

**UREASE ACTIVITY OF SOIL MACROAGGREGATES IN QUERCUS CERRIS AND QUERCUS PETRAEA FORESTS AT THE LANDSCAPE LEVEL**

*Kazakh Research Institute of Soil Science and Agrochemistry named after U.U. Usmanov 75/V  
Al-Farabi avenue 050060, Almaty, Kazakhstan, madgu@indox.ru*

*Annotation.* The main objective of this study was to determine changes in soil urease activity in natural macroaggregates development along a slope in *Q. cerris* and *Q. petraea* forest soils. This study was carried out in Kocadag, Samsun, Turkey. Four landscape positions i.e., summit, shoulder, back slope 1, back slope 2 and foot slope, were selected. For each landscape position, soil macroaggregates were separated into six aggregate size classes using a dry sieving method and then urease activity was analyzed. In this research, topography influenced the macroaggregate size and urease activity within the aggregates. At all landscape positions, the contents of macro aggregates (especially > 6.3 mm and 2.00–4.75 mm) in all soil samples were higher than other macro aggregate contents. In foot slope position, the soils had generally the higher urease activity than the other positions at all landscape positions. In all positions, except for shoulder, urease activity was greater macro aggregates of < 1 mm than in the other macro aggregate size.

*Keywords:* topography, soil, aggregate, urease activity, ferments, forest soil.

**BACKGROUND**

Soil texture has an important role in nutrient management because it influences nutrient retention. For instance, finer textured soils tend to have greater ability to store soil nutrients. The size, quantity and stability of aggregates recovered from soil reflects an environmental conditioning that includes factors which enhance the aggregation of soil [1]. Soil aggregation influences the susceptibility of soil to erosion, organic matter storage, soil aeration, water infiltration and mineral plant supply. Many studies have shown the effects of organic constituents on the amount and stability of soil aggregates. However, understanding the role that soil aggregation plays in fertility recapitalisation also requires a knowledge of how aggregation contributes to organic matter storage in soil. Both processes are mediated by soil biological activity [2].

Soil microorganisms are key players in the recycling of elements, they stabilize soil structure and improve soil water retention. Their activities are essential components of the biotic community in natural forests and are largely responsible for ecosystem functioning [3, 4]. The microbial population and

their activities of the soil surface horizon has been far better studied than that of the soil aggregates [5-7]. Microbes in the deeper horizons also play an important role in ecosystem biogeochemistry [8].

Ferments (enzymes) - protein substances of nature, capable of hundreds of times to accelerate biochemical processes. Enzymes vary widely in molecular size: some have low molecular weight ( $10^4$ ), but most of their molecular weights are in the range from  $1.5 \cdot 10^4$  up to  $1.5 \cdot 10^6$ . Efficiency is high enzyme: catalyzes the conversion of one molecule of  $10^2 - 10^6$  molecules of substrate in 1 min. Most often, the enzymes specific for the type of reaction catalyzed. For example, only the urease decomposes urea polyphe-noloxidase oxidizes polyphenols and derivatives thereof. Urease (urea amidohydrolase, EC 3.5.1.5) is an important enzyme in soil because of the hydrolytic action on urea applied as a fertilizer, or excreted in the urine of grazing animals, but its origins, existence and persistence in soil are obscure. It is generally believed that a significant proportion of urease activity in soil is released from living and lysed microbial cells, and is stabilized as an extracellular enzyme by association

with soil colloids [9], especially soil organic matter [10, 11] listed 10 distinct categories of enzymes in soil. For example, total urease activity in soil may comprise activities associated with viable microorganisms, clay and humic colloids, leaked from extant cells, or released from lysed cells and cell debris. However, urease which become associated with humic colloids due to adsorption, entrapment or copolymerization during organic matter formation would persist for a long period [12].

Clearly then, to understand the role that soil aggregation large or small spatial scales plays between microbiological properties in soils and natural ecosystems such as forest, grassland and agricultural use. At first, necessary to understand the relationship between soil aggregation and biological activity.

The aim of this paper is to evaluate: 1) to assess the influence of topography on soil macro aggregates size distribution, 2) to compare the relationships between macro aggregate size distribution and soil urease activity.

## MATERIAL AND METHODS

### *Site description*

The Kocadağ, located in Samsun (Latitude, 41° 19' N; longitude, 36° 02' W) and has an elevation between 200 – 1200 m above sea level in Northern Anatolia (Figure 1). The climate is semi humid, (Rf = 52.5) with temperatures ranging from 6.6°C in February to 23°C in August. The annual mean temperature is 14°C and annual mean precipitation is 735 mm. Topography and slope show great variations and hilly and Rolling physiographic units are particularly common in the study area. Geology of the study area is dominantly composed of sandstone and limestone. The research area is in the A6 square according to the Davis's grid system [13]. Plant species belonging to the families *Quercus cerris* L. var. *cerris* and *Quercus petraea* (Mattuschka) Liebl. Subsp. *iberica* (Steven ex Bieb) Krasslin are dominant in this forest.

According to the Keys of soil taxonomy, the soils of Kocadağ forest belong to Typic Ustifluvent and Typic Haplustept. Soil samples were collected from five locations; summit, shoulder, backslope 1, backslope 2 and footslope, which differed from each other in altitude, slope, and soil physical and chemical characteristics (Table 1). Some part of natural forest has been fragmented and degraded by such human disturbances. As a result of the destruction *Rhododendron lutetum* Sweet. communities replaced this forest.

### *Soil Sampling*

For study that were carried out in December 2012, soil samples were collected from the organic layer (above the topsoil, thickness approximately 0-15 cm), on the basis of hypothesis that toposequence might be the main controlling factor in soil microbial indices of aggregates. Soils have been studied on along transect (crosswise from South to North direction) with representative five surface soil at summit, shoulder, back slope and foot slope positions were described (figure 1).

Soil samples (~1000 g) were taken with using a sterile soil corer (sterilized with 95% ethanol before use). The samples were transported to the laboratory on the same day. To analyze some soil physicochemical analyses, crop residues, root fragments and stones >2 mm were removed from soil samples. The soil aggregates were separated from different diameter sieves, thus, we obtained total 32 macroaggregate samples. These samples were used to determine microbial response variables of soils at the field moisture condition. Also each sample was stored in polyethylene bags at 4°C in the refrigerator for no longer than 72 h prior to analysis.

### *Soil physico-chemical analysis*

Physical and chemical properties of soils were determined by means of appropriate methods Particle size distribution by hydrometer method [14], pH and electrical conductivity (EC) in 1:1 (w/v) in soil: water

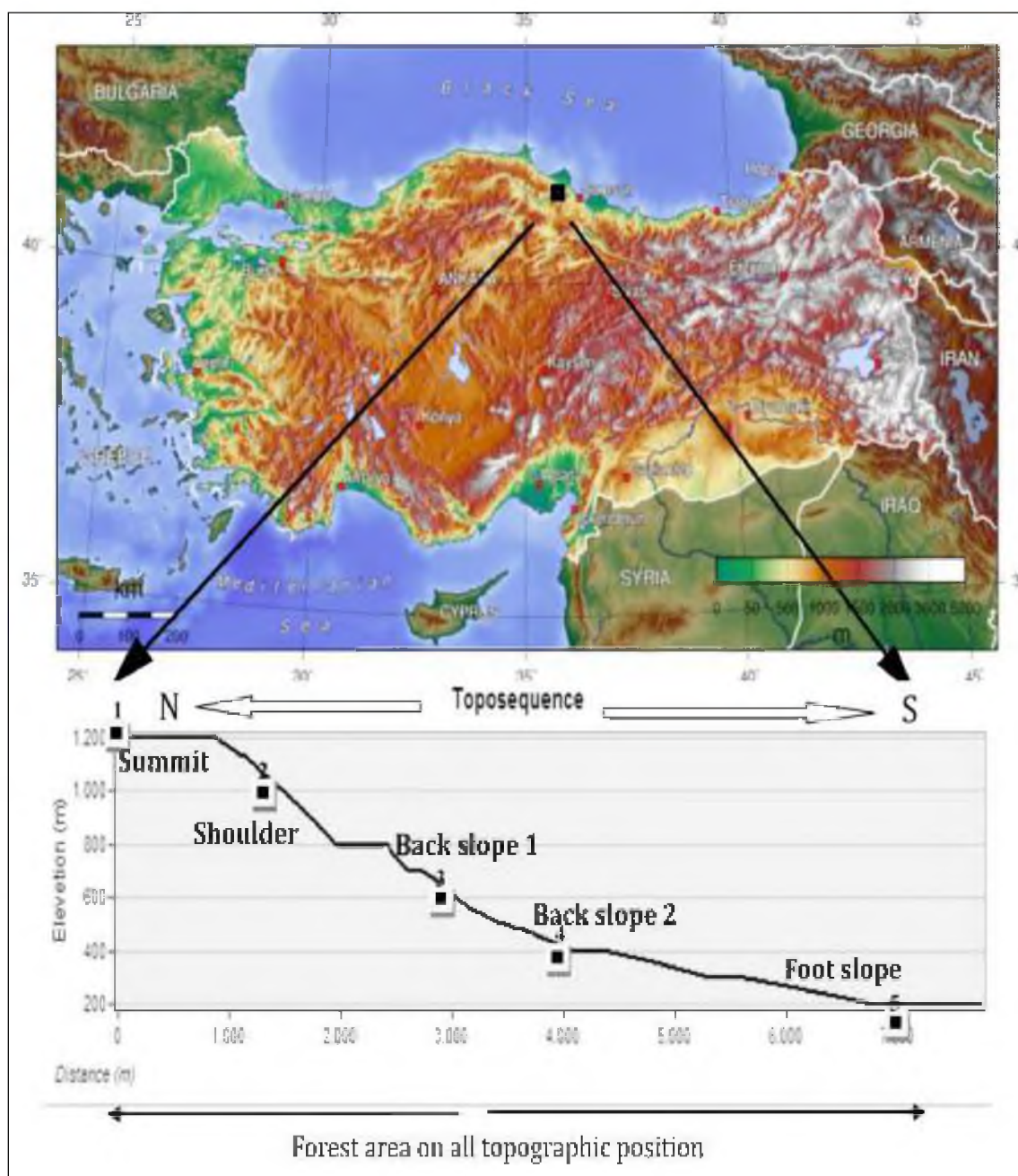


Figure 1 - Transect of the five different soil sample points on the same land cover but different topographic positions

suspension by pH-meter and EC-meter (Soil Survey Staff 1992),  $\text{CaCO}_3$  by Scheibler calcimetric method (Soil Survey Staff 1992), Soil organic carbon by a modified Walkley-Black method (Soil Survey Staff 1992).

#### *Separation of Aggregates*

The initial macro aggregate size distribution was determined by sieving 2 kg soil for 2 min on a stack of sieves with openings 6.30,

4.75, 2.00, 1.40 and 1.00 mm, from the top to the bottom of stack, using an automatic sieve shaker (speed and time of shaker were same) manufactured ELE International. Each size fraction was weighed and eight size classes were obtained: [I]  $>6300 \mu\text{m}$  (extremely macro-aggregate), [II]  $6300 - 4750 \mu\text{m}$  (very strongly macro-aggregate), [III]  $4750 - 2000 \mu\text{m}$  (strongly macro-aggregate), [IV]  $2000 -$

1400  $\mu\text{m}$  (moderately macro-aggregate), [V] 1400 – 1000  $\mu\text{m}$  (slightly macro-aggregate) and [VI] < 1000  $\mu\text{m}$  (very slightly macro-aggregate), indicated by Tisdall and Oades (1982) and Nearing (1995).

#### *Measurement of urease activity*

Urease (EC 3.5.1.5) activity (UA) was measured by the method of Hoffmann and Teicher. 0.25 ml toluene, 0.75 ml citrate buffer (pH 6.7) and 1 ml of 10 % urea substrate solution were added to the 1 g sample and the samples were incubated for 3 h at 37°C. The formation of ammonium was determined spectrophotometrically at 578 nm and results were expressed as  $\mu\text{m N g}^{-1}$  dry sample.

#### *Statistical Analysis*

All data were analyzed using SPSS 11.0 statistical software (SPSS Inc.). Analysis of variance (ANOVA) was carried out using one-factor randomized complete plot design; where significant F-values were obtained, differences between individual means were tested using the LSD (Least Significant Difference) test, with a significance level of  $P < 0.01$ . The asterisks, \*, \*\* and \*\*\* indicate significance at  $P < 0.05$ , 0.01 and 0.001, respectively.

## RESULTS AND DISCUSSION

### *Soil Physical and Chemical Properties*

Soil physical and chemical properties that have been taken into consideration in this research showed variability at short distance in study area formed on accumulated sediment depositions carried by Kızılırmak River. The major physical and chemical properties of the soils in the current study are presented in Table 1. Soil texture varied from sandy loam and loam through clay loam to clay across the surface soils of all profiles. Typic Haplustept had the highest clay content (42.69 %), while Typic Ustifluvent had the highest sand content (62.56 %). On the other hand, Typic Ustifluvent had a higher bulk density than Typic Haplustept due to its high sand content. Soil CEC varied between 8.46

and 35.56  $\text{cmol kg}^{-1}$ . The soil with the highest CEC was the sample no 4 of Typic Haplustept which had the highest clay content and organic matter. Exchangeable Ca and Mg cations accounted for over 95 % of the exchangeable complex as a result of the dissolution of carbonates, whereas exchangeable K and Na levels were rather low. Soil organic matter content ranged from 0.70 % to 1.57 % in upper horizons of all soil maps. These low organic matter levels are attributable to rapid decomposition and mineralization of organic matter due to cultivation practices. According to Table 1, soil reaction varies between 7.66–7.88 and soils have low in electrical conductivity ( $< 1 \text{ dS m}^{-1}$ ) and low nitrogen content. In addition, available phosphorous content of soils is moderate (8–25  $\text{mg.kg}^{-1}$ ) and high level (25–80  $\text{mg.kg}^{-1}$ ). Aggregate Size Distribution in Soils Percentage distribution of soil aggregates in soil samples used dry sieving method in total weight was shown in Figure 2. According to results, it was determined that majority of aggregates generally formed macroaggregates ( $> 250 \mu\text{m}$ ) in both Typic Ustifluvent and Typic Haplustept soils. The contents of macroaggregates (especially  $> 6300 \mu\text{m}$  and 2000–4750  $\mu\text{m}$ ) in all soil samples were higher than microaggregate contents. In addition to that, microaggregates were found higher content in sandy loam texture (Typic Ustifluvent-sample no 2). This case can be explained low clay and low organic matter content of this soil as compared to other soils. In other words, there was a close correlation among aggregate size, organic matter and particle size. Macroaggregate stability depends on management, because of the transient nature of binding agents (Soil Quality Test Kit Guide 1999). Tisdall and Oades (1982) formulated an aggregate hierarchy theory, which explains a gradual break down of macroaggregates, preceding complete dissociation in to primary particles. Another con-

sequence of this principle is that younger and the more labile soil organic matter is contained in macroaggregates than micro aggregates. Our result was also showed coherent with another research carried out by Qian et al. (2004). Their aim is determination of distribution characteristics

for microbial biomass carbon in different soil aggregates. In their studies, different aggregate fractions were obtained with the same method from the seven soil samples and they found that the soil with highest content organic matter had the highest proportion of large soil aggregates.

Table 1 - Selected soil physico-chemical properties at different landscape positions

| Soil properties            | Landscape position |             |              |              |             |
|----------------------------|--------------------|-------------|--------------|--------------|-------------|
|                            | Summit             | Shoulder    | Back slope 1 | Back slope 2 | Foot slope  |
| Coordinate (utm) Longitude | 37T 0257721        | 37T 0258378 | 37T 0259103  | 37T 0260089  | 37T 0264006 |
| Coordinate (utm) Latitude  | 4579115            | 4579806     | 4579500      | 4578216      | 4578763     |
| Clay, %                    | 77,99              | 58,6        | 67,28        | 38,28        | 20,91       |
| Silt, %                    | 14,76              | 25,76       | 19,55        | 28,18        | 21,26       |
| Sand, %                    | 7,25               | 15,64       | 13,17        | 33,54        | 57,83       |
| Texture class              | C                  | SiC         | C            | CL           | SCL         |
| Organic C, %               | 3,35               | 5,38        | 1,66         | 3,025        | 5,45        |
| CaCO <sub>3</sub> , %      | 3,4%               | 0,71%       | 1,82%        | 1,82%        | 1,74%       |
| pH (1:1)                   | 6,70               | 5,90        | 7,60         | 5,70         | 6,60        |
| EC, dS. m <sup>-1</sup>    | 0,24               | 0,20        | 0,47         | 0,29         | 0,93        |

#### *Aggregate Size Distribution in Soils*

Aggregate size distribution in soils percentage distribution of soil macro aggregates in soil samples used dry sieving method in total weight [7]. The contents of macro aggregates (especially > 6.3 mm and 2.00–4.75 mm) in all soil samples were higher than other macro aggregate contents. According to results, it was determined that majority of aggregates generally formed extremely macro aggregate (> 6.3 mm) at the foot slope position. But smallest macro aggregate size (< 1mm, 1.00-1.40 mm and 1.40-2.00 mm) were highest in the crest position. This case can be explained low organic matter content of this soil as compared to other soils. In other words, there was a close correlation among aggregate size and organic matter. Macro aggregate stability depends on management, because of the transient nature of binding agents (Soil Quality Test Kit Guide 1999). Tisdall and Oades (1982) formulated an aggregate hierarchy theory, which explains a gradual break down of macroaggregates, preceding complete dissociation in to primary

particles. Another consequence of this principle is that younger and the more labile soil organic matter is contained in macroaggregates than micro aggregates. Our result was also showed coherent with another research carried out by Qian et al. (2004), [5,7].

#### *Urease activity*

The urease activity distribution in natural soil macro aggregates was given in Table 2. In Except for shoulder position, the urease activity level increased with increasing macro aggregate size ( $P<0,01$ ), reaching a maximum in the < 1.00 mm at all landscape position. Considerable variations in urease activities were found for the different natural macro aggregate size at different landscape positions. Statistically significant variations were found in urease activities at various aggregate size and landscape position. The analysis of variance of the results obtained in our research on the urease activity showed that all factors (different landscape positions and aggregate size) significantly influenced urease activity (table 2).

Table 2 - Changes of urease activity ( $\mu\text{g N g}^{-1}$ ) in natural macro aggregate sizes of soil samples

| Macro aggregate size | Landscape position |                  |                 |                 |                  |
|----------------------|--------------------|------------------|-----------------|-----------------|------------------|
|                      | Summit             | Shoulder         | Back slope 1    | Back slope 2    | Foot slope       |
| >6,30 mm             | 70,7 $\pm$ 0,05    | 89,4 $\pm$ 2,3   | 70,0 $\pm$ 28,5 | 24,8 $\pm$ 16,5 | 54,2 $\pm$ 5,1   |
| 4,75-6,30 mm         | 95,1 $\pm$ 17,5    | 88,5 $\pm$ 3,0   | 32,3 $\pm$ 21,5 | 45,8 $\pm$ 2,7  | 56,3 $\pm$ 37,6  |
| 2,00-4,75 mm         | 90,8 $\pm$ 4,7     | 109,5 $\pm$ 4,5  | 49,6 $\pm$ 5,7  | 47,7 $\pm$ 9,1  | 97,9 $\pm$ 6,5   |
| 1,40-2,00 mm         | 118,7 $\pm$ 12,7   | 143,3 $\pm$ 32,9 | 69,8 $\pm$ 2,9  | 42,4 $\pm$ 28,3 | 154,3 $\pm$ 12,8 |
| 1,00-1,40 mm         | 114,3 $\pm$ 5,5    | 116,6 $\pm$ 17,8 | 48,8 $\pm$ 11,6 | 49 $\pm$ 32,7   | 310,5 $\pm$ 239  |
| <1,00 mm             | 97,0 $\pm$ 9,4     | -                | -               | 72,9 $\pm$ 5,7  | 207,26 $\pm$ 0   |

## CONCLUSION

Topography and aggregate size can influence microorganisms and their activities through affecting soil microclimate, physical and chemical soil characteristics, plant growth, and below ground C inputs. This study demonstrated that changes of macro aggregate size distribution can alter the soil urease activity within the aggregates. The results indicate that the macro aggregate size distribution and urease activity of macro aggregates along a hill slope had great differences in the forest soil depending homogeneous plantation. The foot slope position has greater organic C contents compared to the other positions, because the higher levels in the organic matter content clearly show erosion deposits at the foot slope and denuda-

tion of shoulder. In conclusion, soil properties and urease activity changed depending on landscape positions and aggregate sizes. Therefore, the forests must be used according to site specified management principles. This calls for cautions in large-scale conversions of the native forests to coniferous plantations as a forest management practice on concerns of sustainable soil productivity.

## Acknowledgments

We are grateful to the TUBITAK-BIDEB (The Scientific and Technological Research Council of Turkey) for their financial support. A lot of thanks to the staff of Soil Science & Plant Nutrition Department (Faculty of Agriculture, Ondokuz Mayıs University, Samsun, Turkey) for all their assistance.

## REFERENCES

- 1 Beare M.H., Bruce R.R. A comparison of methods for measuring water-stable aggregates: Implications for determining environmental effects on soil structure // *Geoderma*. - 1993. - Vol.56. - P.87-104.
- 2 Albrecht A., Angers D.A., Beare M., Blanchart E., Soil aggregation, soil organic matter and soil biota interactions : implications for soil fertility recapitalization in the tropics. 16th World Congress of Soil Science. 20 August 1998, Scientific registration n: 212, Symposium n: 12.
- 3 Hackl E, Bachmann G, Zechmeister-Boltenstern S. Microbial nitrogen turnover in soils under different types of natural forest // *Forest Ecology and Management*. - 2004. - Vol. 188. - P. 101-12.
- 4 Kızılkaya R., Aşkın T., Bayraklı B., Sağlam M. Microbiological characteristics of soils contaminated with heavy metals // *European Journal of Soil Biology*. - 2004. - Vol.40. - P. 95-102.
- 5 Aşkın T., Kızılkaya R., Organic and microbial biomass carbon contents of aggregates in a toposequence of pasture Soils // *Asian Journal of Chemistry*. - 2006. 1Vol. 8(2). - P. 1500-1508.
- 6 Dengiz O., Kızılkaya R., Erkoçak A., Durmuş M. Variables of Microbial Response in Natural Soil Aggregates for Soil Characterization in Different Fluvial Land Shapes // *Geomicrobiology Journal*. - 2013. - Vol. 30. - P. 100-107.
- 7 Kussainova M., Durmuş M., Erkoçak A., Kızılkaya R. Soil dehydrogenase activity of natural macro aggregates in a toposequence of forest soil. // *Eurasian Journal of Soil Science*. - 2013. - Vol. 2. - P. 69 - 75.

- 8 Madsen E. Impacts of agricultural practices on subsurface microbial ecology // *Advances in Agronomy*. - 1995. - Vol. 54. - P. 1-67.
- 9 Pinck L. A., Allison F. E. Adsorption and release of urease by and from clay minerals // *Soil Sci.* - 1961. - Vol. 91. - P. 183-8.
- 10 McLaren A.D. Enzyme activity in soils sterilized by ionising radiation and some comments on micro-environments in nature/ ed. N. E. Gibbons. // *Microbiology*. - University of Toronto Press. - 1963. - Vol. 8. - P. 221-9.
- 11 Burns R.G. Enzymes in soil: location and a possible role in microbial ecology // *Soil Biol. Biochem.* - 1982. - Vol. 14. - P. 421-5.
- 12 Pettit N.M., Smith A.R.J., Freedman R.B., Burns R.G. Soil urease: activity, stability and kinetic properties // *Soil Biol. Biochem.* - 1976. - Vol. 8. - P. 479-84.
- 13 Davis P.H. *Flora of Turkey: and the East Aegean Islands*. - Edinburgh University Press. 1965.
- 14 Bouyoucos G.J. A recalibration of the hydrometer method for making mechanical analysis of soils // *Agronomy Journal*. - 1951. - Vol. 43. - P. 435-438.

#### РЕЗЮМЕ

М.Д. Кусаинова

#### АКТИВНОСТЬ УРЕАЗЫ В ПОЧВЕННЫХ МАКРОАГРЕГАТАХ В ЛЕСАХ ПОД QUERCUS CERRIS И QUERCUS PETRAEA НА ЛАНДШАФТНОМ УРОВНЕ

*Казахский научно-исследовательский институт почвоведения и агрохимии им. У.У. Успанова  
050060, Алматы, пр. аль-Фараби, 75в, Казахстан, madgu@inbox.ru*

Основная цель данного исследования была в определении изменений активности уреазы в почве в зависимости от макроагрегатного состава и элементов рельефа лесных почвах на зональном лесном сообществе *Quercus cerris* and *Quercus petraea*. Пробы почвы были отобраны в районе Коджада, города Самсун, Турции с пяти участков, т. е. с вершины, склона, откос 1, откоса 2 и у подножья склона. Почвенные образцы были просеяны сухим методом разделения на шесть фракций, а затем, была проанализирована активность уреазы. По результатам исследования было определено что, топография повлияла на размер макро агрегатов и на активность уреазы в агрегатах на всех ландшафтных участках, за исключением откоса 2. Во всех образцах почв содержание макроагрегатов, во фракциях > 6,3 мм и 2,00-4,75 мм было более высокое, чем в других фракциях. активность уреазы была высокой в макро агрегатах фракции 1,00-1,40 мм у подножья склона, чем в других фракциях.

#### ТУЙІН

М.Д. Кусаинова

#### ЛАНДШАФТҚ ДЕҢГЕЙДЕ QUERCUS CERRIS ЖӘНЕ QUERCUS PETRAEA ӨСЕТІН ОРМАНДАРДАҒЫ ТОПЫРАҚ МАКРОАГРЕГАТТАРЫНДАҒЫ УРЕАЗАНЫҢ БЕЛСЕНДІЛІГІ

*Ө.О. Успанов ат. Қазақ топырақтану және агрохимия ғылыми зерттеу институты,  
050060, Қазақстан, Алматы, аль-Фараби даңғ., 75в, madgu@inbox.ru*

Бұл мақалада табиғи жағдайда *Quercus cerris* and *Quercus petraea* орман топырақтарының құрамында уреазы белсенділігі қандай өзгеріс беретіндігі жөнінде жүргізілген зерттеу жұмастарының түсіндірмесі және алынған нәтижелері туралы анықтама берілген. Зерттелінетін топырақ өрнектері Түркия мемлекетінің, Самсун қаласы, Коджада ауданында бес түрлі нүктелерден алынды. Атап кетсек: шың, баурайы, құлама 1, құлама 2 және тау бөктері. Зерттеу нәтижесінде, зерттелінген жер алаңы топырақ макро агрегаттық құрамына және уреазы белсенділігіне әсері анықталды. Барлық топырақ өрнектерінде, әсіресе > 6,3 мм және 2,00-4,75 мм фракцияларында макроагрегаттар құрамы өте жоғары болды. Тау бөктерінен алынған топырақ өрнектерінде уреазы белсенділігі басқа нұсқалармен салыстырғанда 1,00-1,40 мм фракциялы макроагрегаттарда жоғары көрсеткішті көрсетті.