In recent decades a great attention has been paid for preparation of materials of small size and the technology. Synthesis of nanomaterials especially metal nanoparticles and their application in different field of biological sciences and medicine has provided new hope for development of nanodrugs and their effective delivery in the biological system [1]. In recent decade application of nanoparticles have been increasing due to their unique properties such as ultra-small size, large surface to volume ratio, high reactivity and unique interactions with structural components which improve the pharmacokinetics as well as therapeutic index of the drugs [2]. Among metals, application of silver and its compound in the field of medicine has been experiencing for over 2000 years and they fund nontoxic but safe bactericidal agent [3]. Recently several physical and chemical methods of synthesis of silver nanoparticles have been developed but biological methods such as using plant and fungal material are quite easy because of less expensive, safe and eco-friendly nature [4].

Thyroid hormones are directly associated with metabolic regulation of body and modulate a lot of physiological status of health such as body weight, oxygen requirement, normal growth and development during childhood [5] and patho-physiological dysfunction of thyroid gland is the most common endocrine disorder and in India, and about 42 million people suffer from thyroid associated diseases [6]. It has also been reported that serum cholesterol level increase in hypothyroidism and associated diseases and enhance the risks of coronary artery disease, atherosclerosis and heart failure [7].

Mushrooms of Pleurotus genus are popularly consumed by the peoples of all over the world due to their high nutritional values and some medicinal properties. The Pleurotus mushrooms are rich in proteins, essential amino acids, polysaccharides and essential fatty acids, dietary fibers, minerals,

### IMPACT OF *Pleurotus tuber-regium* (Rumph. ex Fr.) EXTRACT-LOADED NANOPARTICLES ON THYROID PROFILE OF RATS

**Sukumar Dandapat**

Department of Zoology, Ranchi University, Jharkhand, India

**Manoj Kumar**

Department of Zoology, St. Xavier’s College, Jharkhand, India

**Rohit Srivastava**

Department of Zoology, Ranchi University, Jharkhand, India

**Manoranjan Prasad Sinha**

Department of Zoology, Ranchi University, Jharkhand, India

E-mail: dr.sukumar2018@gmail.com

**Aim.** The aim of this study was to investigate the impact of *Pleurotus tuber-regium* extract loaded silver nanoparticles (SNPs) on thyroid profile of rats.

**Methods.** Acute toxicity test was performed following up and down procedure and the analysis of thyroid profile was performed following chemiluminescence assay method.

**Results.** Acute toxicity test showed no mortality and no behavioral changes in rats treated with 2000 mg kg$^{-1}$ of SNPs. High dose (400 mg kg$^{-1}$) of SNPs showed high hyper thyroid activity by increasing T3 = 38.20±4.06 ng dL$^{-1}$; T4 = 4.40±0.32 μg dL$^{-1}$; and by decreasing TSH = 0.73±0.06 μIU mL$^{-1}$ compared to control group (T3 = 30.20±1.02 ng dL$^{-1}$; T4 = 2.94±0.08 μg dL$^{-1}$; TSH = 0.87±0.01 μIU mL$^{-1}$) and animal group treated with 200 mg kg$^{-1}$ dose of SNPs (T3 = 35.69±1.13 ng dL$^{-1}$; T4 = 3.82±0.24 μg dL$^{-1}$; TSH = 0.80±0.02 μIU mL$^{-1}$).

**Conclusions.** *Pleurotus tuber-regium* extract loaded silver nanoparticles are non toxic and can be used in drug delivery as well as in the formulation of drugs used in hypothyroidism.

**Key words:** Pleurotus tuber-regium; silver nanoparticles; thyroid profile.
some vitamins etc. [8]. *Pluortus tuber-regium* has been used as medicinal supplement for headache, stomach ailments, cold and fever, asthma, small pox, high blood pressure as well as for weight gain and malnourishment [9]. The aim of this work was to synthesis silver nanoparticles loaded with aqueous extract of *P. tuber-regium* and to study their impacts of extract loaded silver nanoparticles on profile of rat because the impacts of silver nanoparticles loaded with extract of *P. tuber-regium* has not been explored till dated.

**Materials and Methods**

**Study of impact of silver nanoparticles on thyroid profile on rat**

*Animals:* Wistar albino rats of 175 to 200 g were obtained from the National Institute of Nutrition, Hyderabad, India. They were kept in cage and maintained under standard laboratory conditions at ambient room temperature (22 ± 3 °C) and relative humidity (30–65%), with dark-light cycle of 12 h for 5 days. 15 rats were fed with a commercial pallet diet (Sadguru Shri Shri Industries Pvt. Ltd. Pune, India) and water. The experiment was performed after prior approval of the Ethics committee of Ranchi University, Ranchi (Proceeding no. 46, page no. 137). Acute toxicity study of *P. tuber-regium* extract loaded SNPs on rat was also done according to OECD [10] test guideline 425 (Up and Down procedure), limited test was performed for at the test dose 2000 mg kg⁻¹ on rats and dose was fed according to the body weight.

For the study of impacts of *P. tuber-regium* extract loaded SNPs on thyroid profile of rats seven days experimental period was chosen followed previous method of Garba et al [11]. For the treatment fifteen rats were distributed into three groups and each group contain 5 rats. Two doses (high dose: 400 mg kg⁻¹ and low dose: 200 mg kg⁻¹) of SNPs (average 71.36 nm diameter with −11.2 mV zeta potential) were taken and the doses were feed according to the body weight of the animals [12, 13]. The experiment designed for the study is described below:

**Group-1:** Rats were served as control and they received single dose (1 mL) of distilled water (vehicle) daily orally for 7 days.

**Group 2:** Rats of this group received daily single dose (200 mg kg⁻¹) of nanoparticles orally for 7 days.

**Group 3:** Rats of this group were received daily single dose (400 mg kg⁻¹) of nanoparticles orally for 7 days.

**Analysis of Thyroid Hormone**

Study the impacts of *P. tuber-regium* extract loaded SNPs on thyroid profile of rats were done by chemiluminescence immune assay [14].

**Statistical analysis**

Entire statistical works were done using full proof statistical software WinSTAT (R. Fitch Software, Canal Park, Cambridge, Massachusetts, USA). Data were taken n = 5 and results were expressed as a mean ± standard error of mean. Statistical analysis was performed by one-way ANOVA with post-hoc Student’s t-test, *P* ≤ 0.05 was considered as statistically significant.

**Results and Discussion**

Although *P. tuber-regium* have valuable pharmacological effects and lack toxicity. Therefore, the current study was conducted to assess the acute toxicity of synthesized SNPs loaded with *P. tuber-regium* by following OECD guidelines 425 [10] before study of impact of *P. tuber-regium* extract mediated nanoparticles on thyroid profile of rats. It has been reported that, as the acute oral toxicity study is necessary to determine the safer dose range to manage the clinical signs and symptoms of the drugs [15] and the toxic outcomes of drugs such as decrease body weight, clinical signs and symptoms which are principal observations among various toxicity indicators [16]. In the present extract and SNPs did not show deleterious acute toxicity symptoms such as increased somatomotor activity, convulsion, tremor and itching etc. after till 14 days.

The result of impacts of SNPs loaded with *P. tuber-regium* extract on thyroid profile of rats is presented in the Table. The result shows both low and high doses of SNPs act on the thyroid-pituitary axis and significantly increases T3 and T4 level and decreases TSH level compared to control group. T3 most active form of thyroid hormone than T4, and both are synthesized in the thyroid follicles in low and high amount respectively. T4 is synthesized by monoiodination of T3 in liver [17]. Serum TSH level is inversely correlated with serum T3 and T4 level [18]. It has been reported in hypothyroidism significant decrease in serum T3 and T4 level occurs with increase in TSH level [19]. Sublethal exposure of silver oxide nanoparticles lack capping agent of bioactive substances from medicinal plants or fungi,
Impact of silver nanoparticles loaded with P. tuber-regium extract on thyroid profile of rat

| Treatment groups | Parameters |
|------------------|------------|
|                  | T3 (ng dL\(^{-1}\)) | T4 (μg dL\(^{-1}\)) | TSH (μIU mL\(^{-1}\)) |
| Group-1          | 30.20 ± 1.02    | 2.94 ± 0.08    | 0.87 ± 0.01      |
| Group-2          | 35.69 ± 1.13\(^{*}\) | 3.82 ± 0.24\(^{*}\) | 0.80 ± 0.02\(^{*}\) |
| Group-3          | 38.20 ± 4.06\(^{*}\) | 4.40 ± 0.32\(^{*}\) | 0.73 ± 0.06\(^{*}\) |

Note: “\(^{*}\) — \(P \leq 0.05\) when compared the values of Group-2 (Low dose) and Group-3 (High dose) with Group-1 (Control).

Conclusions

P. tuber-regium extract can be used for synthesis of silver nanoparticles and the synthesized nanoparticles loaded with P. tuber-regium extract showed increased thyroid level. Thus, P. tuber-regium extract loaded nanoparticles can be used for the formulation of pharmaceuticals related to hypothyroidism.

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ВПЛИВ НАНОЧАСТИНОК, НАВАНТАЖЕНИХ ЕКСТРАКТОМ Pleurotus tuber-regium (Rumph. ex Fr.), НА ТИРЕОІДНИЙ ПРОФІЛЬ ЩУРІВ

Sukumar Dandapat¹
Manoj Kumar²
Rohit Srivastava¹
Manoranjan Prasad Sinha¹

¹Кафедра зоології, Університет Ранчі, Джаркханд, Індія
²Кафедра зоології, Колледж Св. Ксавьера, Джаркханд, Індія

E-mail: dr.sukumar2018@gmail.com

Мета. Метою роботи було з’ясувати вплив екстракту Pleurotus tuber-regium, який містить наночастинки срібла (SNP), на профіль щитоподібної залози щурів.

Методи. Тест на гостру токсичність проводили після процедури впливу екстрактом у мінімальній та максимальній кількості, а аналіз профілю щитоподібної залози здійснювали методом гемілюмінесцентного аналізу.

Результати. Тест на гостру токсичність показав відсутність смертності та зміни у поведінці щурів, що отримали 2000 мг/кг НЧС. Висока доза (400 мг/кг) НЧС показала високу гіпертироїдну активність за рахунок збільшення T₃ = 38,20 ± 4,06 нг дл⁻¹; T₄ = 4,40 ± 0,32 мкг дл⁻¹; і за рахунок зниження ТТГ = 0,73 ± 0,06 мкМЕ/мл порівняно з контрольною груповою (T₃ = 30,20 ± 1,02 нг дл⁻¹; T₄ = 2,94 ± 0,08 мкг дл⁻¹; T₄ = 0,87 ± 0,01 мкМЕ/мл) та групою тварин, що отримали дозу НЧС у 200 мг/кг (T₃ = 35,69 ± 1,13 нг дл⁻¹; T₄ = 3,82 ± 0,24 мкг дл⁻¹; ТТГ = 0,80 ± 0,02 мкМЕ/мл).

Висновки. Екстракт P. tuber-regium, що містить наночастинки срібла, нетоксичний, і його можна використовувати для виготовлення лікарських засобів, які застосовують при гіпотиреозі.

Ключові слова: Pleurotus tuber-regium; наночастинки срібла; профіль щитоподібної залози.

ВПЛЯННЯ НАНОЧАСТИЦ, НАГРУЖЕНИХ ЕКСТРАКТОМ Pleurotus tuber-regium (Rumph. ex Fr.), НА ТИРЕОІДНИЙ ПРОФІЛЬ КРЫС

Sukumar Dandapat¹
Manoj Kumar²
Rohit Srivastava¹
Manoranjan Prasad Sinha¹

¹Кафедра зоологии, Университет Ранчи, Индия
²Кафедра зоологии, Колледж Св. Ксавьера, Джаркханд, Индия

E-mail: dr.sukumar2018@gmail.com

Цель. Целью работы было изучение влияния экстракта Pleurotus tuber-regium, содержащего наночастицы серебра (НЧС), на профиль щитовидной железы крыс.

Методы. Тест на острую токсичность выполняли после процедуры воздействия экстрактом в минимальном и максимальном количестве, а анализ профиля щитовидной железы выполняли методом хемилюминесцентного анализа.

Результаты. Тест на острую токсичность показал отсутствие смертности и изменений в поведении крыс, получавших 2000 мг/кг НЧС. Высокая доза (400 мг/кг) НЧС показала высокую гипертироидную активность за счет увеличения T₃ = 38,20 ± 4,06 нг дл⁻¹; T₄ = 4,40 ± 0,32 мкг дл⁻¹; за счет снижения ТТГ = 0,73 ± 0,06 мкМЕ/мл по сравнению с контрольной группой (T₃ = 30,20 ± 1,02 нг дл⁻¹; T₄ = 2,94 ± 0,08 мкг дл⁻¹; T₄ = 0,87 ± 0,01 мкМЕ/мл) и группой животных, получивших дозу НЧС в 200 мг/кг (T₃ = 35,69 ± 1,13 нг дл⁻¹; T₄ = 3,82 ± 0,24 мкг дл⁻¹; TSH = 0,80 ± 0,02 мкМЕ/мл).

Выводы. Наночастицы серебра, содержащие экстракт P. tuber-regium, нетоксичны, и их можно использовать для приготовления лекарственных средств, применяемых при гипотиреозе.

Ключевые слова: Pleurotus tuber-regium; наночастицы серебра; профиль щитовидной железы.