

**Japanese Spotted Fever**  
**Recent situation of occurrence**  
**in Mie Prefecture, Japan**  
**and diagnosis by PCR method using**  
**eschar and erythema from patients**

**Katsuhiko Ando** : Faculty of Medicine, Mie University  
**Shigehiro Akachi** : Mie Prefectural Institute  
**Shigetoshi Sakabe** : Yamada Red Cross Hospital  
**Yasuo Chinzei** : Faculty of Medical Engineering,  
Suzuka University

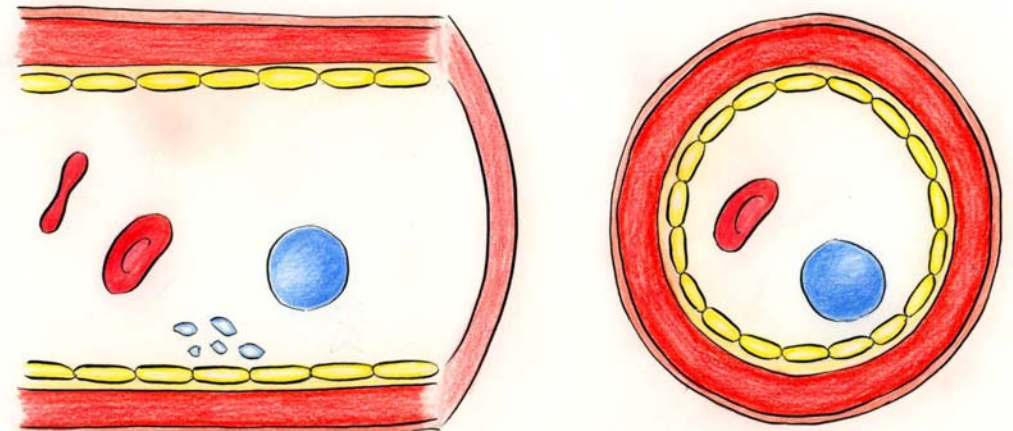
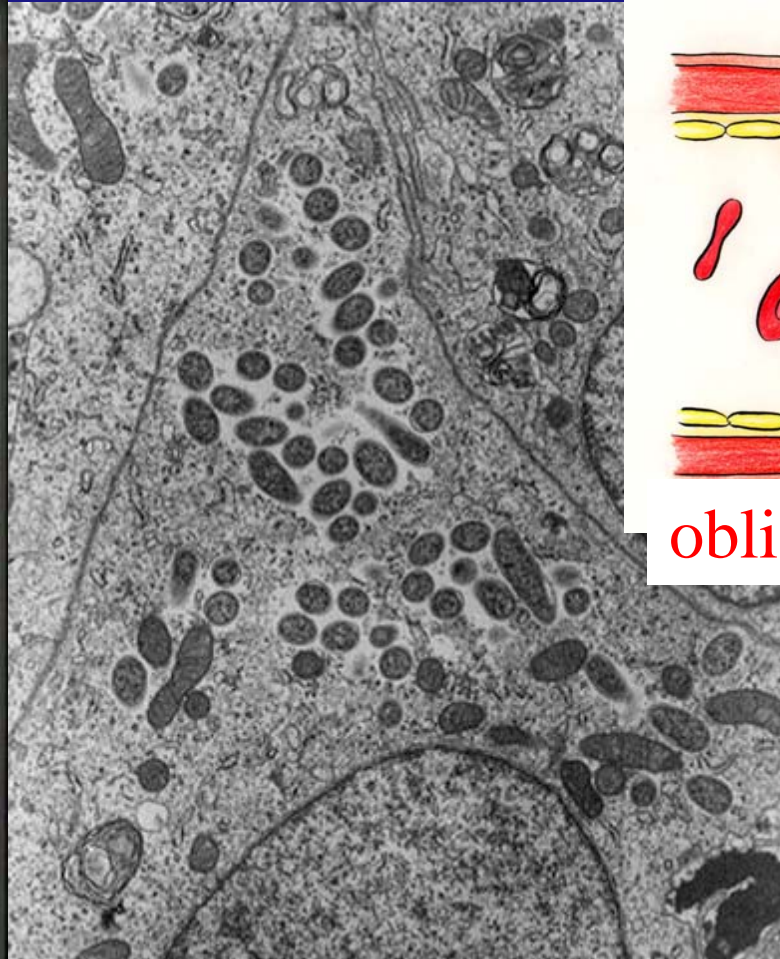
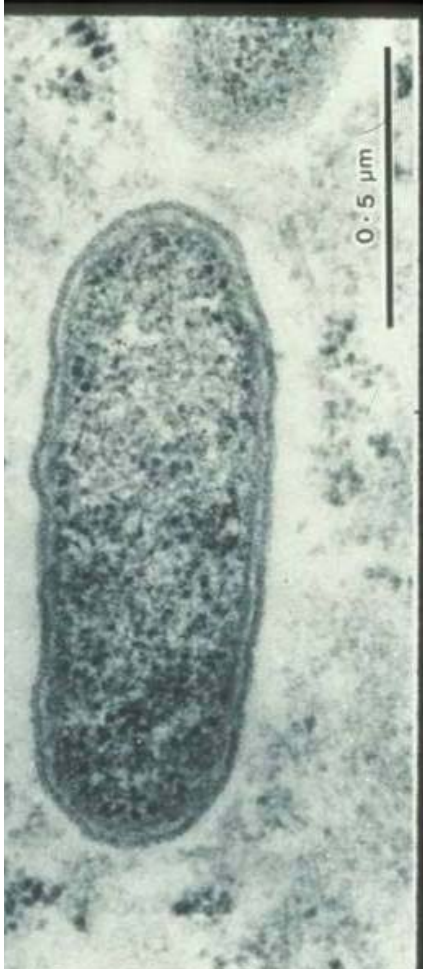
# Distribution and host

Disease (organism)	Geographic Area	Host
<b>Spotted fever group</b>		
RM spotted fever ( <i>R. rickettsi</i> )	Western hemisphere	Tick Wild rodent
Japanese spotted fever ( <i>R. japonica</i> )	Japan, Korea	Tick ?
Queensland tick typhus ( <i>R. australis</i> )	Australia	Tick Wild rodent
North Asia tick typhus ( <i>R. sibirica</i> )	Siberia, Mongolia	Tick Wild rodent
<b>Tsutsugamushi group</b>		
Scrub typhus ( <i>Orientia tsutsugamushi</i> )	Asia, Australia	Mite Wild rodent

# Morphology of Pathogen of Japanese spotted fever, *Rickettsia japonica*

Size :  $2 \mu\text{m} \times 0.5 \mu\text{m}$

Proliferation : vascular endothelial cell in blood vessel



obligately intracellular parasites

# History of Discovery in Japan and Mie Prefecture

- Japanese spotted fever (JSF) is a tick borne disease.
- Causative agent is *Rickettsia japonica*.
- The first case was reported in Tokushima in 1984.
- Since then, more than 700 cases are reported for 24 years, mainly western Japan.
  
- In Mie Prefecture, first case was reported in 1988.
- Since then, no case was reported until 2000.
- Unknown rickettsial diseases occurred from 2001
- Confirmed 2 cases in 2005 and fatal case in 2006.

# Clinical features

- JSF is transmitted by a tick bite from spring to autumn (April to Nov.) in Japan
- One week later
  - High fever (39~40C)**
  - Erythema (extremities to trunk)**
  - Tick bite eschar**
- Severe case : Convulsion, Consciousness disturbance by toxin, DIC, Death
- Treatment: **Minocycline + Levofloxacin**

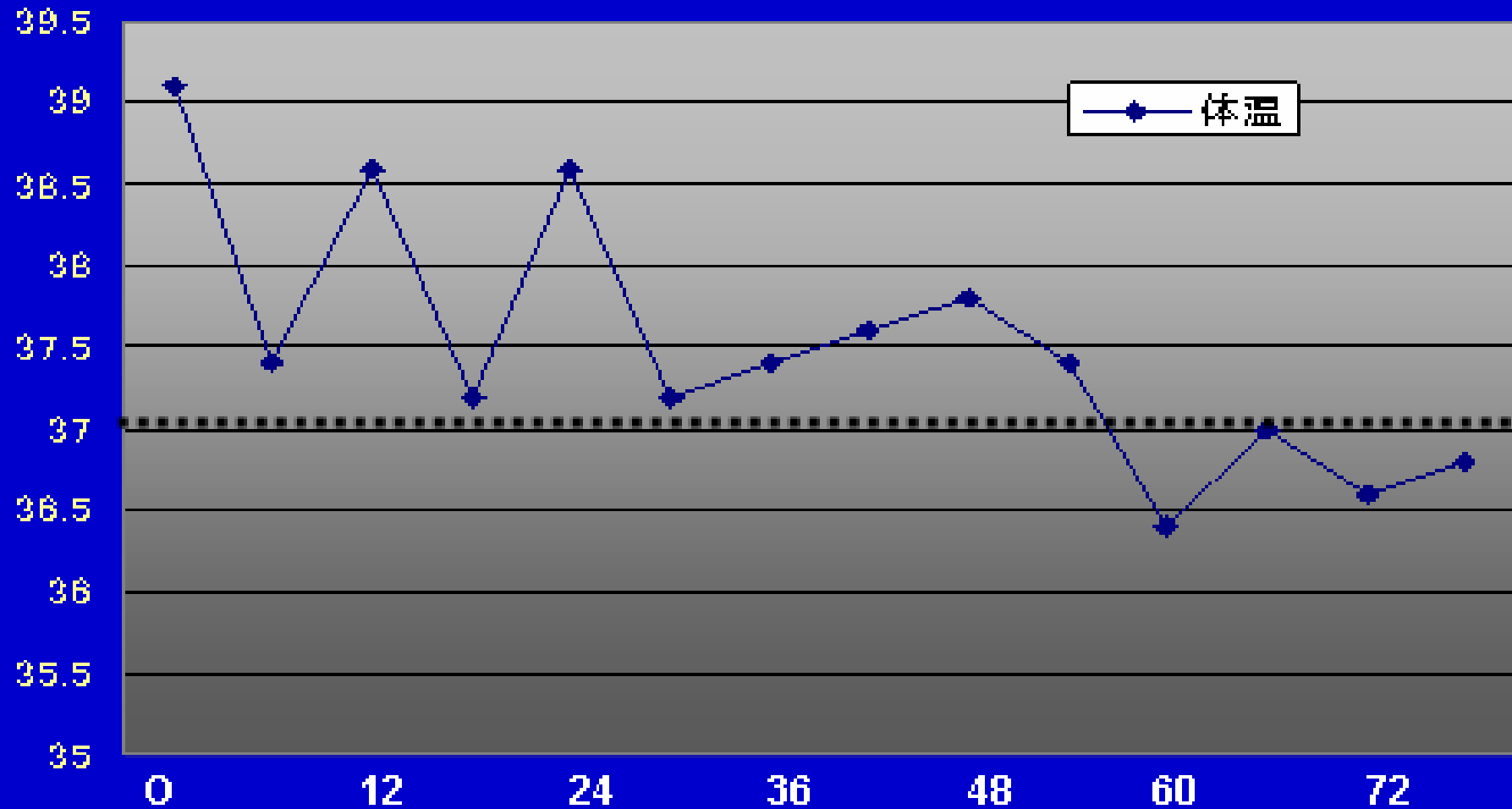


# Treatment

progress

Treatment: Minocycline + Levofloxacin (Oral administration)

体温(℃)



Time from treatment (hours)

# Diagnosis

- **Indirect immunofluorescence**
  - Antibody for acute phase –low
  - Antibody for recovery phase- high
- **Monoclonal antibody**
  - Specificity low
- **PCR using whole blood**
  - Sensitivity low
  - ↓
  - (Acute phase)
  - PCR using biopsied skin of eschar and erythema**

# Method of PCR

**First PCR**  
(for SF group)



**17-kDa Membrane Protein**



## Primer

**R2: TTTACAAAATTCTAAAACG**

**R1: TCAATTCACAACCTTGCCATT**

**Rj10: ATTCTAAAACCATATACTG**

**Rj5 : CGCCATTCTACGTTACTACC**

**1 step 1cycle 95C 2min**

**2 step 30cycle 94C 45sec**

**52C 30sec**

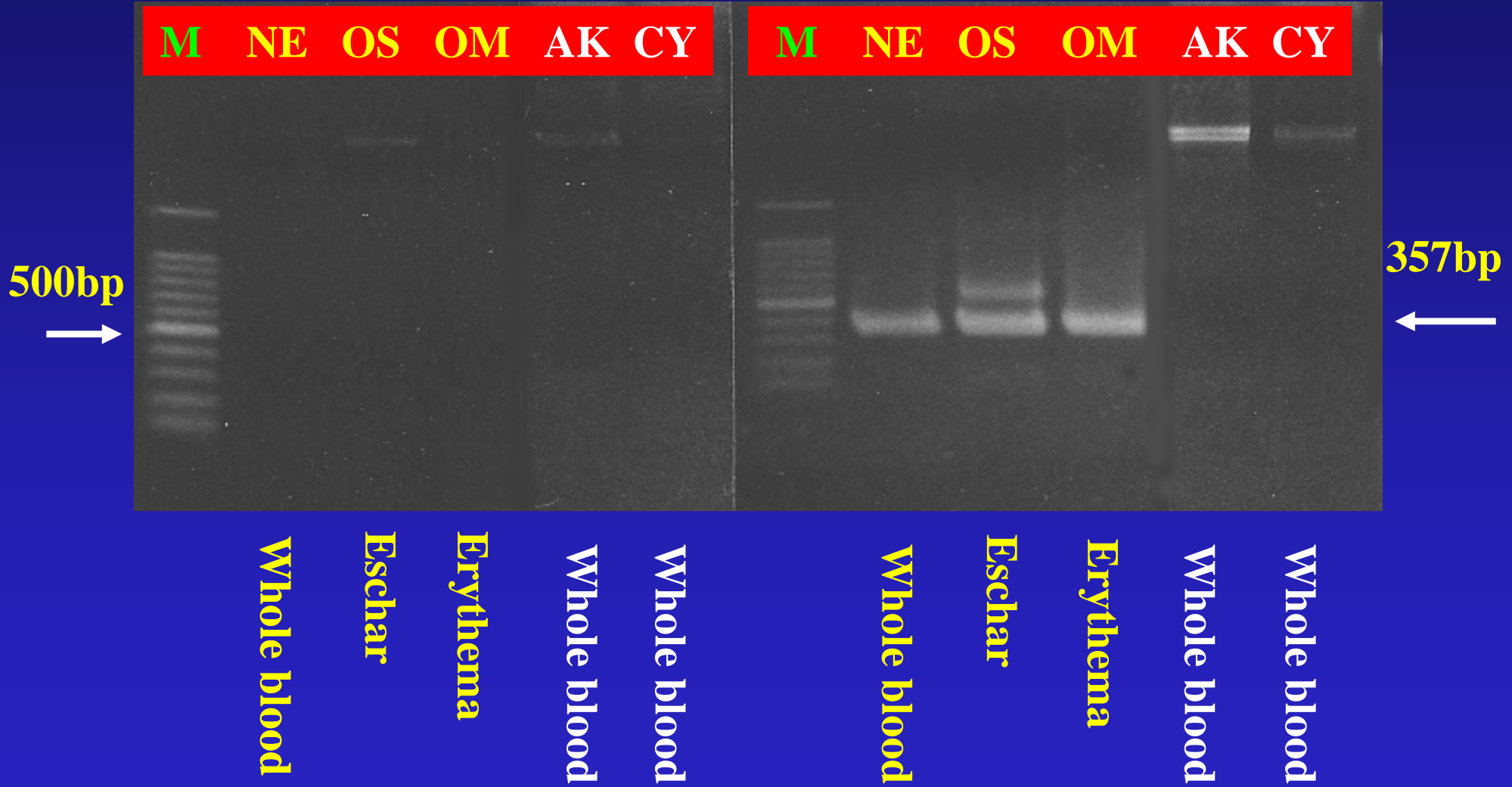
**72C 45sec**

**3 step 1cycle 72C 5min**

# Result of PCR

## First PCR

## Nested PCR



# Nucleotide sequence of 317bp of 17kDa membrane protein

*R. japonica* 1 ATTAATTATATATTAATAGAGAGAATTATATGAAACTATTATCTAAAATTATGATTATAGCTCTTGCAACTTCTATGTTA  
W. Blood 1 ATTAATTATATATTAATAGAGAGAATTATATGAAACTATTATCTAAAATTATGATTATAGCTCTTGCAACTTCTATGTTA  
Eschar 1 ATTAATTATATATTAATAGAGAGAATTATATGAAACTATTATCTAAAATTATGATTATAGCTCTTGCAACTTCTATGTTA  
Erythema 1 ATTAATTATATATTAATAGAGAGAATTATATGAAACTATTATCTAAAATTATGATTATAGCTCTTGCAACTTCTATGTTA  
\*\*\*\*\*

*R. japonica* 81 CAAGCCTGTAACGGTCCGGGCGGTATGAATAACAAGGTACAGGAACACTTCTTGCGGTGCTGGTGGCGCATTACTTGG  
W. Blood 81 CAAGCCTGTAACGGTCCGGGCGGTATGAATAACAAGGTACAGGAACACTTCTTGCGGTGCTGGTGGCGCATTACTTGG  
Eschar 81 CAAGCCTGTAACGGTCCGGGCGGTATGAATAACAAGGTACAGGAACACTTCTTGCGGTGCTGGTGGCGCATTACTTGG  
Erythema 81 CAAGCCTGTAACGGTCCGGGCGGTATGAATAACAAGGTACAGGAACACTTCTTGCGGTGCTGGTGGCGCATTACTTGG  
\*\*\*\*\*

*R. japonica* 161 TTCTCAATTCGGTAAGGGCACAGGACAGCTTGTGGGAGTAGGTGTAGGTGCATTACTTGGAGCAGTTCTTGGTGGACAAA  
W. Blood 161 TTCTCAATTCGGTAAGGGCACAGGACAGCTTGTGGGAGTAGGTGTAGGTGCATTACTTGGAGCAGTTCTTGGTGGACAAA  
Eschar 161 TTCTCAATTCGGTAAGGGCACAGGACAGCTTGTGGGAGTAGGTGTAGGTGCATTACTTGGAGCAGTTCTTGGTGGACAAA  
Erythema 161 TTCTCAATTCGGTAAGGGCACAGGACAGCTTGTGGGAGTAGGTGTAGGTGCATTACTTGGAGCAGTTCTTGGTGGACAAA  
\*\*\*\*\*

*R. japonica* 241 TCGGTGCAGGTATGGATGAGCAGGATAGAAGACTTGCAGAGCTTACCTCACAGAGAGCTTTAGAAACAGCTCCTAGT  
W. Blood 241 TCGGTGCAGGTATGGATGAGCAGGATAGAAGACTTGCAGAGCTTACCTCACAGAGAGCTTTAGAAACAGCTCCTAGT  
Eschar 241 TCGGTGCAGGTATGGATGAGCAGGATAGAAGACTTGCAGAGCTTACCTCACAGAGAGCTTTAGAAACAGCTCCTAGT  
Erythema 241 TCGGTGCAGGTATGGATGAGCAGGATAGAAGACTTGCAGAGCTTACCTCACAGAGAGCTTTAGAAACAGCTCCTAGT  
\*\*\*\*\*

# Summary of Results in 2008

	Sample	No. Exam.	No. Posi.	Posi.(%)
	Whole blood (PCR)	45	21	46.7
	Eschar (PCR)	18	17	94.4
<b>Acute phase</b>	Erthema (PCR)	16	13	81.3
	Serum (IFA)			
	IgM	43	13	30.2
	IgG	43	9	20.9
-----				
	Serum (IFA)			
<b>Recovery phase</b>	IgM	28	27	96.4
	IgG	28	24	85.7
<b>Total</b>		<b>45</b>	<b>35</b>	

# Analysis of Positive Cases

---

<b>Year</b>	<b>No. positive Whole Blood PCR</b>	<b>No. positive Eschar or Erythema or PCR</b>	<b>No. positive Antibody PCR</b>	<b>Positive (%)</b>
<b>2008</b>	<b>19</b>	<b>19</b>	<b>19</b>	<b>100</b>

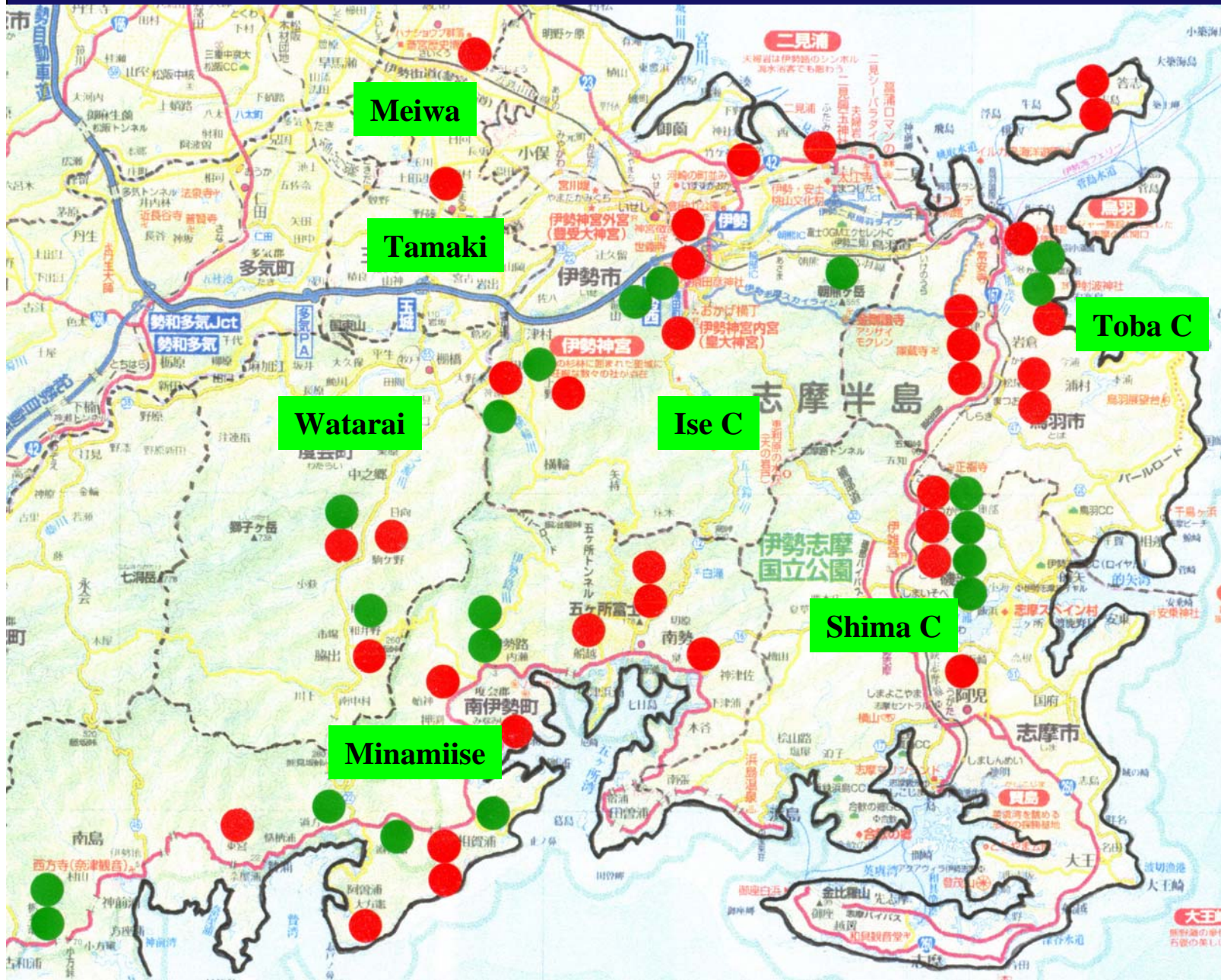
---

---

<b>Year</b>	<b>No. positive Eschar or Erythema or PCR</b>	<b>No. positive Antibody PCR</b>	<b>No. positive Whole Blood PCR</b>	<b>Positive (%)</b>
<b>2008</b>	<b>33</b>	<b>33</b>	<b>19</b>	<b>57.5</b>

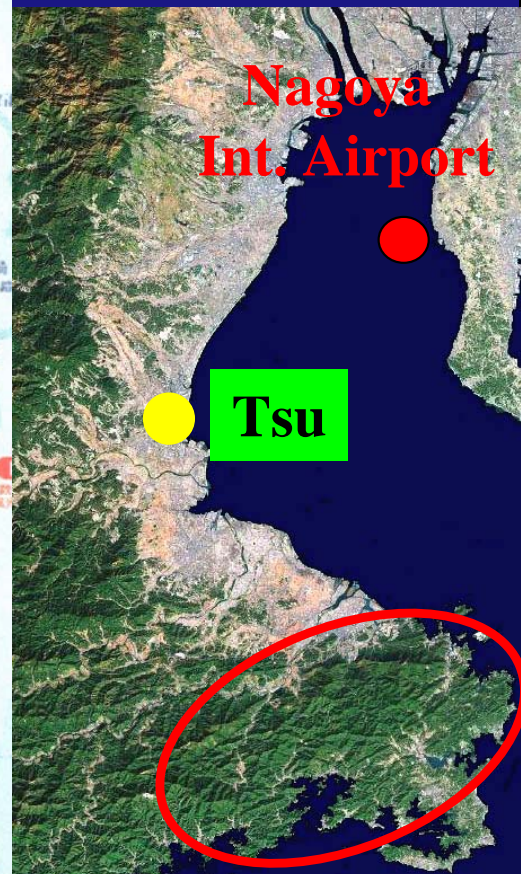
---

# Distribution of Patients



● 2008

● 2007



Nagoya  
Int. Airport

Tsu

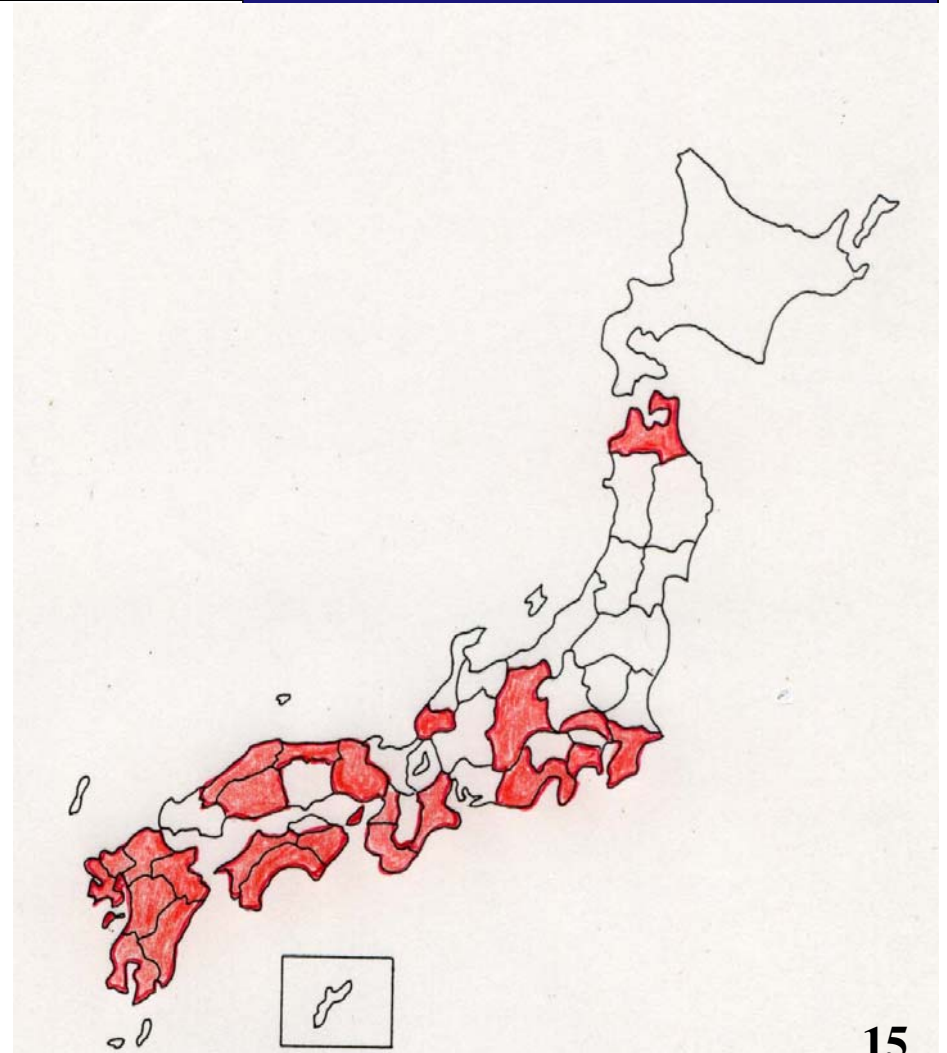
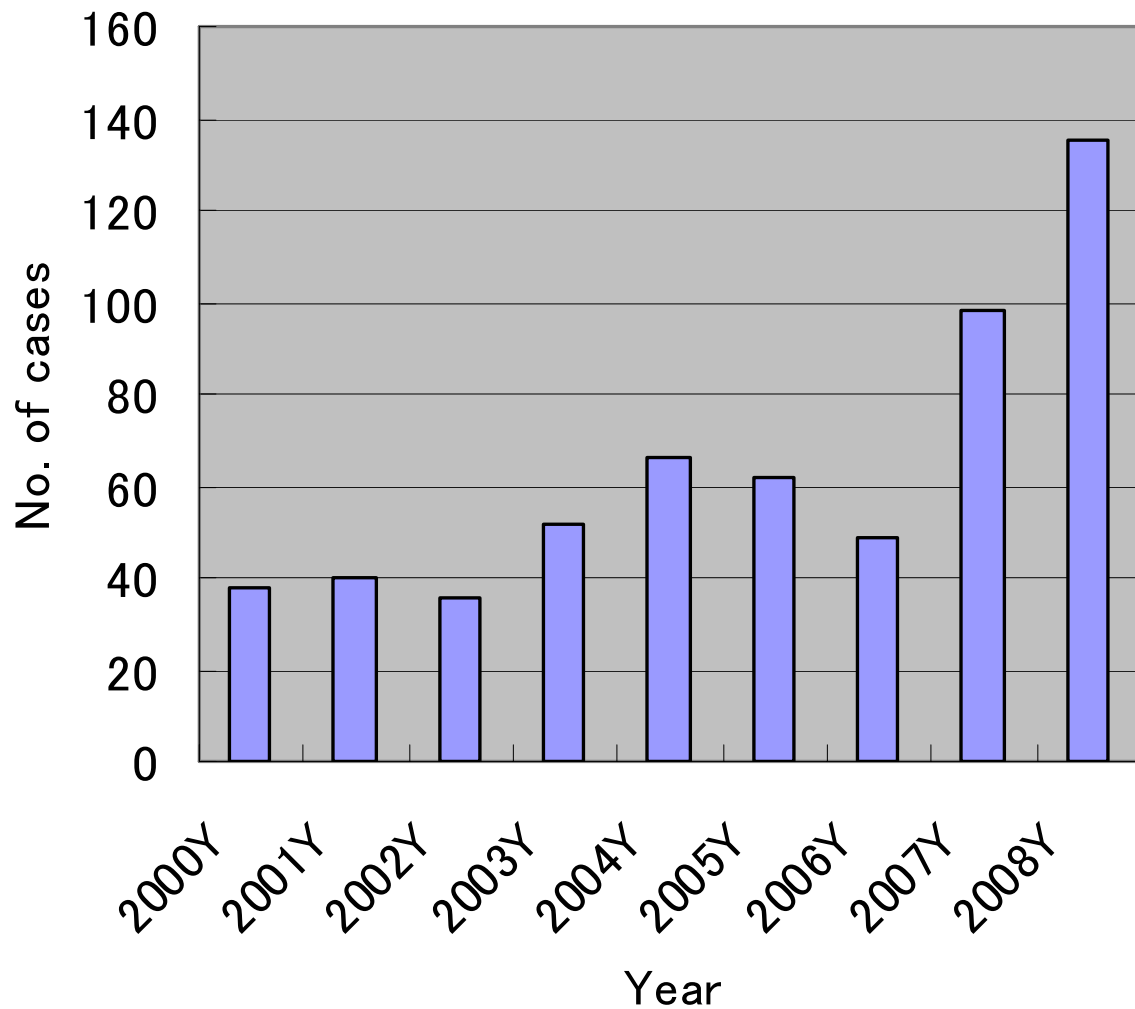
# Occurrence in each prefecture

---

	2007	2008		2007	2008
<b>Mie</b>	<b>20</b>	<b>35</b>	Miyazaki	4	<b>7</b>
Wakayama	16	16	Tokushima	2	2
Kagoshima	16	<b>11</b>	Nagasaki	2	2
Kumamoto	11	<b>18</b>	Aomori	1	0
Shimane	10	<b>13</b>	Chiba	1	<b>7</b>
Hiroshima	5	<b>4</b>	Tottori	1	2
Hyogo	4	4	Kochi	1	<b>6</b>
Ehime	4	<b>5</b>			
			<b>Total</b>	<b>98</b>	<b>135</b>

---

# Number of cases and distribution in Japan

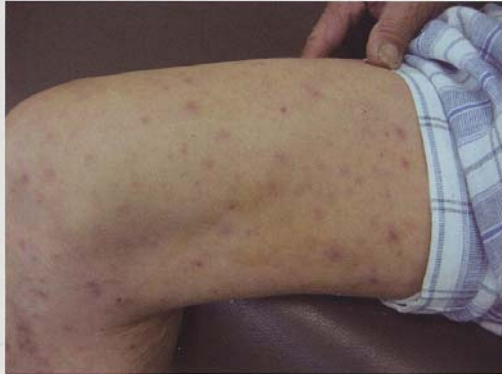


にほんこうはんねつ  
「日本紅斑熱」に注意しましょう！

1. 「日本紅斑熱」とは、野山に生息している「リケッチアという病原体をもっているダニ」に刺されることで感染する病気です。
2. 春から秋にかけて感染し、2～8日後に次のような症状がでます。

<日本紅斑熱の症状>

体がだるい 高熱（39～40度以上）がでる 発疹がでる



↑<刺し口>

←日本紅斑熱の発疹（痒くないのが特徴的）

3. **症状が出たら直ちに医療機関で診察してもらいましょう！**
4. 速やかに治療する必要があります！
5. 治療が遅れると命にかかわることもあるので気をつけて！
6. **人から人に感染することはありません！**



<ヤマトマダニ>



<野山に立ち入る際の注意事項（ダニに刺されないことが大切！）>

1. 肌をださないよう、長袖、長ズボン、手袋などをする。
2. ダニの忌避剤を使用する。
3. 地面に直接寝ころんだり、腰を下ろしたりしない。
4. 立ち入った後入浴し、体についたダニを落としましょう。ダニに刺されているときは、指でつぶさず、頭部をピンセットなどで摘んで慎重に取り除くか、医療機関で取ってもらいましょう。

<問い合わせ先>

伊勢保健福祉事務所保健衛生室（伊勢保健所）地域保健課

三重県伊勢市勢田町6 2 2 番地（TEL：0596-27-5148, FAX：0596-27-5253）

※発疹及び刺し口の画像提供：山田赤十字病院

※ヤマトマダニの出典：「日本の有害節足動物（加納六郎、篠永哲著）」、協力：三重大学医学部医動物学教室

# Pamphlet to prevent tick bite

**Thank you very much**

