

## Revised Abstract

**Objectives:** Tigecycline, a glycolcycline, has been shown to have potent broad spectrum activity against most commonly encountered respiratory pathogens. This report focuses on *in vitro* susceptibility trends of Asian *Haemophilus influenzae* (HI) isolates using data from the Tigecycline Evaluation and Surveillance Trial (TEST). **Methods:** From 2005-2010, 566 HI isolates were collected in 8 Asian countries from various infection sources. MICs were performed at each site per CLSI guidelines using commercially-prepared microbroth panels. Tigecycline results were interpreted according to FDA breakpoints. Trends in the proportion of  $\beta$ -lactamase producers were assessed by the Cochran-Armitage test and MIC trends by Spearman's correlation. **Results:** Of 566 *H. influenzae* strains, 173 (30.6%) were  $\beta$ -lactamase positive (24.4% in 2005, 29.6% in 2006, 38.3% in 2007, 31.5% in 2008, 20% in 2009, 27.3% in 2010,  $p$  for trend = 0.51). The % susceptible, MIC<sub>90</sub> (mcg/ml), geometric mean MIC, and  $p$  values for MIC trends for tigecycline are shown below.

Species/phenotype (n per year)	% Susceptible MIC <sub>90</sub> /geometric mean MIC						MIC trend $p$
	2005	2006	2007	2008	2009	2010	
<i>H. influenzae</i> (45/135/167/89/75/55)	100 0.25/0.125	99.3 0.25/0.126	98.2 0.25/0.128	100 0.25/0.147	100 0.12/0.091	100 0.25/0.149	0.85
HI $\beta$ -lactamase neg. (34/95/103/61/60/40)	100 0.25/0.141	100 0.25/0.121	97.1 0.25/0.120	100 0.25/0.145	100 0.25/0.088	100 0.25/0.146	0.48
HI $\beta$ -lactamase pos. (11/40/64/28/15/15)	100 0.12/0.086	97.5 0.25/0.139	100 0.25/0.141	100 0.25/0.152	100 0.12/0.104	100 0.25/0.157	0.31

**Conclusions:** Tigecycline showed excellent *in vitro* activity against all *H. influenzae* isolates, with 100% of strains susceptible in the last 3 years, including all  $\beta$ -lactamase positive isolates. No trend over time toward increasing proportions of  $\beta$ -lactamase producers or increasing tigecycline MICs could be found.

## Introduction

Tigecycline has been shown to have potent broad spectrum activity against most commonly encountered respiratory pathogens. Tigecycline is a glycolcycline antimicrobial that inhibits protein synthesis by binding to the 30S ribosomal subunit. Tigecycline resistance is very infrequent and is also difficult to induce in the laboratory [1, 2]. This analysis was undertaken to assess whether there has indeed been no decrease in tigecycline activity against *H. influenzae* over the past 6 years, using isolates from Asia. The study is part of the Tigecycline Evaluation and Surveillance Trial (TEST), a large global surveillance study that has been monitoring antimicrobial susceptibility and resistance trends of tigecycline and comparator agents since 2004.

## Materials & Methods

- Isolates were derived mainly from respiratory tract (82%), head (e.g., otitis, conjunctivitis -- 10%), and blood (5%). Only one isolate per patient was accepted into the study. From 2005-2010, 566 clinical *H. influenzae* isolates were collected from 29 medical centers in eight Asian countries. This sample included 173  $\beta$ -lactamase positive strains (30.6%). Isolates were identified to the species level and tested at each site by the participating laboratory.
- Organism collection, transport, confirmation of organism identification, and development and management of a centralized database were coordinated by Laboratories International for Microbiology Studies (LIMS), a division of International Health Management Associates, Inc., located in Schaumburg, IL, USA.
- Minimum inhibitory concentrations (MICs) were determined by the Clinical and Laboratory Standards Institute (CLSI) recommended broth microdilution testing method [3]. Tigecycline was supplied by Pfizer, Inc. (Collegeville, PA, USA). All other agents were supplied by the panel manufacturers MicroScan (Siemens Medical Solutions Diagnostics., West Sacramento, CA, USA) and TREK (TREK Diagnostic Systems, Cleveland, OH). The following antimicrobial agents were included on the panels with their dilution ranges (expressed in mcg/ml): amikacin (0.5-64); amoxicillin-clavulanic acid (0.12/0.06-32/16); ampicillin (0.06-16); cefepime (0.5-32); ceftazidime (8-32); ceftriaxone (0.06-64); meropenem (0.12-16); levofloxacin (0.008-8); minocycline (0.5-16); piperacillin-tazobactam (0.06/4-128/4); tigecycline (0.008-16).
- Quality control (QC) of broth microdilution panels followed manufacturers and CLSI guidelines using *H. influenzae* ATCC 49247 and ATCC 49766. Results were included in the analysis only when corresponding QC isolates tested within the acceptable range according to CLSI guidelines [4].
- MIC interpretive criteria followed published breakpoints defined by CLSI [4] and the United States Food and Drug Administration (FDA) package insert for tigecycline [5].

## References

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## Acknowledgements

We gratefully acknowledge the contributions of the investigators, laboratory personnel, and all members of the Tigecycline Evaluation Surveillance Trial program group. This study was sponsored by Pfizer Inc.

## Results

Figure 1. Number of ALL *H. influenzae* and  $\beta$ -lactamase positive phenotypes in the study sample by country, 2005-2010.

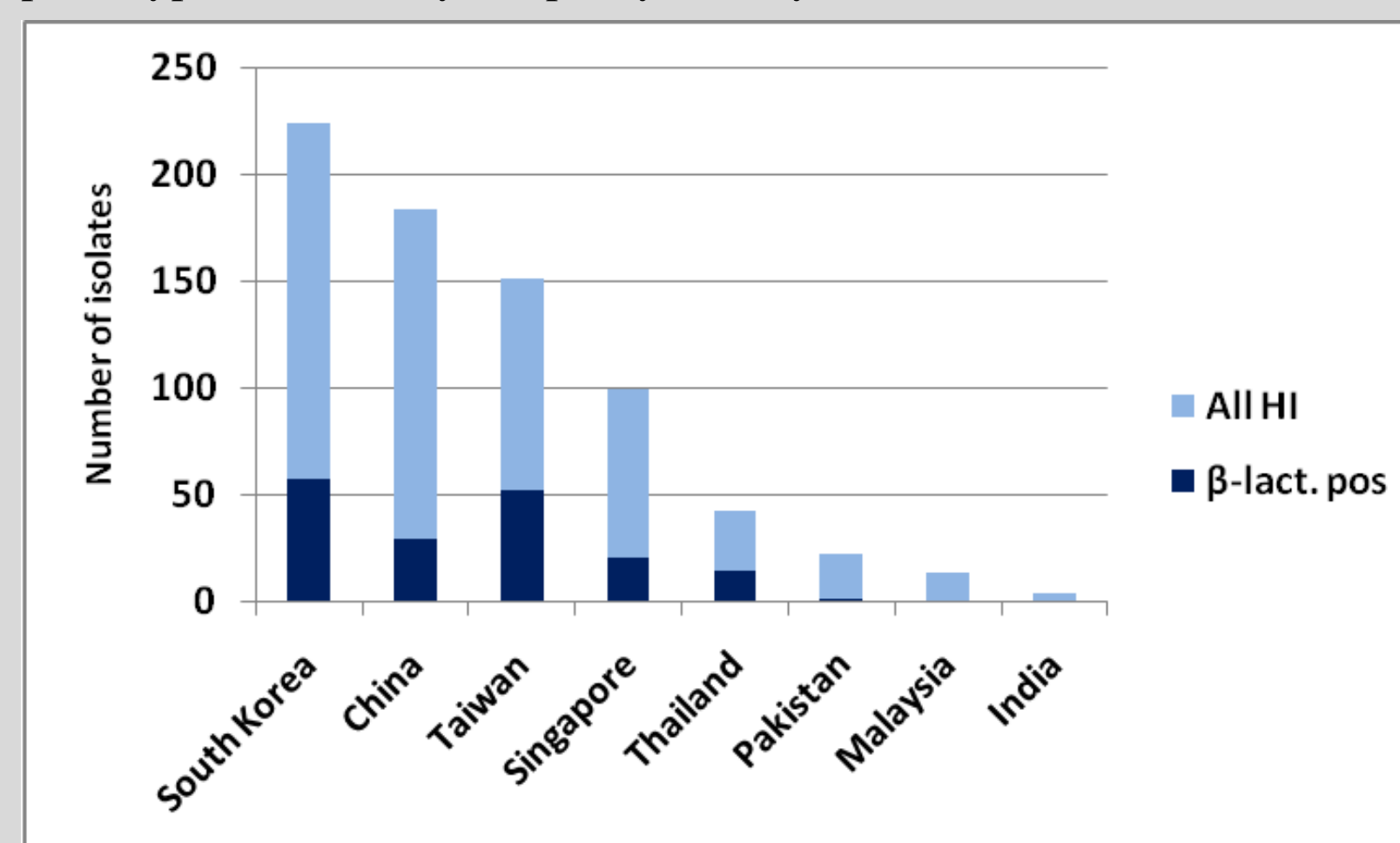


Figure 2. Proportion of *H. influenzae* that were  $\beta$ -lactamase positive, 2005-2010.

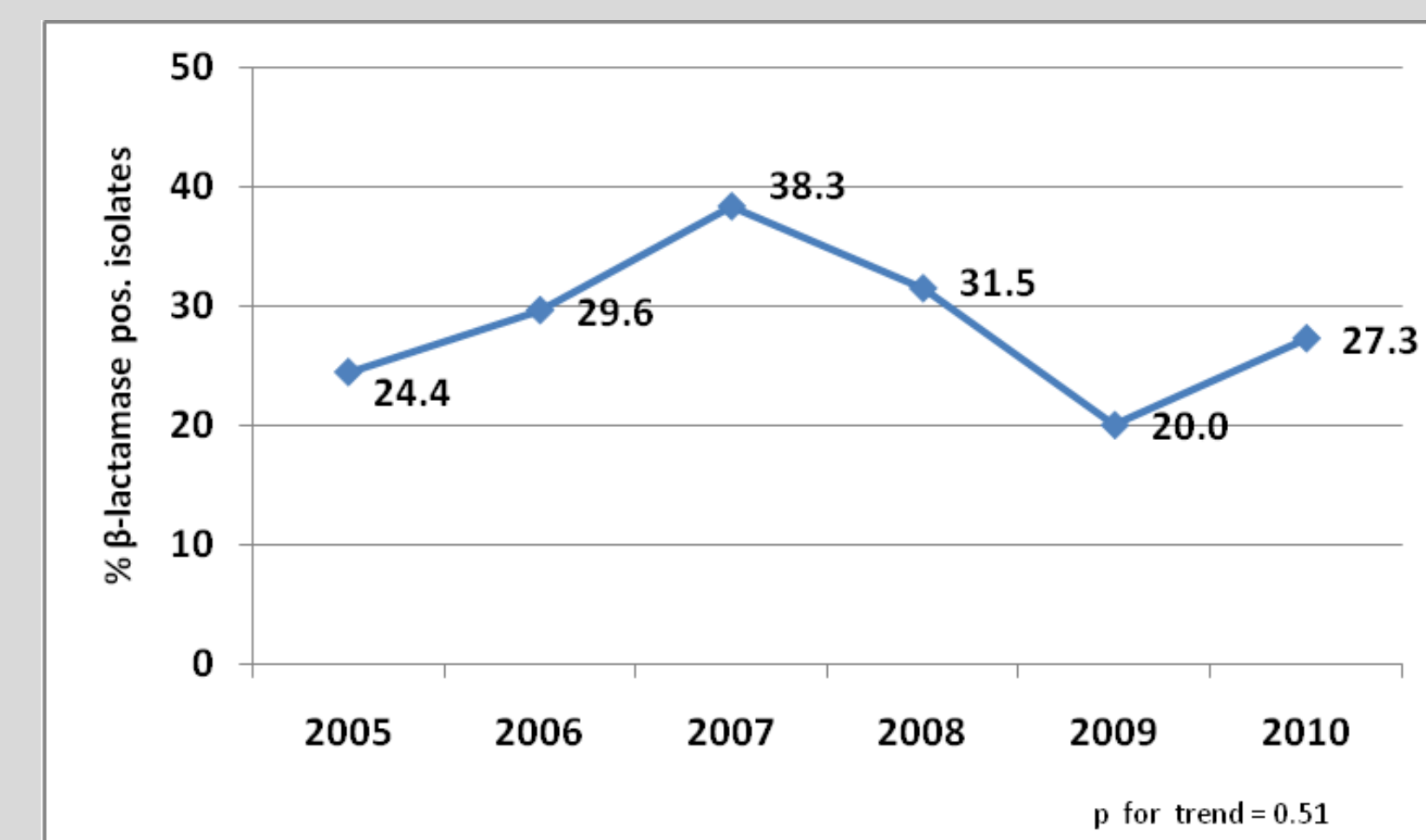
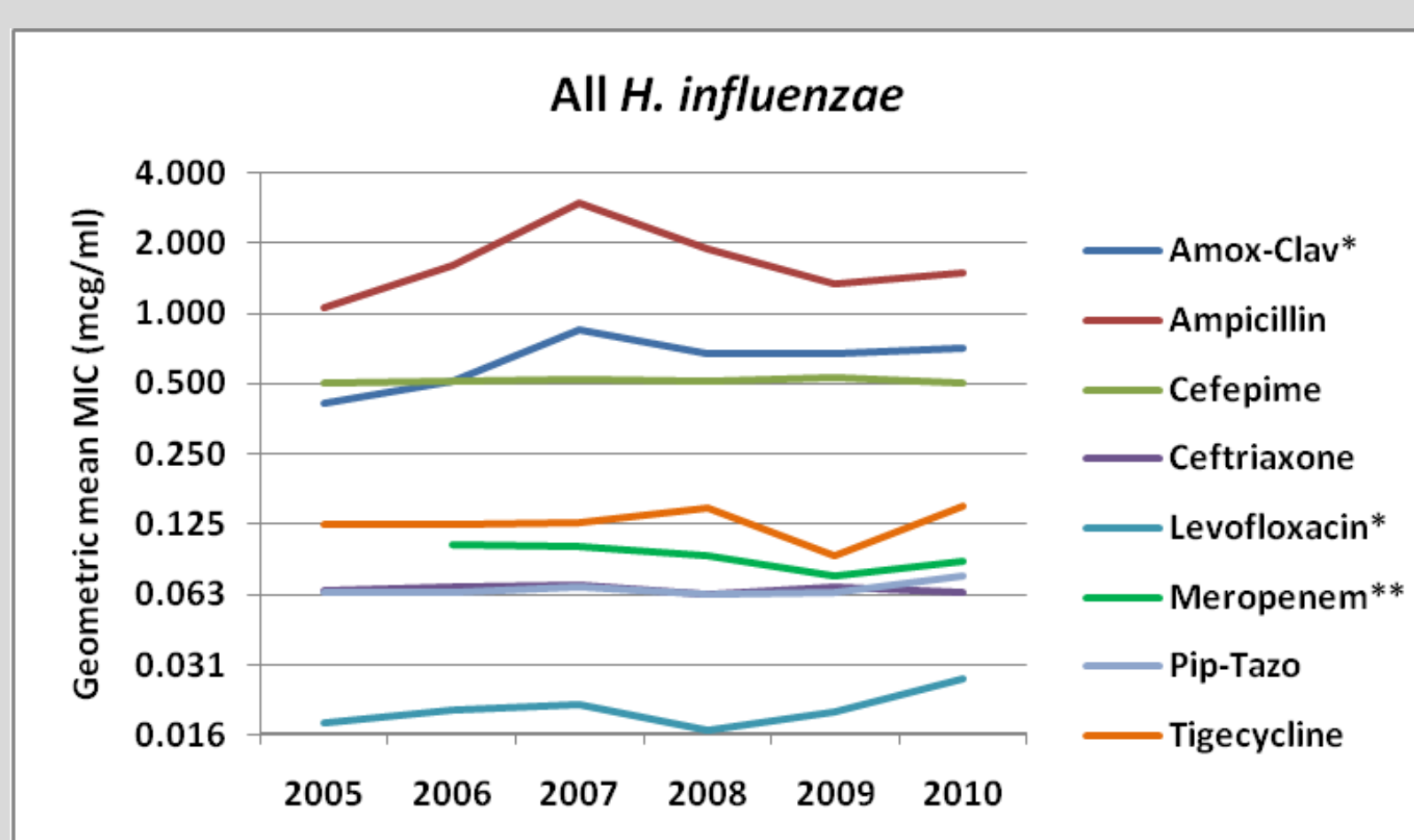


Table 1. *In vitro* activity of tigecycline and comparators, 2005-2010.

Species/phenotype (n per year) <sup>1</sup>		MIC <sub>90</sub> / % Susceptible					
		2005	2006	2007	2008	2009	2010
All <i>H. influenzae</i> (45/135/167/89/75/55)	Amox-Clav	1/100	2/100	2/98.2	2/100	2/98.7	2/96.4
	Ampicillin	16/75.6	>32/69.6	>32/56.3	>32/64.1	32/74.7	32/72.7
	Cefepime	≤0.5/100	≤0.5/99.3	≤0.5/100	≤0.5/100	≤0.5/100	≤0.5/100
	Ceftriaxone	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100
	Levofloxacin	0.06/100	0.06/99.3	0.06/95.8	0.015/100	0.06/98.7	0.12/100
	Meropenem	-- <sup>2</sup>	0.25/100	0.25/100	0.25/100	0.12/100	0.12/100
	Pip-Tazo	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/98.2
	Tigecycline	0.25/100	0.25/99.3	0.25/98.2	0.25/100	0.12/100	0.25/100
HI, $\beta$ -lactamase neg. (34/95/103/61/60/40)	Amox-Clav	0.5/100	1/100	2/100	2/100	2/100	1/100
	Ampicillin	≤0.5/100	≤0.5/99.0	1/91.3	1/93.4	1/93.3	≤0.5/100
	Cefepime	≤0.5/100	≤0.5/100	≤0.5/100	≤0.5/100	≤0.5/100	≤0.5/100
	Ceftriaxone	≤0.06/100	≤0.06/100	0.12/100	≤0.06/100	≤0.06/100	≤0.06/100
	Levofloxacin	0.06/100	0.06/100	0.03/99.0	0.06/100	0.03/98.3	0.12/100
	Meropenem	-- <sup>2</sup>	0.25/100	0.25/100	0.25/100	0.12/100	0.12/100
	Pip-Tazo	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/97.5
	Tigecycline	0.25/100	0.25/100	0.25/97.1	0.25/100	0.25/100	0.25/100
HI, $\beta$ -lactamase pos. (11/40/64/28/15/15)	Amox-Clav	1/100	2/100	4/95.3	4/100	4/93.3	8/86.7
	Ampicillin	32/0	>32/0	>32/0	>32/0	>32/0	>32/0
	Cefepime	≤0.5/100	≤0.5/97.5	≤0.5/100	≤0.5/100	≤0.5/100	≤0.5/100
	Ceftriaxone	0.12/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100
	Levofloxacin	0.03/100	0.03/97.5	0.5/90.6	0.015/100	0.06/100	0.5/100
	Meropenem	-- <sup>2</sup>	0.25/100	0.25/100	0.25/100	0.12/100	0.25/100
	Pip-Tazo	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100	≤0.06/100
	Tigecycline	0.12/100	0.25/97.5	0.25/100	0.25/100	0.12/100	0.25/100

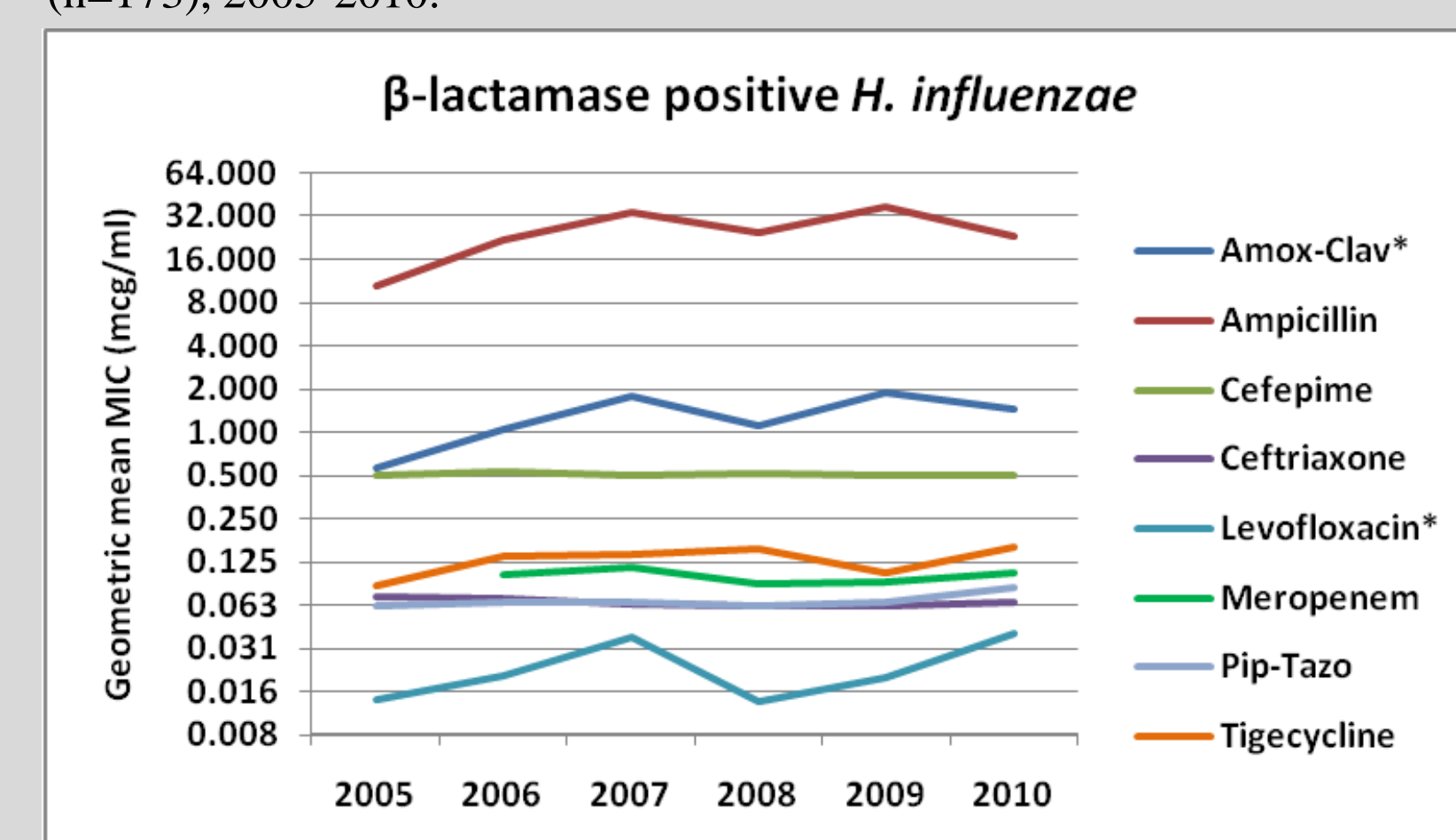
<sup>1</sup> n per year for meropenem: All HI 3/53/167/89/75/55, HI  $\beta$ -lactamase neg. 3/32/103/61/60/40, HI  $\beta$ -lactamase pos. 0/21/64/28/15/15  
<sup>2</sup> not reported since n<10

Figure 3. Geometric mean MIC for all *H. influenzae* (n=566), 2005-2010.



\*  $p$  for trend (increasing MICs) < 0.05; \*\*  $p$  for trend (decreasing MICs) < 0.05

Figure 4. Geometric mean MIC for  $\beta$ -lactamase positive *H. influenzae* (n=173), 2005-2010.



\*  $p$  for trend (increasing MICs) < 0.05

## Conclusions

- No trend over time toward increasing proportions of  $\beta$ -lactamase producing *H. influenzae* was found in Asia between 2005 and 2010.
- Tigecycline showed excellent *in vitro* activity, with 100% of *H. influenzae* susceptible in the last three years, including all  $\beta$ -lactamase positive isolates.
- No trend over time toward increasing tigecycline MICs could be found ( $p < 0.05$ ). Of the other agents tested, only amoxicillin-clavulanic acid and levofloxacin demonstrated a statistically significant increase in MICs between 2005 and 2010 both for *H. influenzae* overall and the  $\beta$ -lactamase producing subgroup ( $p < 0.05$ ), while meropenem showed a slight but statistically significant decreasing trend in MICs ( $p = 0.003$ ).