

## A Molecular and Comparative Study of Type-able and Non-type-able *Haemophilus influenzae* isolated from different clinical samples in Hilla, Iraq.

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### Abstract

<b>Background</b>	<i>H. influenzae</i> bacteria classified as type-able and non-type-able according to the presence or absence of capsule which is correlated with site of isolation.
<b>Objectives</b>	To isolate <i>H. influenzae</i> from different clinical samples and differentiate both capsulated (type-able) and non-capsulated (non-type-able) one by molecular detection method and to make a comparison between the two types by cultural, molecular, and clinical aspects.
<b>Methods</b>	A total of 220 clinical samples were aseptically taken from throat, ear, eye, sputum and CSF of patients attended three main hospitals in Hilla city, Iraq during the period from Feb. 2012 to Jun. 2012. All samples were subjected to bacterial cultivation, standard bacteriological method and molecular detection method. Other primers were used to detect the presence or absence of capsule using Bex A, Bex B, while p2 primer was used to detect non-type-able. Among capsulated one a specific primers (Hib, bex) targeting <i>H. influenzae</i> type b were used.
<b>Results</b>	29 (13.2%) out of 220 clinical samples give presumptive detection and isolation of <i>H. influenzae</i> , of these only 10 (34.5%) were positive using X (haemin) and V (Nad) tests. Using PCR, only 6 out of 10 (60%) were positive, out of 6 isolates only 2 (33.3%) were capsulated (type-able) and 4(66.6%) non-type-able, out of 2 capsulated 1 (50%) was type b <i>H. influenzae</i> .
<b>Conclusion</b>	Specific genetic marker should be used to detect both types. Many non-type-able <i>H. influenzae</i> isolates are also important cause of upper respiratory tract diseases including pharyngitis, otitis media, and conjunctivitis. Using serotype specific gene targeting type b (Hib) is important among patient with meningitis and lastly separation between type-able and non-type-able according to the site of isolation.
<b>Keyword</b>	<i>H. influenzae</i> , genetic marker, serotype specific gene, Hib, Bex( A, B) P6, NTHI, X,V factors

### Introduction

**H**aemophilus influenzae (*H. influenzae*) is a Gram negative coccobacillus whose environmental niche is primarily restricted to the human respiratory tract. The genus name reflects its absolute requirement for heme and the species name reflects the erroneous initial belief that this organism was the causative agent of influenzae<sup>(1)</sup>. Generally, it

is difficult and time-consuming to detect *H. influenzae* by using traditional method. So, PCR-based method targeting the outer membrane protein (OMP) represents specific diagnostic targets. OMP P6 is highly conserved among most strains of *H. influenzae*<sup>(2)</sup>. It is a peptidoglycan-associated lipoprotein constitutes 1-5% of all OMPs. Unlike P2 protein, P6 protein shown a very high homology (97%) in the amino acid