Journal de Mycologie Médicale (2014) 24, e143—e148

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ORIGINAL ARTICLE/ARTICLE ORIGINAL

Immunomodulatory efficacy of ethanol extract of propolis on tumor-bearing mice with disseminated candidiasis

Efficacité immunomodulatrice de l'extrait éthanolique de propolis sur les souris porteuses de tumeurs avec une candidose disséminée

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Received 1 January 2014; received in revised form 26 April 2014; accepted 15 May 2014
Available online 22 October 2014

KEYWORDS
Disseminated candidiasis;
Candida albicans;
Cancer;
Propolis;
Immunomodulation

Summary
Objective. — This study was aimed at investigating the effect of propolis on immunosurveillance by measuring the levels of serum interleukin (IL)-4, IL-10, IL-17, tumor necrosis factor (TNF)-α and interferon (IFN)-γ in tumor-bearing mice with disseminated candidiasis.
Methods. — The ethanol extract of propolis was selected for this study. Balb/C female mice were infected with *Candida albicans* (*C. albicans*) and inoculated with spontaneous mouse mammary tumor (SMMT). The serum levels of tissue inhibitor of metalloproteinase-1(TIMP-1) were assessed by enzyme- linked immunosorbent assay (ELISA). Mice were treated daily with propolis solution (100 mg/kg, 0.1 mL, orally) for 3 days before IV challenge with *C. albicans* and SC challenge with SMMT and continued for 10 days. The rates of survival and tumor growth of understudy mice were investigated as well. The levels of TNF-α, IFN-γ, IL-4, IL-10 and IL-17 cytokines in culture supernatants were determined by ELISA.
Results. — The mean tumor size was significantly increased in tumor-bearing mice infected with *C. albicans* (16.98 ± 0.49 mm^2) as compared to other mice groups ($P < 0.05$). The results showed a significant decline of IL-4 and IL-10 levels after propolis administration to tumor-bearing mice infected with *C. albicans* (53.41 pg/mL, 156.81 pg/mL and 63.45 pg/mL) ($P < 0.05$). The increment of TNF-α (433.85 pg/mL) and IFN-γ (120.43 pg/mL) levels were also observed.

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http://dx.doi.org/10.1016/j.mycmed.2014.05.002

Conclusion. — Data revealed that propolis has remarkable immunomodulatory effect, which provides a scientific validation for the popular use of this natural substance, and further investigation will help to understand propolis usefulness during immunosuppressive conditions.

MOTS CLÉS
Candidose disséminée ;
Candida albicans ;
Cancer ;
Propolis ;
Immunomodulation

Résumé
Objectif. — Cette étude visait à étudier l'effet de propolis sur l'immunosurveillance en mesurant les niveaux sériques de l'interleukine (IL) -4, IL-10, IL-17, du facteur de nécrose tumorale (TNF)-α et de l'interféron (IFN)-γ de souris porteuses de tumeurs avec une candidose disséminée.
Matériel et méthodes. — L'extrait l'éthanolique de propolis a été sélectionné pour cette étude. Des souris femelles Balb/c étaient infectées avec *Candida albicans* (*C. albicans*) et inoculées avec une tumeur mammaire spontanée de souris (SMMT). Les taux sériques d'une métalloprotéinase-1 (TIMP-1) inhibiteur cellulaire ont été évalués par *enzyme-linked immunosorbent assay* (ELISA). Les souris ont été traitées quotidiennement avec la solution de propolis (100 mg/kg, 0,1 mL, oralement) pendant 3 jours avant l'injection IV avec *C. albicans* et SC avec la SMMT, traitement poursuivi pendant 10 jours après challenge. Les taux de survie et la croissance des tumeurs des souris ont été enregistrés. Les niveaux de TNF-α, IFN-γ, et des cytokines IL-4, IL-10, IL-17 dans les surnageants de culture ont été déterminés par la technique ELISA.
Résultats. — La taille de la tumeur a été augmentée de manière significative chez les souris porteuses de tumeurs et infectées par *C. albicans* ($16{,}98 \pm 0{,}49$ mm^2) par rapport aux autres groupes de souris ($p < 0{,}05$). Les résultats ont montré une diminution significative des niveaux de l'IL-4 et de l'IL-10 après administration de propolis aux souris porteuses de tumeurs et infectées par *C. albicans* (53,41 pg/mL, 156,81 pg/mL et 63,45 pg/mL) ($p < 0{,}05$). L'incrément des niveaux de TNF-α (433,85 pg/mL) et IFN-γ (120,43 pg/mL) a également été observé.
Conclusion. — Ces données ont révélé que la propolis a un remarquable effet immunomodulateur, et représentent une validation scientifique pour l'utilisation de cette substance naturelle. Des études complémentaires aideront à comprendre l'intérêt de la propolis lors de conditions immunosuppressives.

Introduction

Propolis has gained popularity in alternative medicine, and it is used in food and beverages to improve health and to prevent diseases. Propolis is a chemically complex bee product, containing material collected from buds or exudates of plants, volatile substances and wax [3]. It is composed of 30% wax, 50% resins and vegetable balsams, 10% essential oils, 5% pollen and other substances [5]. Because of its various bioactivities, including antibacterial [21], antiviral [9], antifungal [7], anti-inflammatory [28] and antitumoral [4,22] effects, propolis has been used in folk medicine and has attracted many investigators' interest. However, little is known about propolis effect on various cytokines production by immune cells. In vitro and in vivo assays demonstrated that propolis may activate macrophages, increasing their microbicidal activity and enhance the lytic activity of natural killer cells against tumor cells [20,26].

The polymorphic yeast *Candida albicans* (*C. albicans*) is an important opportunistic human pathogen causing infections that range from superficial mucosal lesions to life-threatening systemic disease. It is by far the most common cause of fungal invasive infections, which could be attributed to the little immunosuppression required to predispose an individual to invasive *Candida* infections [23]. Resistance to disseminated candidiasis requires the coordinated action of innate and adaptive immune defenses. Neutrophils and macrophages can clear the yeast via phagocytosis, and macrophage activation also leads to the release of several key mediators, which are important for protecting the host against disseminated candidiasis. Cytokines appear to play a major role, acting not only as modulators of antifungal effector functions but also as key regulators in the development of the different Th subsets from precursor Th cells. Studies in mice have shown that development of protective anticandidal Th1 responses requires the concerted actions of several cytokines, such as IFN-γ and TNF-α in the relative absence of inhibitory Th2 cytokines, such as IL-4 and IL-10, which inhibit development of Th1 responses [29]. The effect of natural products possessing immunomodulatory properties provided a potential therapeutic modality for reducing the risk of various diseases and cancers [14]. Several studies have indicated the role of immune responses in *C. albicans* infection [16,24,30], but few or no reports have addressed the dual effect of *Candida* infection and tumor development. Therefore, this study was aimed at developing an experimental model system for the evaluation of propolis on host — fungal pathogen interplay in *Candida*-infected tumor-bearing mice. We investigated the immunomodulatory effects of propolis by measuring the levels of IL-4, IL-10, IL-17, TNF-α and IFN-γ in culture supernatants, tumor size and TIMP-1 levels in mice understudy.