IDENTIFICATION OF LEPTOSPIRA STRAIN IN RAT AT LEPTOSPIROSIS AREA IN GRESIK DISTRICT, EAST JAVA PROVINCE

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Abstrak. *Leptospirosis merupakan penyakit bersumber binatang (zoonosis), dapat ditularkan oleh hewan domestik (anjing, kucing, babi, sapi) dan binatang pengerat, terutama tikus. Jenis bakteri Leptospira yang ditularkan oleh tikus merupakan bakteri yang paling berbahaya bagi manusia dibandingkan semua bakteri yang ada pada hewan domestik. Tujuan penelitian ini untuk mengetahui keberhasilan penangkapan, species tikus yang berada di lokasi penelitian, strain leptospira pada tikus, mengetahui kepositifan leptospira berdasarkan kelompok Rattus atau non Rattus, penempatan perangkap dan lokasi penangkapan, serta mengetahui hubungan antara jenis spesies yang didapatkan, penempatan perangkap dan lokasi penangkapan dengan kepositifan leptospira pada tikus. Penelitian observasional dengan desain potong lintang. Penangkapan tikus dilakukan pada dua lokasi yaitu di Kecamatan Dukun dan Duduk Sampeyan, pada bulan Agustus-November 2010. Sebanyak 300 perangkap dipasang pada masing-masing lokasi yaitu di dalam rumah 2 perangkap dan di luar rumah 2 perangkap, selama 3 malam berturut-turut. Tikus yang tertangkap diidentifikasi, dihitung kepadatannya dan diambil serum dari darah jantung untuk diperiksa keberadaan bakteri leptospira secara Microscopic Aglutination Test (MAT). Keberhasilan penangkapan tikus di Dukun dan Duduk Sampeyan tinggi (di atas 7%). Ditemukan 7 strain leptospira interogans pada 13 tikus dengan persentase terbanyak pada Rattus norvegicus. Hasil uji statistik menunjukkan adanya hubungan kelompok Rattus dengan angka kepositifan bakteri leptospira pada α=5% bahwa ada hubungan kelompok Rattus untuk positif bakteri leptospira. Kelompok non Rattus berisiko 0,147 kali dibandingkan Rattus pada 95% CI 0,040-0,534, dengan kata lain Rattus lebih berisiko dibandingkan non Rattus untuk positif bakteri leptospira. Kelompok Rattus lebih berisiko untuk positf bakteri leptospira dibandingkan non Rattus*

Kata kunci: *leptospira, tikus, gresik*

INTRODUCTION

Leptospirosis is one of zoonotic diseases, transmitted by domestic animal (dogs, cats, pigs and cows) and rodent specially rat. Kind of leptospira that transmitted by rat was the most dangerous bacteriaa for human compared with another bacteriaa in domestic animals. Leptospira bacteria transmitted to human direct via skin lesion, mucosae of nose, mouth or eye or indirect via water, soil, mud, plants and food contaminated leptospira bacteria. Incubation period about 2 days until 4 weeks. This bacteria lives in proximal renal tubules and excreted in urine [1]

Data from Gresik Health District Agency, [2] showed leptospirosis case in the year of 2007 as much 27 cases with *Case Fatality Rate* (CFR) 7.4%, in 2008 as much 24 cases with CFR 29.17% and in 2009 as much 34 cases with CFR 28.13%, and until October 2010 as much 25 cases

with CFR 60%. Location of rat survey in Dukun and Duduk Sampeyan Sub district with delibration in last of 3 years at this area always found leptospirosis cases and at 2010 there were new cases at this location.

Survey in Semarang city, Central Java Province found 8% (5 from 62 rat blood serra sample had been examined by MAT) showed positive leptospira. As much 80% of rat that posititive leptospira were R. norvegicus. Strain that showed positive reaction in MAT were *L. bataviae* dan *L. hardjo*.[3]

Rat is main reservoirs of leptospirosis transmission. This study aimed to describe : trap success, rat species, leptospira strain bacteria, existence of leptospira bacteria at rat according *Rattus* group, rat position (indoor or outdoor) and research location, to find out relationship between *Rattus* group, rat position and research location with existence of leptospira bacteria in rat.

METHODS

Cross sectional design by rat trap survey had been done in two location there were Sambogunung Village, Dukun Sub district and Panjungan Village, Duduk Sampeyan Subdistrict, Gresik District on Augst-November 2010. Population of this survey were rat, mus and Suncus in research area. Sample in this survey were rats and suncus that had been trapped. As much 300 traps were put at research area in and outdoor during 3 days. Every morning trap were checked. Rat and suncus that had been trapped and than euthanated using atropine 0,02-0,05 mg/kg rat weight followed by ketamin HCL 50-100 mg/kg rat weight by injected in thick muscle of rat or suncus thigh. [4] After that, take rat or suncus sera from cardiac for leptospira bacteria identification by used *Microscopic Aglutination Test* (MAT) technique [5], identification of rats and counting of trap success. [6] Rattus group divided in to Rattus and non Rattus group, trap position divided in to indoor or outdoor and research location divided in to Dukun and Duduk Sampeyan. Data analyzed and presented in table and statistically by using chi square. Ethical clearance got from Ethic Commission, National Institute Health Research and Development, Health Ministry, Republic of Indonesia.

RESULTS

Insectivora *Suncus murinus* and *Mus* also trapped in this survey. Because of it's role in leptospirosis transmission *Suncus* and *Mus* joined to analysis. Trap success in Sambogunung Village, Dukun Sub district as much 19% and in Panjungan Village, Duduk Sampeyan Sub district as much 9%. Rat and suncus species that had been trapped in this research.

Table 1 showed rat and suncus that had been found in high percentage identified as *Rattus tanezumi* and *R. norvegicus* as much 36.54%. *Mus cervicolor* found rarely. Trap success indoor foun higher than outdoor. Female rat found higher than male.

As much 132 *Rat* and *Suncus* blood sera examine in laboratory by using MAT technique. Table 2 showed rat and suncus that positive leptospira bacteria by MAT. *Rattus norvegicus* or riol rat that found indoor was mostly showed positive leptospira in Dukun Subdistrict. Leptospira bacteria strain that showed positive reaction with sample from Dukun area were 5 strain that were *Leptospira hardjo, L. bataviae, L.icterohaemorhagie, L. australis, L.gryphotyphosa.* Two of nine sample that positive leptospira was *Suncus*

murinus. showed that insectivore *Suncus murinus* take apart in leptospirosis transmission. Rat trap find in door higher than outdoor.

Table 3 showed rat species that had positive reaction with leptospira strain bacteria that used in MAT. Strain of leptospira bacteria that showed positive reaction were : *L.icterohaemorhagie, L. rachmat, L. australis, L. pomona, L. hardjo,* and *L. bataviae*. Rat that mostly found was *Bandicota bengalensis*. Rat trap that positive leptospira by MAT found indoor and outdoor in same proportion, it was indicate that leptospira transmission indoor and outdoor had the same possibility

Table 1. Species and Number of Rat and Suncus that had been Trapped

Num	Species	Number of Rat Trapped			M	F	Y	Sum	%
		Out	In	NL					
1	*Rattus tanezumi*	32	39	2	26	46	1	73	46.79
2	*Rattus norvegicus*	16	40	1	16	41	0	57	36.54
3	*Mus musculus*	2	0	0	2	0	0	2	1.28
4	*Bandicota bengalensis*	4	3	0	2	5	0	7	4.49
5	*M. cervicolor*	0	1	0	1	0	0	1	0.64
6	*Suncus murinus*	7	8	1	5	11	0	16	10.26
	Trap success	21.18%	31.60%						
	Total	61	91	4	52	103	1	156	100

Note :
Out : outdoor NL : No label F : Female
In : Indoor M : Male Y : Young

Table 2. Result of Positive Leptospira by Laboratory Examinated using Microscopic Aglutination Test Technique (n=83), Sample from Dukun Subdistrict

Num.	Species	in/outdoor	Strain of Leptospira bacteria
1	*R. norvegicus*	indoor	*L. hardjo, L. Bataviae*
2	*R. norvegicus*	indoor	*L. bataviae*
			L.hardjo, L. bataviae,
3	*R. norvegicus*	indoor	*L. icterohaemorhagie*
4	*R. tanezumi*	indoor	*L. hardjo*
			L.hardjo, L. bataviae,
5	*Suncus murinus*	indoor	*L. icterohaemorhagie*
			L.hardjo, L. bataviae,
6	*R. norvegicus*	outdoor	*L. icterohaemorhagie*
7	*R. norvegicus*	indoor	*L. bataviae*
8	*R. norvegicus*	indoor	*L.hardjo, L. bataviae, L.icterohaemorhagie, L. australis, L.gryphotyphosa*
9	*Suncus murinus*	outdoor	*L. bataviae*

Table 3. Result of Positive Leptospira by Laboratory Examinated Using Microscopic Aglutination Test Technique (n=49), Sample from Duduk Sampeyan Subdistrict

Num.	Species	in/outdoor	Strain of Leptospira bacteria
1	*B. bengalensis*	outdoor	*L. bataviae*
2	*B. bengalensis*	outdoor	*L. hardjo, L. Bataviae*
3	*R. tanezumi*	indoor	*L.hardjo, L. rachmat, L.gryphotyphosa, L. hardjo, L. Bataviae*
4	*B. bengalensis*	indoor	*L.icterohaemorhagie, L. rachmat, L. australis, L. pomona, L. hardjo, L. Bataviae*

Table 4. Existence of Leptospira Bacteria at Rat According *Rattus* Group, Trap Location and Research Location

Variale	Positivity of leptospira bacteria positive		negative		Odds Ratio	95% Confidence Interval	p
	n	%	n	%			
Species							
Rattus	8	61.5	109	91.6	1.00	Referensi	
Non rattus	5	38.5	10	8.4	0.147	0.040-0.534	0.001
Trap location							
Di dalam rumah	9	69.2	72	60.5	1.00	Referensi	
Di luar rumah	4	30.8	47	39.5	1.469	0.428-5.044	0.504
Research location							
Dukun	9	69.2	74	62.2	1.00	Referensi	
Duduk Sampeyan	4	30.8	45	37.8	1.368	0.398-4.703	0.618

Leptospira positive bacteria according to species, trap location and area of rat trap survey described in Table 4.

DISCUSSION

A limitation of this study was observation done in one moment, so result of this research just describe condition when survey had been done. But, result of this survey showed there was leptospirosis transmitted cycle that related with rat as main reservoir.

Trap success showed in Dukun Subdistrict as much 19%, higher than in Duduk Sampeyan Subdistrict as much 9 %. Trap success described crude rat population. Trap success higher than 7% showed that crude rat population was high. [7]

Table 4 showed positive leptospira bacteria in *Rattus* group higher than non

Rattus group. Location of rat that positive leptospira bacteria indoor higher than outdoor. In Dukun area rat with positive leptospira bacteria more than in Duduk Sampeyan. Statistical analysis showed there was relationship between *Rattus* group and positive leptospira bacteria with p value 0.001, Non *Rattus* had risk to positive leptospira bacteria 0.147 rather than Rattus. (Odds Ratio 0.147 Confidence Interval 0.040-0.534), in another word Rattus had more risk to positive leptospira bacteria.

Strain that showed positive reaction in MAT in Dukun Subdistrict were *Leptospira hardjo, L. bataviae, L. icterohaemorhagie, L. australis, L.gryphotyphosa* and in Duduk Sampeyan Sub district were *L.icterohaemorhagie, L. rachmat, L. australis, L. pomona, L. hardjo, L. bataviae.* From strain that showed positive reaction, *L. icterohaemorhagie* was a kind of leptospira strain that most virulence to human.[8]

Existance of leptospira bacteria at rat showed one of leptospirosis transmission way to human was from rat. *Rattus norvegicus* (riol rat) was mostly species found in Dukun Subdistrict. This rat had main habitat in riol or water seepage so had high possibility to infected leptospira from water. Infected rat had very high possibility to infected their child caused have activity in same place (nested). *Rattus norvegicus* that positive leptospira by MAT that mostly found indoor showed there were transmission of leptospirosis indoor. Research in Semarang City showed the same pattern that *R. norvegicus* dominantly positive leptospira by MAT (80% from 5 rat positive leptospira).[3]

Survey in Duduk Sampeyan Subdistrict showed rat species that mostly found positive leptospira was *Bandicota bengalensis.* This rat had medium size. Survey area in Duduk Sampeyan was settlement area that near with rice field. *Bandicota bengalensis* had natural habitat : grassland, rice field and coppice. This species even found in Kudus and Semarang City (Central Java), Wlingi (East Java).[9] Trap location from rat positive leptospira had the same proportion indoor and outdoor, so there was possibility for leptospira transmission indoor and outdoor. Research in Madurai, India also showed leptospira interogans found at rat.[10] Leptospirosis in Peru showed something that same if rat as reservoir of leptospirosis, by PCR *(Polymerase Chain Reaction)* technique found that species *Rattus rattus* dan *Rattus norvegicus* positive leptospira.[11] Statistic analysis showed there were relationship between *Rattus* group and positivity of leptospira bacteria. In conclusion, *Rattus* group had more risk to positive leptospira bacteria than non *Rattus.*

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