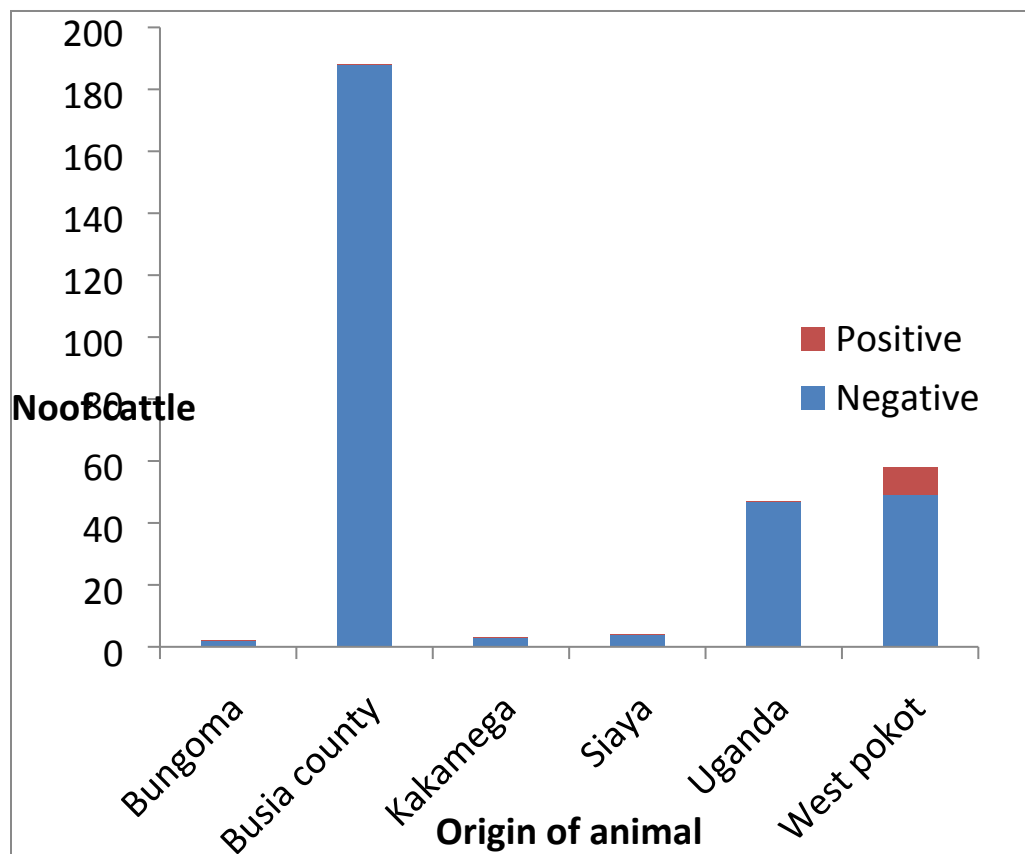
## 228 **Ethical issues**

229 We obtained written permission from Busia County Director of Veterinary Services (CDVS) to  
230 access the abattoirs and sought verbal consent from abattoir managers to access abattoir  
231 compounds. Participation by cattle owners was sought through signing informed consent. Ethical  
232 approval for this study was obtained from institutional research and ethics committee (IREC) of  
233 Moi University (IREC No. 0001850).

## 234 **Results**

### 235 **Descriptive statistics**

236 In the study, a total of 302 cattle carcasses were inspected. Of these, 188 (62.25%) originated  
237 from Busia County while others originated from Uganda and other parts of Kenya (Figure 2).  
238 Local breeds of cattle comprised 295 (97.68%, 95% Confidence Interval (CI): 95.08–98.98). The  
239 inspected carcasses consisted of 222 (73.51%, 95% CI: 68.09 – 78.32) male cattle.



240

241 **Fig 2: Distribution of source of cattle by county and their infection status, Busia town**  
 242 **abattoirs 2018 (n=302).**

243 Majority of the carcasses inspected, 144 (47.68%) were aged between 4–6 years and 18  
 244 (5.96%, 95% CI: 3.67–9.42) were aged above nine years. Hydatid cysts were visualized in nine  
 245 (2.98%) of the inspected carcasses. Among these, eight were female (Table 1).

246

247

248

249

250

251 **Table 1: Proportions of cattle by age, gender and infection status at Busia town abattoirs**  
 252 **2018 (n=302)**

Age grp	Male		Total male	Female		Total female	Total carcasses
	Positive	Negative		Positive	Negative		
1-3	0	13	13	0	5	5	<b>18</b>
4-6	0	101	101	2	41	43	<b>144</b>
7-9	0	93	93	6	23	29	<b>122</b>
>9	1	14	15	0	3	3	<b>18</b>

253  
 254 Out of the nine carcasses with cysts, five (55.56%) had multiple organ infestations. The  
 255 main infested organs were liver (n=7) and lung (n=4). The total number of samples collected for  
 256 PCR confirmation was 14 out of which 13 (92.86%) samples turned positive on PCR test  
 257 (sensitivity = 92% and specificity = 95%). From the laboratory results, the samples whose bands  
 258 were visualized in ultra violet (UV) trans-illuminator were interpreted as positive on the PCR  
 259 test. One sample calcified during preservation and therefore the DNA could not be extracted  
 260 from it for PCR testing. All the positive carcasses were animals from West Pokot County.

### 261 **Knowledge, attitude and practices**

262 We interviewed 310 cattle owners, of whom 260 (83.87%, 95% CI: 79.19 – 87.69) were male.  
 263 The overall median age was 41 years (range 21-69 years). Livestock farmers comprised 158  
 264 (50.97%, 95% CI: 45.27 – 56.65) of the study participants (Table 2). When asked about their  
 265 level of education, 116 (37.42%, 95% CI: 32.06 – 43.06) said that they had completed primary  
 266 education, 92 (29.67%, 95% CI: 24.71 – 35.15) had completed secondary education and 58  
 267 (18.71%, 95% CI: 14.62 – 23.60) did not have any formal education (Table 2).

268 **Table 2: Literacy level of participants and their role in the value chain, Busia town**  
 269 **abattoirs, 2018**

Factor	Frequency	Percentage
<b>Level of education (n=310)</b>		
None	58	18.70
Primary completed	116	37.42
Secondary completed	92	29.68
Tertiary completed	44	14.20
<b>Role in value chain (n=310)</b>		
Livestock farmer	158	50.97
Butchery owner	65	20.97
Livestock trader	39	12.58
Butchery attendant	21	6.77
Butcher man/flayer	17	5.48
Meat inspector	5	1.61
Intern/student	3	0.97
Abattoir cleaner	2	0.65

270

271 On knowledge of cystic hydatidosis, 197 (63.55%, 95% CI: 57.89 – 68.86) participants  
 272 said that they had heard about the disease. The participants who knew the role of dogs in  
 273 transmission of the disease were 53 (17.10%, 95% CI: 13.37 – 23.84). The effects of hydatidosis  
 274 on livestock were known to 175 (56.45%) of the respondents, out of which 161 (92.00%) were  
 275 butchery owners. On average, the participants who scored  $\geq 5$  were 40 (12.90%).

276           When we assessed the attitude of the participants on the disease, 162 (54.00%) disagreed  
277 that there is a risk of hydatid disease transmission to livestock or humans by having a dog on the  
278 same compound with livestock; 91 (30.33%) agreed, 32 (10.67%) strongly agreed and 15 (5%)  
279 strongly disagreed to the question. Among the participants who answered the question regarding  
280 the importance of deworming dogs to control the disease, 130 (43.33%) disagreed, 119 (39.67%)  
281 agreed, 43 (14.33%) strongly agreed and eight (2.67%) strongly disagreed. The participants who  
282 disagreed that disposing or condemning infected organs was a waste of food were 159 (51.96%),  
283 while 95 (31.05%) strongly disagreed, 42 (13.73%) agreed, and 10 (3.27%) strongly agreed.  
284 Those who agreed that keeping their livestock dewormed and clean was a reflection of their  
285 social status were 177 (57.65%), while 88 (28.66%) strongly agreed, 42 (13.68%) disagreed, and  
286 none strongly disagreed. A good attitude towards the disease was held by 123 (39.68%).

287           Assessment of study participants on their practice on the disease revealed that 256  
288 (85.62%, 95% CI: 81.12 – 89.39) dewormed their livestock and 124 (48.44%, 95% CI: 42.17 –  
289 54.74) of them dewormed their livestock after every 3 months. Dogs were kept at home by 221  
290 (71.29%, 95% CI: 66.55 – 76.87) participants. Among the dog keepers, 93 (42.08%, 95% CI:  
291 35.49 – 48.89) dewormed their dogs and 37 (39.78%, 95% CI: 29.78 – 50.46) dewormed at an  
292 interval of three months. Methods of disposing dog faeces included burying, 95 (42.99%, 95%  
293 CI: 36.37 – 49.80), doing nothing and open disposal, 83 (37.56%, 95% CI: 28.33 – 48.52).  
294 Feeding their dogs on raw meat was admitted by 120 (54.30%, 95% CI: 47.48 – 61.00) dog  
295 keepers, while 196 (63.23%, 95% CI: 58.78 – 69.80) cattle owners admitted that they sometimes  
296 slaughtered animals at home. However, 115 (58.67%, 95% CI: 51.44 – 65.64) of the meat  
297 slaughtered at home was not inspected by qualified meat inspectors and 85 (43.37%, 95% CI:  
298 36.32 – 50.62) of raw organs of animals slaughtered at home were fed to dogs. At bivariate

299 analysis, religion ( $P<0.0447$ ), gender ( $P<0.0354$ ), age ( $P<0.0046$ ), education (0.0012) and  
 300 occupation (0.0085) had a statistical association with adequate knowledge (Table 3).

301 **Table 3: Bivariate analysis with knowledge as a coefficient of other variables**

Variable	OR (95% CI)	P value
Age (> 35 years)	88.44 (51.79 – 945.74)	0.0046
Education (primary or less)	56.13 (40.33 – 194.67)	0.0012
Gender	83.87 (0.87 – 112.44)	0.0354
Marital status (single)	8.71 (2.45 – 44.12)	6.5437
Occupation (butchery owner)	51.94 (33.56 – 98.89)	0.0085
Religion (none)	0.97 (0.56 – 2.44)	0.0447

302 **OR: Odds Ratio, CI: Confidence Interval**

303 On multivariate analysis, occupation (being a butchery owner) ( $P<0.002$ ), age above 35 years  
 304 ( $P<0.002$ ) and literacy level ( $P<0.001$ ) were independently associated with adequate knowledge  
 305 on CH (Table 4).

306 **Table 4: Multivariate logistic regression with knowledge as random effect variable to age,**  
 307 **level of education and occupation**

Coefficient	AOR (95% CI)	Standard error	P value
Age (> 35 years)	73.43 (21.71 – 1342.72)	1.0216	0.002
Education (primary or less)	56.13 (40.33 – 194.67)	0.27990	0.001
Gender	83.87 (0.87 – 112.44)	1.0293	1.581
Marital status (single)	8.71 (2.45 – 44.12)	1.0319	4.056
Occupation(butchery owner)	0.47 (0.12 – 0.86)	0.4361	0.002

Religion (none)	0.97 (0.56 – 2.44)	0.4361	0.223
-----------------	--------------------	--------	-------

308 *AOR: Adjusted Odds Ration, CI: Confidence Interval*

### 309 **Discussion**

310 The study found that there is a risk of cystic hydatidosis spreading to Busia town via cattle trade  
 311 from endemic areas like West Pokot County. Some of these cattle slaughtered in Busia Town  
 312 originate from Uganda and other parts of Kenya that are infested with the parasite. In Busia, the  
 313 parasite may spread further through improper disposal of infested organs. The study identified  
 314 gaps in the participants' knowledge, attitude and practice regarding awareness on CH, risks of  
 315 keeping dogs on same compound with livestock, practicing poor meat hygiene, improper  
 316 disposal of dog faeces and infested organs of animals slaughtered at home in absence of qualified  
 317 meat inspectors and not deworming of their dogs and livestock regularly. These gaps on KAP  
 318 can increase the risk of infestation with the parasite.

319 Most of the cattle slaughtered at Busia town abattoirs during the study period came from  
 320 within Busia County. Male cattle formed higher percentage of the cattle slaughter figures than  
 321 females because they grow faster are heavier in weight and attain their mature weight earlier than  
 322 females. Thus males give higher returns in profit after sale of their carcasses as butchery owners  
 323 buy cattle for slaughter based on weight and body size. The local breeds of cattle (zebu), being  
 324 the majority of cattle in Busia County [6], have slow growth rate and get their optimum weight at  
 325 an average age of four years [9]. This could explains why most of the slaughtered cattle at the  
 326 two abattoirs were between 4-6 years old.

327 The reported prevalence of 3% in this study shows the extent to which the infestation can  
 328 easily spread over time from its known endemic areas. The possibility of spread is very  
 329 important to the public health stakeholders in the country to take action so as to avoid the spread

330 of this disease to non-endemic regions in Busia [5,10]. The trend of spread over time should be a  
331 warning to Kenya's public health players that the disease is no longer a problem to pastoral  
332 communities only. The study findings confirmed that the infestation rates of cattle increased  
333 significantly with age [11,12]. This study revealed that liver was the main infested tissue; a  
334 similar finding was seen in studies done in Kermanshah province, west of Iran and in slaughter  
335 houses in Maasailand and Turkana in Kenya [10,13]. Other studies have found that lungs are the  
336 main infested tissues [11,12,14]. Other findings in this study are that female cattle are more  
337 likely to be infested with CH than males, similar to a study done in Tabriz area, Northwest of  
338 Iran [12], a study done in Central Ethiopia [11] and a study done in Libya [15]. The higher  
339 prevalence in female cattle may be correlated to the fact that the females are kept for  
340 reproductive purpose hence they live for longer periods while most male cattle are slaughtered at  
341 an early age. However, both male and female are at risk of contracting the disease [5].

342         The findings of the KAP survey showed that the beef value chain in the study area was  
343 dominated by men, who have culturally been cattle traders and they tend to dominate livelihood  
344 activities that generate financial income as was seen in studies done in Uganda [16] and Pakistan  
345 [7]. Most of the participants, especially farmers, did not have much knowledge about  
346 hydatidosis. Among those who knew the effects of CH, the majority were butchery owners as  
347 they were more familiar with the direct losses due to condemnation of infested organs and  
348 carcasses [7]. Results from this study showed that non-pastoral communities like those found in  
349 Busia Kenya are unfamiliar with CH. Participants above the age of 35 years were more aware of  
350 the disease than younger people. The statistical association between age and knowledge on  
351 hydatid disease could be due to the cumulative experience and insights about the disease that  
352 accrues with age [17].”

353 A large number of the participants disagreed that there is a risk of transmission of the  
354 disease by keeping a dog on the same compound with the livestock. A majority of them also did  
355 not find it important to deworm dogs as a control measure for hydatidosis, though more than half  
356 of the participants agreed that deworming livestock and keeping them clean is a true reflection of  
357 someone's social status. There was poor attitude on the disease by participants, which may be  
358 contributed to by low literacy level as more than half of them had primary education and below.  
359 These barriers related to knowledge and information could hamper the effectiveness of  
360 interventions in prevention and control of CH [18]".

361 Approximately half of respondents dewormed their livestock; however, more than a  
362 quarter of them did not have a regular deworming interval for their livestock. The findings on the  
363 number of dog keepers who dewormed their dogs is consistent with a study done in Uganda [16],  
364 but was in contrast to studies done in Ethiopia where dog keepers were 71% of the study  
365 participants and none of them dewormed their dogs [19] and in Pakistan where dog keepers were  
366 64% and 68% of them dewormed their dogs [7]. The dog owners did not know the risk contained  
367 in improper disposal or inappropriate handling of dog faeces in terms of transmission of CH.  
368 This makes controlling the disease difficult being that the dog faeces, with infective larvae of the  
369 parasite, contaminates the environment hence exposing the livestock and human population to  
370 risk of infestation. Control of cystic hydatidosis is less effective without the support of dog-  
371 owners, and this support can only be obtained if the people have a clear understanding of the life  
372 cycle of the parasite and of risk factors for human and livestock infestations [20].

373 We noted that slaughtering animals at home was a common practice by respondents, but  
374 qualified meat inspectors were rarely contacted to inspect such carcasses. Failure to call a  
375 qualified meat inspector to inspect meat slaughtered at home leads not only to improper disposal

376 of infected organs and carcasses, but also risks transmission of other zoonotic diseases to  
377 humans. This observation was also made in a study conducted among pastoral communities in  
378 Greater Kapoeta of South Sudan [21]. Infested organs and carcasses of cattle slaughtered at  
379 home are eaten by the people, fed to dogs, or disposed of in places where dogs can readily access  
380 them. Feeding dogs on possibly infested raw meat or organs as done by majority of dog keepers  
381 also promote perpetuation of the parasite in dogs and the environment through dog faeces [20].  
382 Our findings revealed that inadequate deworming of dogs and livestock, poor dog faecal disposal  
383 and poor disposal of infested organs of animals slaughtered at home are risky practices by Busia  
384 communities [19,22].

385         The prevalence which has been estimated by this study might be lower than expected due to  
386 failure to include cattle whose owners could not be traced and therefore not having a chance to establish  
387 their infestation status and so the actual risk may be higher than reported. Failure to get positive results  
388 on PCR in two occasions might be explained by the fact that using strains to characterize  
389 *Echinococcus* is essential to establish a preventive and control strategy in every endemic area,  
390 but using DNA based methods for strain/genotype characterizations of *E. granulosus* have some  
391 difficulties, especially access to an efficient and pure concentration of DNA and proper primers  
392 [23].

393         There is a non-negligible risk of CH in Busia communities which might worsen with time  
394 given that the county is connected to areas perceived to be endemic for the disease (West Pokot  
395 and Turkana counties) via livestock trade. The local people also have poor KAP on the disease  
396 and hence there is need to implement information, education and communication campaigns to  
397 improve KAP on CH in the area. Cystic hydatidosis is an important but neglected zoonotic  
398 disease which should be put under surveillance by public health authorities in Kenya. The  
399 authors recommend commencement of Busia community public health education (PHE) to

400 improve knowledge, attitude and practices on the disease. The community PHE may also  
401 improve veterinary public health activities like deworming dogs, disposing of dog faeces,  
402 slaughter hygiene, meat inspection and sanitation measures. Future studies should focus on  
403 prevalence of CH in humans and dogs in Busia.

#### 404 **Acknowledgements**

405 We acknowledge the following institutions and programs for their funding and/or collaboration  
406 in this study; Ministry of Health (Kenya Field Epidemiology and Laboratory Training Program),  
407 Moi University, International Livestock Research Institute, county governments of Busia and  
408 Migori.

409 We acknowledge the following individuals for their participation or contribution in the  
410 development and review of the proposal and/or manuscript; Prof. Eric Fevre, Dr. Dalmas Oyugi,  
411 Dr. Mark Nanyingi, Dr. Annie Cook, Dr. Austin Bitek, Dr. Allan Ogendero, Dr. Kelvin Momanyi,  
412 Mrs. Mary Midida Owade, Mr. Benard Owade and Mr. Gilbert Nyandiga. We thank Dorothy L  
413 Southern for her critical review of the manuscript and her scientific writing support. We also  
414 acknowledge the immense contribution made by Kenya Field Epidemiology and Laboratory  
415 Training Program (K-FELTP) lecturers and Cohort 12 residents throughout the study period.

#### 416 **Author Contributions**

417 **Conceptualization:** Henry Joash Ogutu, Bernard Bett, Christina Otieno.

418 **Data curation:** Henry Joash Ogutu, Bernard Bett, Christina Otieno.

419 **Formal analysis:** Henry Joash Ogutu, Maurice Owiny.

420 **Investigation:** Henry Joash Ogutu.

421 **Methodology:** Henry Joash Ogutu, Maurice Owiny, Bernard Bett, Christina Otieno.

422 **Software:** Henry Joash Ogutu. Supervision: Bernard Bett, Christina Otieno.

423 **Supervision:** Bernard Bett, Christina Otieno, Maurice Owiny

424 **Writing – original draft:** Henry Joash Ogutu, Bernard Bett, Christina Otieno.

425 **Writing – review & editing:** Henry Joash Ogutu, Maurice Owiny, Bernard Bett, Christina  
426 Otieno.

427

#### 428 **Conflict of interest**

429 The authors declare that they do not have any competing interest.

#### 430 **Disclaimer**

431 The findings and conclusions in this manuscript are those of the authors and do not necessarily  
432 represent the official position of the Kenyan Ministry of Health or Moi University.

#### 433 **Reference**

- 434 1. Dinkel A, Njoroge EM, Zimmermann A, Wälz M, Zeyhle E, Elmahdi IE, et al. A PCR  
435 system for detection of species and genotypes of the *Echinococcus granulosus*-complex,  
436 with reference to the epidemiological situation in eastern Africa. *Int J Parasitol.* 2004;34:  
437 645–653. doi:10.1016/j.ijpara.2003.12.013
- 438 2. Grosso G, Gruttadauria S, Biondi A, Marventano S, Mistretta A. Worldwide epidemiology  
439 of liver hydatidosis including the Mediterranean area. *World J Gastroenterol.* 2012;18:  
440 1425–1437. doi:10.3748/wjg.v18.i13.1425
- 441 3. Pedro L. M. Treatment of echinococcosis [Internet]. 2018 [cited 12 Apr 2019]. Available:  
442 <https://www.uptodate.com/contents/treatment-of-echinococcosis>

- 443 4. Thatcher VE, Sousa OE. *Echinococcus oligarthrus* Diesing, 1863, in Panama and a  
444 comparison with a recent human hydatid. *Annals of Tropical Medicine & Parasitology*.  
445 1966;60: 405–416. doi:10.1080/00034983.1966.11686430
- 446 5. Odero JK, Japhet K. Magambo, Kutima HL, Ndahi L, Njonge FK. The burden of Cystic  
447 *Echinococcus* in selected regions in Kenya. 2015; doi:10.13140/rg.2.2.19423.05282
- 448 6. KNBS. 2009 Kenya Population and Housing Census: Volume 1A Population Distribution  
449 by Administrative Units. In: Kenya National Bureau of Statistics [Internet]. 13 May 2013  
450 [cited 12 Apr 2019]. Available: [https://www.knbs.or.ke/2009-kenya-population-and-](https://www.knbs.or.ke/2009-kenya-population-and-housing-census-volume-1a-population-distribution-by-administrative-units/)  
451 [housing-census-volume-1a-population-distribution-by-administrative-units/](https://www.knbs.or.ke/2009-kenya-population-and-housing-census-volume-1a-population-distribution-by-administrative-units/)
- 452 7. Khan A, Naz K, Ahmed H, Simsek S, Afzal MS, Haider W, et al. Knowledge, attitudes and  
453 practices related to cystic echinococcosis endemicity in Pakistan. *Infect Dis Poverty*.  
454 2018;7. doi:10.1186/s40249-017-0383-2
- 455 8. Schwartz HJ. *The One Humped Camel in Eastern Africa. A Pictorial Guide to Disease,*  
456 *Health Care and Management*. Available:  
457 [https://www.academia.edu/20539443/The\\_One\\_Humped\\_Camel\\_in\\_Eastern\\_Africa.\\_A\\_Pi](https://www.academia.edu/20539443/The_One_Humped_Camel_in_Eastern_Africa._A_Pictorial_Guide_to_Disease_Health_Care_and_Management)  
458 [ctorial\\_Guide\\_to\\_Disease\\_Health\\_Care\\_and\\_Management](https://www.academia.edu/20539443/The_One_Humped_Camel_in_Eastern_Africa._A_Pictorial_Guide_to_Disease_Health_Care_and_Management)
- 459 9. Nogueira GP. Puberty in South American *Bos indicus* (Zebu) cattle. *Anim Reprod Sci*.  
460 2004;82–83: 361–372. doi:10.1016/j.anireprosci.2004.04.007
- 461 10. Addy F, Alakonya A, Wamae N, Magambo J, Mbae C, Mulinge E, et al. Prevalence and  
462 diversity of cystic echinococcosis in livestock in Maasailand, Kenya. *Parasitol Res*.  
463 2012;111: 2289–2294. doi:10.1007/s00436-012-3082-8

- 464 11. Assefa H, Mulate B, Nazir S, Alemayehu A. Cystic echinococcosis amongst small  
465 ruminants and humans in central Ethiopia. *Onderstepoort J Vet Res.* 2015;82: E1-7.  
466 doi:10.4102/ojvr.v82i1.949
- 467 12. Mirzaei M, Rezaei H, Nematollahi A. Role of ruminants in the epidemiology of  
468 *Echinococcus granulosus* in Tabriz area, Northwest of Iran. *Trop Biomed.* 2015;32: 269–  
469 275.
- 470 13. Chalechale A, Hashemnia M, Rezaei F, Sayadpour M. *Echinococcus granulosus* in humans  
471 associated with disease incidence in domestic animals in Kermanshah, west of Iran. *J*  
472 *Parasit Dis.* 2016;40: 1322–1329. doi:10.1007/s12639-015-0681-1
- 473 14. Tembo W, Nonga HE. A survey of the causes of cattle organs and/or carcass condemnation,  
474 financial losses and magnitude of foetal wastage at an abattoir in Dodoma, Tanzania.  
475 *Onderstepoort J Vet Res.* 2015;82. doi:10.4102/ojvr.v82i1.855
- 476 15. Elmajdoub L, A. Rahman W. Prevalence of Hydatid Cysts in Slaughtered Animals from  
477 Different Areas of Libya. *Open Journal of Veterinary Medicine.* 2015;05: 1–10.  
478 doi:10.4236/ojvm.2015.51001
- 479 16. Omadang L, Chamai M, Othieno E, Okwi A, Olaki Inangolet F, Ejobi F, et al. Knowledge,  
480 attitudes and practices towards cystic echinococcosis in livestock among selected pastoral  
481 and agro-pastoral communities in Uganda. *Tropical Animal Health and Production.*  
482 2017;50. doi:10.1007/s11250-017-1394-5

- 483 17. Nyakarahuka L. Knowledge, attitude and practices towards cystic echinococcosis in  
484 pastoral communities in Kasese District, Uganda [Internet]. Thesis, Makerere University.  
485 2011. Available: <http://makir.mak.ac.ug/handle/10570/2095>
- 486 18. Battelli G. Echinococcosis: costs, losses and social consequences of a neglected zoonosis.  
487 *Vet Res Commun.* 2009;33 Suppl 1: 47–52. doi:10.1007/s11259-009-9247-y
- 488 19. Gebremichael D, Feleke A, Gebremaryam G, Awel H, Tsigab Y. Knowledge, attitude and  
489 practices of hydatidosis in pastoral community with relation to public health risks in  
490 Ayssaita, northeastern of Ethiopia. *Global Veterinaria.* 2013;11: 272–279.  
491 doi:10.5829/idosi.gv.2013.11.3.7570
- 492 20. Yang YR, McManus DP, Huang Y, Heath DD. Echinococcus granulosus Infection and  
493 Options for Control of Cystic Echinococcosis in Tibetan Communities of Western Sichuan  
494 Province, China. *PLoS Negl Trop Dis.* 2009;3. doi:10.1371/journal.pntd.0000426
- 495 21. Wumbiya SD, Francis M, Wilfred E, Nasinyama GW, Eystein S, Adrian M, et al.  
496 Knowledge Attitude and Practices towards Cystic Echinococcosis among Pastoral  
497 Communities in Greater Kapoeta South Sudan. 2017.
- 498 22. Ernest E, Nonga HE, Kassuku AA, Kazwala RR. Hydatidosis of slaughtered animals in  
499 Ngorongoro district of Arusha region, Tanzania. *Trop Anim Health Prod.* 2009;41: 1179–  
500 1185. doi:10.1007/s11250-008-9298-z
- 501 23. Harandi MF, Hobbs RP, Adams PJ, Mobedi I, Morgan-Ryan UM, Thompson RCA.  
502 Molecular and morphological characterization of Echinococcus granulosus of human and  
503 animal origin in Iran. *Parasitology.* 2002;125: 367–373. doi:10.1017/S0031182002002172

504

505