

**Bee propolis as a new modality for treatment of H1N1 Influenza**Ali Farid M.Ali<sup>1</sup>, Ermelando V.Cosmi<sup>2</sup>, Sanaa M. Ali<sup>3</sup> and LailaFarid<sup>4</sup><sup>1</sup>Heliopolis research Center, Cairo, Egypt<sup>2</sup> La Sapienza, Roma Italy<sup>3</sup>Faculty of Veterinary Medicine Kafr Elshiekh University, Egypt<sup>4</sup>Department of obstetrics and Gynecology, Faculty of Medicine, Ain Shams University Cairo Egypt[elshayb1950@yahoo.com](mailto:elshayb1950@yahoo.com)

**Abstract:** Influenza is a respiratory infection with significant morbidity and mortality. Drug which inhibits the uncoating process of influenza virus growth have been available for treatment of the virus there as common concern about the development of drug resistant virus strains hence the need of a new drug. Propolis is a natural product collected by bees from plant sources which is used to seal holes and repair many structures in the hive. The biological activities of propolis include antibacterial antifungal, antiportozoan, antitumoural and other therapeutic properties. The Aim of this work is to introduce Bee propolis for the first time in the literatures for treatment of H1N1 virus to compare between Bee propolis and oseltamivir regarding its therapeutic and adverse effect. One hundred mice (50 received oseltamivir, 75mg and 50 received Bee propolis 0.5 gm) *Plaque Assay*, Virus regrowth after removal of the compound (Bee propolis- Oseltamivir) and estimation of telomere length were performed. Plaque formation of influenza A/PR/8/34 virus was completely inhibited by 15mg/ml of bee propolis after 12h incubation, whereas visible plaque formation was detected in the plate treated with oseltamivir. In mice infected with a high challenge dose of influenza A/PR/8/34 virus orally administered 500 and 1gm/kg/day completely prevented the death of mice and the survival rate mice was significantly higher than those in mice treated with oseltamivir ( $P < 0.01$ ), the weights of mice were increased on bee propolis this effect is not seen with oseltamivir. ( $P < 0.01$ ), Cost Benefit ratio of Bee Propolis is less than that with oseltamivir., There is a statistically significant decrease in telomere length with oseltamivir ( $P < 0.01$ ). **Conclusion** Bee propolis is new modality of treatment of human influenza virus infection H1N1, with more positive results than oseltamivir.

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**Keywords:** Influenza virus H1N1, oseltamivir, Bee propolis, antiviral activity

**1. Introduction**

Influenza is a respiratory infection with significant morbidity and mortality<sup>(7)</sup>, oseltamivir<sup>(2)</sup> has been available for treatment of the virus there as common concern about the development of drug resistant virus strains hence the need of a new drug.

During any influenza season antigenic drift in the virus may occur after formulation of the year's vaccine has taken place, rendering the vaccine less protective, and outbreaks can more easily occur among high- risk populations, It is likely that vaccine would not be available for the first wave of spread of virus, So Antiviral agents are mandatory, **Propolis** is a natural product collected by bees from plant sources which is used to seal holes and repair many structures in the hive, The biological activities of propolis include antibacterial, antifungal, antiportozoal, **antiviral(3,4)**, antitumoural, **Immunomodulation, anti-inflammatory**, Caffeic acid phenethyl ester (CAPE) extracted from honey bee. Propolis is a biologically active ingredient of propolis with several interesting biological properties. Besides their well-known antioxidant

CAPE inhibits certain enzyme activities such as lipoxygenases, cyclooxygenase, glutathione S-transferase and xanthine oxidase, Caffeic acid phenethyl ester has also been reported to have antitumor activity, Anti-inflammatory properties, Apoptosis inducible functions, **inhibitory effects of HIV replication**<sup>(6)</sup> and antimetastatic activity We isolated for the first time in literature 10 compounds that had antiviral effect for influenza H1N1, This antiviral activity is due to preventing the binding of virions<sup>^</sup> to target cells by inhibiting proprotein convertase and marking infected cells for destruction by complement or T cells, Blocking the M2 ion channel, preventing virion uncoating and the release of genome segments in the cytoplasm, lowering intracellular GTP levels through the inhibition of inosine monophosphate dehydrogenase and by interfering with transcription and genome replication, it inhibits RNA dependent RNA polymerase, it blocks the translation of mRNA, markedly long half life of binding to the NA active site one day while with oseltamivir is 1.25 hours, it can be given in an Aerosolized form, it has anabolic

effect, No gastrointestinal effect, other advantages than its antiviral effect include antibacterial, antifungal, antiportozoal, **antiviral**, antitumoral, **Immunomodulation**, **anti-inflammatory**, So we conducted a study which is in accordance of international guidelines for use of laboratory animals experimentation, this study is aiming to introduce Bee propolis for the first time in the literatures for treatment of influenza H1N1 virus, and to compare between Bee propolis and oseltamivir regarding its therapeutic and adverse effect.

## 2. Material and methods

The components of this study includes Bee Propolis and oseltamivir, Cells, Virus, Plaque Assay (8,9), Determination of LD<sub>50</sub> of H<sub>1</sub>N<sub>1</sub> in mice group, Haemagglutination inhibition assay, Lung virus titer determination, *In vivo* toxicity determination, Evaluation of Antiviral effects in a prophylaxis model, Evaluation of antiviral effects in a therapeutic model, Telomere length (10) measured by fluorescence *insitu* hybridization (FISH) (Fig.1).

## 3. Results and Discussion

This study showed that Plaque formation of influenza A/PR/8/34 virus was completely inhibited by 15µg/ml of bee propolis after 72h incubation, whereas visible plaque formation was detected in the plate treated with oseltamivir 15µg/ml, The antiviral activity of Bee propolis was not influenced by an increase in multiplicity of infection (MOI) from 0.0001 to 1 but that of oseltamivir was influenced in a yield reduction assay, No increase in

viral yield was seen in either culture supernatant or cells after removal of bee propolis (15 µg /ml) but in contrast, production of infection recurred in culture, supernatant and in cells after removal of oseltamivir, In mice infected with a high challenge dose of influenza A/PR/8/34 virus orally administrated 1gm/kg/day completely prevented the death of mice and the survival rate mice was significantly higher than those in mice treated with oseltamivir  $P < 0.01$ , the weight of mice was increased on bee propolis this effect is not seen with oseltamivir.  $P < 0.01$ , Cost Benefit ratio of Bee Propolis is less than that with oseltamivir., There is a statistically significant decrease in telomere length with oseltamivir  $P < 0.01$ , short telomeres are more vulnerable to oxidative damage, In the prophylaxis model efficacy was demonstrated more in Bee propolis than oseltamivir  $P < 0.01$  in all disease parameters studied in these experiment including prevention of death, increase in MDD, reduction in the body weight decline, inhibition of lung consolidation and reduction of titer of virus recovered from the lung, In the therapeutic model oseltamivir is less effective than bee propolis against lethality when treatment began 24 hours after viral infection, Statistically significant inhibition of viral adsorption on to Red cells in bee propolis  $P < 0.01$ . suggesting that bee propolis was capable of directly interfering with viral hemagglutination to block binding to cellular receptors,

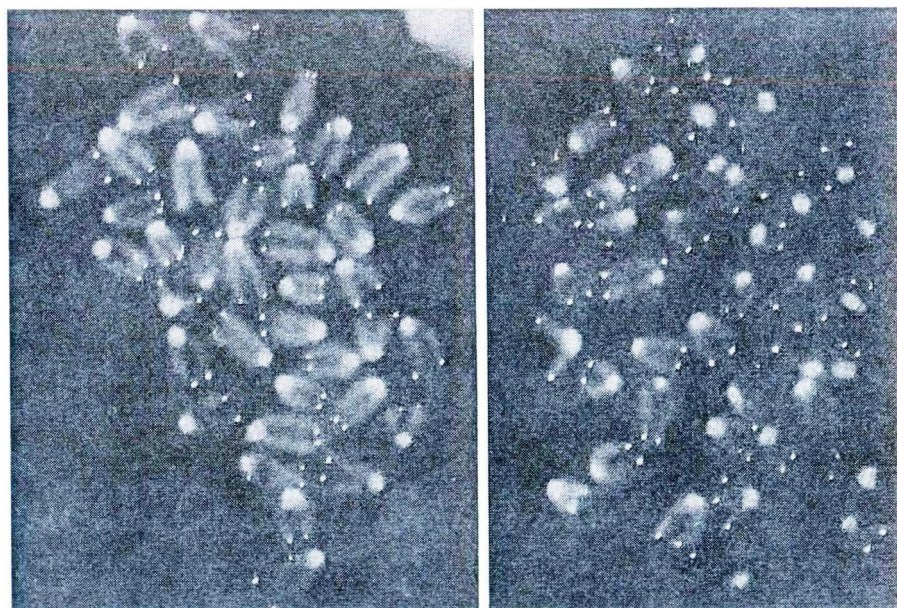


Figure Telomere length with  
bee propolis

Telomere length with  
oseltamivir

Figure 1.

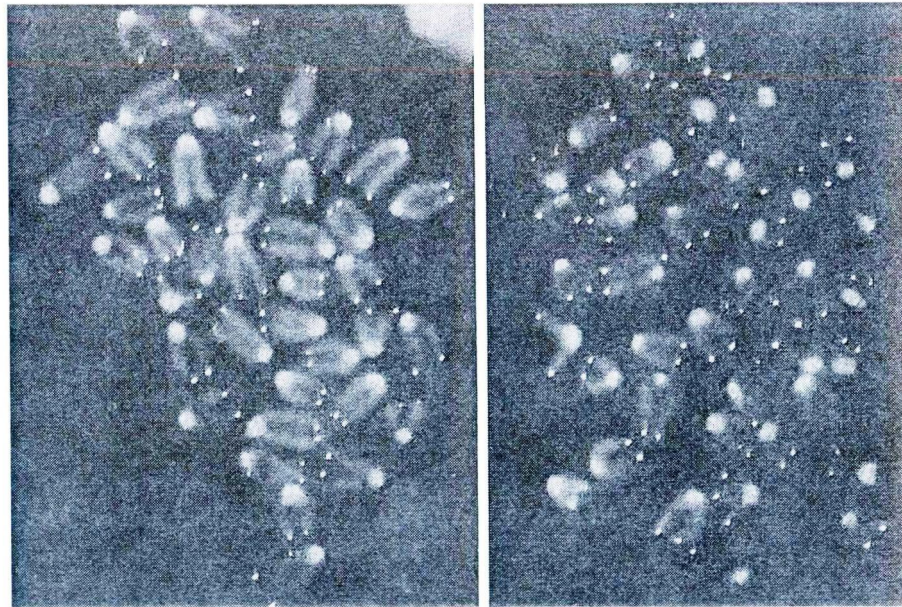


Figure Telomere length with  
bee propolis

Telomere length with  
oseltamivir

Figure 2.

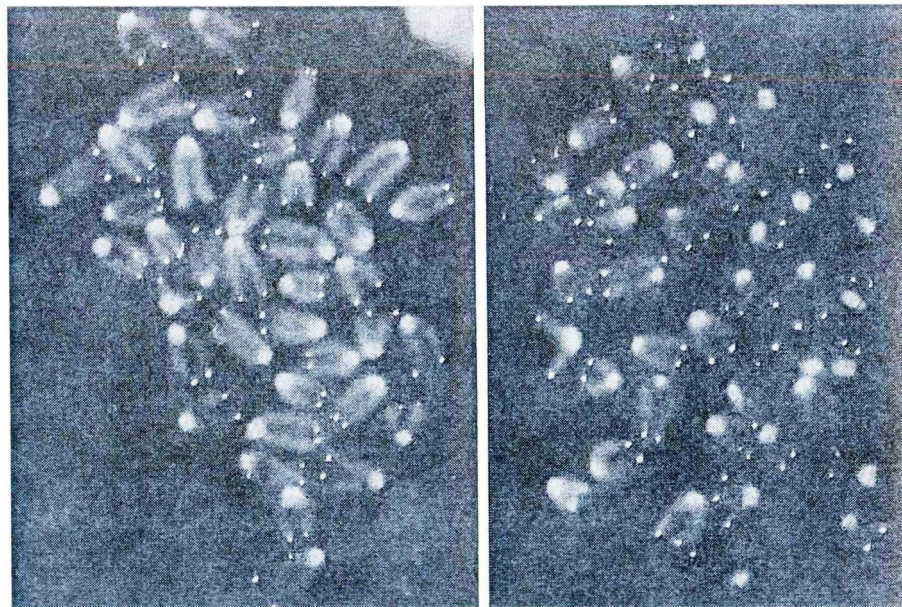


Figure Telomere length with  
bee propolis

Telomere length with  
oseltamivir

Figure 3.

## Conclusion

Bee propolis is a new modality for treatment of human influenza virus infection H1N1, with more positive results than oseltamivir, it has many action rather than its antiviral activity, it has about 10 compounds had antiviral activity these multiple mechanisms of action didn't induce drug resistance in addition it has many beneficial health effect than oseltamivir, it does not affect the telomere length.

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