

원 저

글라이포세이트의 유전자 독성에 대한 멜라토닌의 유전자 보호 효과

계명대학교 의과대학 동산의료원 응급의학교실¹, 계명대학교 의과대학 해부학교실²

김정규¹ · 최우익¹ · 이재호² · 최인장² · 진상찬¹

Genoprotective Effect of Melatonin Against to the Genotoxicity of Glyphosate on Human Blood Lymphocytes

Jung-Gyu Kim, M.D.¹, Woo-Ik Choi, M.D.¹, Jae-Ho Lee, M.D.²,
In-Jang Choi, M.D.², Sang-Chan Jin, M.D.¹

Department of Emergency Medicine, School of Medicine, Keimyung University, Dongsan Medical Center, Daegu¹,

Department of Anatomy, College of Medicine, Keimyung University, Daegu², Korea

Purpose: Glyphosate is a widely used non-selective herbicide. Previous studies have shown that glyphosate has genotoxicity, and that even low-doses of glyphosate can cause DNA damage. Melatonin is a hormone produced and secreted by the pineal gland that is known to be a potent anti-carcinogen, anti-oxidant, and genetic protector. This study was conducted to investigate the genoprotective effect of melatonin against glyphosate in human blood lymphocytes.

Methods: Human peripheral blood was obtained from 15 young, healthy volunteers and cultured under four different toxicologic conditions. The four groups consisted of a control group, glyphosate only group (300 ng/mL), glyphosate with low level of melatonin group (50 μ M), and glyphosate with high level of melatonin group (200 μ M). The mean Sister Chromatid Exchange (SCE) frequency of each group was then analyzed.

Results: Glyphosate exposed groups had a higher mean SCE frequency (10.33 ± 2.50) than the control group (6.78 ± 2.31 , $p < 0.001$). Interestingly, the group that received a low-level of melatonin had a lower mean SCE frequency (8.67 ± 2.58) than the glyphosate-only group, while the group that received a high level of melatonin had a much lower mean SCE frequency (8.06 ± 2.50) than the glyphosate-only group. There was statistical significance.

Conclusion: Melatonin exerted a potent gene protective effect against the genotoxicity of glyphosate on human blood lymphocytes in a dose-dependent fashion.

Key Words: Glyphosate, Genotoxic, Sister chromatid exchange, Melatonin

Introduction

Glyphosate is widely used nonselective herbicide for both agricultural and non-agricultural purpose. The chemical name of glyphosate is 'N-(Phosphonomethyl) glycine'. It was discovered to be an herbicide by Monsanto chemist John Franz in 1970. Many previous studies about the safety of glyphosate formulation have concluded that there is little toxicity to humans. Past review concluded that there is no

책임저자: 진 상 찬

대구광역시 중구 달성로 56

계명대학교 동산의료원 응급의학과

Tel: 053) 250-7610, Fax: 053) 250-7028

E-mail: jchan98@hanmail.net

투고일: 2016년 9월 22일

1차 심사일: 2016년 9월 27일

게재 승인일: 2016년 10월 11일

potential for Roundup® (the registered trademark of Monsanto, One of the most widely used glyphosate commercial formulation) herbicide to pose a health risk to humans¹⁾. According to more extensive use of glyphosate, glyphosate intoxicated patients visited emergency room more frequently. However, recent studies showed a harmful effect of glyphosate variously. Especially, their harmful effect is vary depending upon absorption route. Small amount of oral intake may cause nausea, vomiting, and diarrhea, however, large amount of ingestion may cause severe systemic effects. Severe systemic symptoms may occur from cardiotoxicity, hepatotoxicity, renal toxicity, non-cardiogenic pulmonary edema, mental change, metabolic acidosis and even to cardiac arrest and death. And inhalation exposure may cause airway discomfort, laryngeal burn, and acute pneumonitis²⁾. But most of those symptoms are acute clinical chief complaints. Glyphosate has much toxicity in addition to acute clinical chief complaints. Glyphosate herbicide is potential endocrine disruptor to human. And cytotoxicity of glyphosate herbicide on placental cells could induce reproductive disability³⁾. Moreover, glyphosate herbicide induces the breakage of DNA strands and chromosomal damage⁴⁾. Previous study showed that glyphosate has the genotoxicity by sister chromatid exchange (SCE) test, and even a low-dose of glyphosate may cause DNA damage⁵⁾. Several studies about glyphosate supported that glyphosate has a negative effect for human⁶⁻⁸⁾. Glyphosate may have various toxicities but antidote does not exist. So, conservative management is almost only treatment in acute intoxicated patient. And there are no preventable agent for genotoxicity and carcinogenicity of glyphosate.

Melatonin, chemically N-acetyl-5-methoxytryptamine, is a manmade form of hormone produced and secreted by the brain, exactly pineal gland. Known effects of melatonin is regulating circadian rhythm of physiological functions including bedtime, seasonal reproduction, blood pressure, core temperature, and so on^{9,10)}. In addition to widely known effect above, melatonin has many beneficial effects physiologically. Melatonin increases activity of anti-oxidative

enzymes and scavenges free radicals as an antioxidant^{11,12)}. And, melatonin has been known potent anti-carcinogen. Many studies were performed about anti-carcinogenicity of melatonin through various methods. These studies showed oncostatic action or even tumor size reduction effect of melatonin for breast cancer, colon cancer, cervicovaginal cancer, and hepatocellular cancer¹³⁻¹⁷⁾. Randomized controlled trial demonstrated that melatonin reduced the risk of death at 1 year in solid cancer patients¹⁸⁾. Melatonin is also potent genetic protector. Previous study proved that melatonin reduced DNA damage induced by gamma radiation¹⁹⁾. And melatonin reduced genotoxic effects by hypoxia both in vitro and in vivo²⁰⁾. Anisimov et al.²¹⁾ showed that melatonin protected the cells from DNA damage, not only by oxidative mutagens, but also by different alkylating agents.

It is difficult to know the detail mechanisms of both genotoxic effect of glyphosate and genetic protective effect of melatonin. This study was done to clarify protective effect of melatonin in human blood lymphocyte exposed to genotoxicity of glyphosate by SCE frequency method.

Methods

1. Chemicals

Roundup UltraMax® (Monsanto, Roseville, CA, USA) was used as representative product of glyphosate herbicide. It contains 570 gram of active ingredient glyphosate in 1 liter. And Melatonin made by Sigma-Aldrich (Saint Louis, MO, USA) was used. Phytohemagglutinin (PHA, Gibco, Waltham, MA, USA) as mitogen, RPMI 1640 (Gibco, Waltham, MA, USA) as culture medium, fetal bovine serum (Gibco, Waltham, MA, USA) as growth supplement and colcemid (Gibco, Waltham, MA, USA) were used. Bromodeoxyuridine (BrdU, Sigma-Aldrich, Saint Louis, MO, USA) was also used.

2. Blood Sampling

Peripheral blood of young and healthy volunteers