

A glass of milk and a bowl of cottage cheese.

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## ASSESSMENT OF 17B-ESTRADIOL CONTENT IN MILK AND DAIRY PRODUCTS AS A SAFETY CRITERION

MONOGRAPH



**MINISTRY OF EDUCATION AND SCIENCE OF UKRAINE  
HIGHER EDUCATIONAL INSTITUTION  
«PODILLIA STATE UNIVERSITY»**

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**Kamianets-Podilskyi– 2025**

**UDC 637.074**

Recommended for publication by the academic council of the Higher educational institution «Podillia State University» (Protocol No. 6 dated 29 May 2025)

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**Assessment** of  $17\beta$ -estradiol content in milk and dairy products as a safety criterion : monograph. / KOCHETOVA H., KUKHTYN M., HORIUK Y., SALATA V. Kamianets-Podilskyi : HEI «PSU». PE Zvoleiko D.H., 2025. 96 p.

ISBN 978-617-620-368-1

The monograph investigated the content of  $17\beta$ -estradiol in milk and its impact on the safety of dairy products. It was found that the concentration of this hormone depends on the fat content of the milk, the lactation period, and the stage of pregnancy of cows. Milk with higher fat content contains more  $17\beta$ -estradiol, and thermal treatment does not significantly affect its level. The lack of control over this hormone in milk in Ukraine highlights the need for regulatory standards to ensure consumer safety.

**UDC 637.074**

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**ISBN 978-617-620-368-1**

## ABSTRACT

Milk safety assessment is an essential task to ensure that the population has access to nutritious, high-quality dairy products. The consumption of dairy products containing high levels of estrogens raises concerns among scientists from various fields because of the direct relationship between chronic exposure to estrogenic hormones and the prevalence of various types of cancer. Among steroid hormones,  $17\beta$ -estradiol is the most biologically active. According to Codex Alimentarius Commission requirements, the maximum amount of external estradiol that enters the body through food should not exceed 50,000 pg/kg/day. An analysis of Ukrainian regulatory documents revealed that there is no control over the  $17\beta$ -estradiol content in raw milk during its processing. However, milk and dairy products contribute approximately 60-80% of all dietary estrogens. Given the significant impact of high estrogen concentrations on the consumer's body, the current regulatory standards for  $17\beta$ -estradiol in milk and dairy products, considering the intensive milk production technologies, are not fully substantiated. Therefore, conducting systematic research, considering the influence of multiple factors on the level of  $17\beta$ -estradiol in raw milk, is promising and relevant. This will provide a scientific basis for determining the maximum allowable concentration of this natural hormone in raw milk during its transfer for processing.

The study aims to determine the content of  $17\beta$ -estradiol in cow milk and develop a methodology for defining and evaluating the maximum allowable concentration of this hormone in raw milk during the acceptance for processing. It was established that the average content of  $17\beta$ -estradiol in pooled raw milk obtained within a day at a single farm does not depend on the time of milk collection. At the same time, the content of  $17\beta$ -estradiol in milk collected from a single herd undergoes significant changes over the course of the year. Mixing milk from different farms at a processing plant does not lead to a significant difference in the  $17\beta$ -estradiol content compared to milk obtained at a specific farm in the region. It was found that milk with a higher fat content contains more  $17\beta$ -estradiol. The milk separation process affects the distribution of  $17\beta$ -estradiol between cream and skim milk. The amount of  $17\beta$ -estradiol in skim milk was nearly 5-7 times lower than in cream.

The study also examined changes in  $17\beta$ -estradiol content during the pregnancy period of cows, showing that estrogen hormone concentration gradually increased, with significant changes ( $p < 0.05$ ) compared to the first month of pregnancy. Specifically, during the first three months of pregnancy, the amount of  $17\beta$ -estradiol did not exceed 100 pg/ml, with an average range of  $42.4 \pm 7.7$  to

68.3±7.8 pg/ml. Starting from the fourth month of pregnancy, the steroid hormone level increased on average to 139.4±11.8 pg/ml, which was 3.2, 2.7, and 2.0 times higher ( $p<0.05$ ) than during the first, second, and third months of pregnancy, respectively. The growth in 17 $\beta$ -estradiol concentration continued to rise significantly in the following months of pregnancy. By the fifth month, the hormone level was 497.3±36.7 pg/ml, which was 3.5 times ( $p<0.05$ ) higher than in the fourth month and nearly 10 times higher ( $p<0.05$ ) than in the first month of pregnancy. The maximum levels of 17 $\beta$ -estradiol were observed during the late stages of pregnancy, with the highest concentrations recorded at the end of the seventh and beginning of the eighth months of pregnancy, reaching 1105.3±78.5 and 1209.8±82.4 pg/ml, respectively.

The research on the content of 17 $\beta$ -estradiol in milk during the estrous cycle of cows showed that the lowest concentration of the hormone was found during the first seven days of the cycle, ranging from 57.1 to 65.6 pg/ml. From the 15th to the 19th day of the estrous cycle, the amount of 17 $\beta$ -estradiol ranged from 365.5 to 391.3 pg/ml, with the highest concentration of 17 $\beta$ -estradiol recorded on the 19th day of the cycle, reaching 407.3±39.5 pg/ml. Thus, milk produced at the end of lactation and during the final stages of the estrous cycle serves as a significant source of estrogenic hormones, which enter the human body through dairy products.

Further analysis of 17 $\beta$ -estradiol content in different dairy products revealed a correlation between increased hormone concentration and higher fat content in milk. In particular, milk with a fat content of 3.2% and 3.8% contained 395.9±34.1 and 547.8±49.8 pg/ml of 17 $\beta$ -estradiol, respectively, nearly 10 times higher ( $p<0.05$ ) than milk with 1.0% fat and 1.8 to 2.5 times higher ( $p<0.05$ ) than milk with 2.5% fat. Similar trends were observed in fermented dairy products (yogurt, kefir), where the concentration of 17 $\beta$ -estradiol also depended on the fat content. In yogurt and kefir with 1.0% fat, the hormone concentration ranged from 25.5±3.0 to 36.1±3.6 pg/ml, which was, on average, 4.7 times lower ( $p<0.05$ ) than in products with 2.0 and 2.5% fat content. The study found significant changes in 17 $\beta$ -estradiol content in cream and butter, depending on fat content. It was found that the concentration of 17 $\beta$ -estradiol in cream and butter was several times (5-10) higher than in milk and fermented dairy products, regardless of their fat content. Thus, the quantity of 17 $\beta$ -estradiol in dairy products depends primarily on its concentration in raw milk used for production and the fat content in the final product.

The research on the acute toxicity of milk with 17 $\beta$ -estradiol concentrations ranging from 20.0 to 2500 pg/ml showed no significant changes in the movement activity, body shape, or reproductive inhibition of infusoria. At 17 $\beta$ -estradiol

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Scientific edition

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Monograph

Signed before printing 29.05.2025.

Format 60x84/16. Offset paper. Digital printing.  
Auth. sheet 7,56. Mind. printing sheet 5,58. Electronic edition. Deputy No 637

Printed PE Zvoleiko D.H.

9 Kn. Koriatovychiv St.; Kamianets-Podilskyi, Khmelnytskyi Region, 32301;  
e-mail: abetka.svit@gmail.com

Certificate of entry into the State Register of Publishing Entities  
series DK No. 2276 dated 31. 08. 2005.

