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# Περιβαλλοντική Βιοτεχνολογία-Environmental Biotechnology

## Ενότητα 6: Biological Removal of Nitrogen

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# Nitrification (Microbiology)

**Nitrification** is the microbiological oxidation of $NH_4^+$-N to $N0_2^-$-N and $NO_3^-$ -N. Because of its oxygen demand (up to 4.57 g $O_2$/g $NH_4^+$-N) and toxicity to aquatic microorganisms, **$NH_4^+$-N removal is a mandated process** for some wastewaters. In addition, wastewater treatment that involves denitrification of $NO_3^-$ -N frequently requires nitrification to convert the input $NH_4^+$-N to $NO_3^-$ -N. Nitrification to remove $NH_4^+$-N from drinking-water supplies also is practiced in order to make the water biologically stable and to eliminate the free-chlorine demand that produces chloramines when free chlorine is desired.

The nitrifying bacteria are **autotrophs, chemolithotrophs, and obligate aerobes**. Each factor is crucial for understanding when nitrifiers can be selected and accumulated in a biological process.

# Nitrification (Microbiology)

Nitrifiers are **obligate aerobes**, and they use $O_2$ for respiration and as a direct reactant for the initial monooxygenation of $NH_4^+$ to $NH_20H$ (hydroxylamine). The latter use of oxygen may be the reason why nitrifiers are relatively intolerant of low dissolved-oxygen concentrations; nitrifier catabolism is slowed by oxygen limitation at concentrations that have no effect on many heterotrophs.

Nitrification is a two-step process. In the first step, $NH_4^+$ is oxidized to $N0_2^-$ according to the following energy-yielding reaction, which is normalized to one electron equiv.

$$\frac{1}{6}NH_4^+ + \frac{1}{4}O_2 = \frac{1}{6}NO_2^- + \frac{1}{3}H^+ + \frac{1}{6}H_2O$$

$$\Delta G^{0'} = -45.79\,kJ\ per\ e^-\ eq.$$

The most commonly recognized genus of bacteria that carries out the first step is *Nitrosomonas; however, Nitrosococcus, Nitrosopira, Nitrosovibrio, and Nitrosolobus* are also able to oxidize $NH_4^+$ to $N0_2^-$.

# Nitrification (Microbiology)

**The second stage of the nitrification reaction-is the oxidation of $NO_2^-$ to $NO_3^-$:**

$$\frac{1}{2}NO_2^- + \frac{1}{4}O_2 = \frac{1}{2}NO_3^-$$

$$\Delta G^{0'} = -37.07\,kJ\ per\ e^-\ eq.$$

Although *Nitrospira, Nitrospina, Nitrococcus*, and *Nitrocystis* are known to sustain themselves from the second-stage reaction, *Nitrobacter* is the most famous genus of the $NO_2^-$ oxidizers. Within the Nitrobacter genus, several subspecies are distinct, but closely related genetically within the alpha subdivision of the proteobacteria . Recent findings using oligonucleotide probes targeted to the 16S rRNA of Nitrobacter indicate that Nitrobacter is not the most important nitrite-oxidizing genus in most wastewater treatment processes. Nitrospira more often is identified as the dominant nitrite oxidizer.

# Nitrification (Kinetic parameters)

The $f_s^0$ is very low for ammonium and nitrite oxidizers. Compared to the typical $f_s^0$ value of 0.6-0.7 for aerobic heterotrophs, nitrifiers conserve very few electrons in biomass. The low $f_s^0$ values translate directly to low Y values. While the numerical value of Y for the ammonium oxidizers might not appear to be much lower than for aerobic heterotrophs, the apparent closeness is an illusion caused by using different units in the denominator. In units of g $VSS_a$/g OD, Y for the ammonium oxidizers is approximately **0.1, compared to about 0.45 for heterotrophs**.

The maximum specific growth rates of both organisms are low, with both of them less than 1/d at 20°C. With such small values of $\hat{\mu}$ , the limiting value of $\theta_x^{min}$ must be large: All values are greater than 1 d. On the other hand, nitrifiers are able to drive effluent $NH_4^+$ or $NO_2^-$ concentrations to very low levels, since $S_{min}$ values are well below 1 mg N/L/

# Nitrification (Kinetic parameters)

The temperature effects are quite important, because nitrification is sometimes considered impossible for low-water temperatures. In fact, stable nitrification can be maintained at 5°C or lower, as long as the SRT remains high enough. For 5°C, a safety factor of only 5 requires that $\theta_x$ be 3.6 . 5 = 18 d. One problem with low-temperature nitrification is that $\hat{\mu}$ becomes quite small, making recovery of nitrification after a washout a very slow process. Thus, avoiding nitrifier washout due to excess sludge wasting, low D.O., or inhibition must be an absolute priority, particularly for low temperatures.

The directly comparable parameters-such ·as $f_s^0$, $\hat{\mu}$ , $[\theta_x^{min}]_{lim}$, and $S_{min}$-are very similar for the two types of nitrifiers. This circumstance is completely logical, since both are aerobic chemolithoautotrophs oxidizing N. Furthermore, they almost always coexist in the same habitats, experiencing the same SRT and oxygen concentrations. The correspondence of limiting parameters reflects their biochemical and ecological similarities.

# Nitrification (Kinetic parameters)

The following equation is an overall, balanced reaction for the complete **oxidation of $NH_4^+$ to $NO_3^-$-N** by nitrifiers having $\theta_x$=15 d and / $f_s$ = 0.067. It represents a typical situation for both nitrifier types together.

$$NH_4^+ + 1.815O_2 + 0.1304CO_2 = 0.0261C_5H_7O_2N + 0.973NO_3^- + 0.921H_2O + 1.973H^+$$

Besides the low net formation of nitrifier biomass ($Y_{net}$ = 0.21 g $VSS_a$/g N), this stoichiometric equation illustrates the two other important features of nitrification. First, nitrification creates a major oxygen demand. Second, nitrification produces almost two strong-acid equivalents per mole of $NH_4^+$ removed. In common mass units, the alkalinity consumption is 1.973·50/14 = 7.05 gas $CaC0_3$/g $NH_4^+$-N. The first step, ammonium oxidation, is responsible for the acid production.

Nitrifiers produce soluble microbial products, which can be consumed by heterotrophic bacteria

# Nitrification (Kinetic parameters)

Nitrifiers are reputed to be highly sensitive to **chemical inhibition**. This belief is partly accurate. The very slow growth rate of nitrifiers magnifies the negative impacts of inhibition and, in part, makes it appear that nitrifiers are more sensitive than are faster growing bacteria. Furthermore, some apparent inhibitors are electron donors whose oxidation depletes the D.O. and may cause oxygen limitation. However, nitrifiers are sensitive to inhibition from a range of organic and inorganic compounds. Among the most relevant ones are: **unionized $NH_3$ (at higher pH), undissociated $HNO_2$ (usually at low pH), anionic surfactants, heavy metals, chlorinated organic chemicals, and low pH.**

# Nitrification (Set-up)

A successful nitrification process-suspended growth or biofilm-must account for the reality that heterotrophic bacteria always are present and competing with the nitrifiers for dissolved oxygen and space. The nitrifiers' relatively high $K_O$ value puts them at a disadvantage in the competition for oxygen. Their slow growth rate is a disadvantage when competing for any space that requires a high growth rate. These two disadvantages are overcome by ensuring that the nitrifiers have a **long SRT, typically greater than 15 d**, although larger values may be needed in the presence of toxic materials, a low D.O. concentration, or low temperature. In activated sludge, maintaining a SRT of 15 d or greater corresponds to the loading condition termed extended aeration. Thus, maintaining an extended aeration loading usually is synonymous with having a nitrifying process. For biofilm processes, the BOD flux and the detachment rate indirectly control the nitrifiers' SRT.

# Nitrification (Set-up)

**One-sludge nitrification** can be carried out in sequencing batch reactors, which involve sequential periods of filling, aerobic reaction, settling, and effluent draw-off in one tank. As for any SBR system, multiple units are almost always needed, in order that storage requirements for incoming wastewater are not excessive. In most ways, SBR nitrification resembles any other one-sludge system. However, the fill and- draw-feeding scheme can create conditions resembling plug flow in a continuous feed system.

# Nitrification (The role of the ratio $BOD_L$/TKN)

Nitrifying systems are affected by the influent **$BOD_L$ :TKN ratio** in three ways. **The first way** is that synthesis of heterotrophic biomass sequesters nitrogen and reduces the flow of nitrogen from ammonium, to nitrite, and to nitrate. If the influent $BOD_L$:TKN ratio is large enough-say, greater than about 25 g $BOD_L$/g TKN-little or no reduced nitrogen is available for nitrification.

**Second**, the $BOD_L$:TKN ratio determines what fraction of the active biomass is comprised of nitrifiers. Due to the low $f_s^0$ for the nitrifiers, their fraction normally is Jow. For typical $BOD_L$:TKN ratios in municipal sewage (5 to 10 g $BOD_L$/g N), the nitrifiers normally constitute less than 20% of the active biomass and are a smaller fraction of the volatile suspended solids.

**Finally**, the $BOD_L$:TKN ratio exerts some control over how heterotrophs and nitrifiers compete for common resources: dissolved oxygen and space in flocs or biofilms. In the long term, a higher $BOD_L$ :TKN ratio tends to force the nitrifiers deeper into the floc or biofilm. This incurs greater mass transport resistance for the nitrifiers' substrates, particularly $NH_4^+$ and $O_2$. In the short term, a high growth rate for heterotrophs could create negative impacts on nitrifiers by sequestering nitrogen, consuming oxygen, or physically sweeping nitrifiers out of the floc or biofilm

# Nitrification (ANAMMOX process)

Recently, a novel bacterium in the planctomycetes group has been discovered for its ability to anaerobically oxidize $NH_4^+$-N to $N_2$, not to $NO_2^-$ . It is called the **ANAMMOX** microorganism because it does **ANaerobic AMMonium OXidation**. The ANAMMOX bacterium uses ammonium as its electron donor and nitrite as its electron acceptor. The energy reaction is:

$$NH_4^+ + NO_2^- = N_2 + 2H_2O$$

The yield and specific growth rate reported for ANAMMOX are low, about 0.14 g $VSS_a$/g $NH_4^+$-N and O.065/d, respectively. This gives an overall **stoichiometry** of approximately:

$$NH_4^+ + 1.26NO_2^- + 0.085CO_2 + 0.02H^+ = N_2 + 0.017C_5H_7O_2N + 0.24NO_3^- + 1.95H_2O$$

# Denitrification (Physiology of bacteria)

**Denitrification** is widespread in nature. Denitrifiers are common among the Gram-negative Proteobacteria, such as ***Pseudomonas, Alcaligenes, Paracoccus*,** and ***Thiobacillus***. Some Gram-positive bacteria, including Bacillus, can denitrify. Even a few halophilic Archaea, such as Halobacterium, are able to denitrify.

The denitrifiers used in environmental biotechnology are chemotrophs that can **use organic or inorganic electron donors**. Those that utilize organic electron donors are heterotrophs and are widespread among the *Proteobacteria*. A more limited group of autotrophs can **utilize $H_2$** and **reduced sulfur**. Because of their great metabolic diversity, denitrifiers are commonly found in soils, sediments, surface waters, ground-waters, and wastewater treatment plants.

Denitrification proceeds in a stepwise manner in which nitrate ($NO_3^-$) is sequentially reduced to nitrite ($NO_2^-$), nitric oxide (NO), nitrous oxide ($N_2O$), and $N_2$ gas. Each half-reaction and the enzyme catalyzing it are shown below.

$$NO_3^- + 2e^- + 2H^+ = NO_2^- + H_2O \qquad Nitrate\,\mathrm{Re}\,ductase$$
$$NO_2^- + e^- + 2H^+ = NO + H_2O \qquad Nitrite\,\mathrm{Re}\,ductase$$
$$2NO + 2e^- + 2H^+ = N_2O + H_2O \qquad Nitric\,Oxide\,\mathrm{Re}\,ductase$$
$$N_2O + 2e^- + 2H^+ = N_{2(g)} + H_2O \qquad Nitrous\,Oxide\,\mathrm{Re}\,ductase$$

# Denitrification (Effect of $O_2$)

The oxygen concentration controls whether or not the facultative aerobes respire nitrogen. Oxygen can control denitrification in two ways. The first is **repression** of the several nitrogen-reductase genes. The second control mechanism is **inhibition** of the activity of the reductase by D.O. concentrations greater than a few tenths of a mg $O_2$/L. The fact that the D.O. concentrations that repress the reductase genes are much higher than the concentrations that inhibit their activity means that **denitrification can occur when D.O. concentrations are well above zero**. This situation is enhanced when the denitrifying bacteria are located inside flocs or biofilms, where the oxygen concentration is lower than in the bulk liquid.

Very low concentrations of the electron donor or too high concentrations of D.O. concentration can lead to accumulation of the denitrification intermediates: $NO_2^-$, $NO_2$, and $N_2O$.

# Denitrification (Effect of pH- organic substrates)

Although denitrifiers are not especially pH sensitive, pH values outside the optimal **range of 7 to 8 can lead to accumulation of intermediates**. In low alkalinity waters, pH control can become an issue, because denitrification produces strong base. Base production is illustrated by the balanced reactions in which acetate and H2 are electron donors for the heterotrophic and autotrophic denitrifiers, respectively:

$$CH_3COOH + \frac{8}{5}NO_3^- + \frac{4}{5}H_2O \rightarrow \frac{4}{5}N_2 + 2H_2CO_3 + \frac{8}{5}OH^-$$

$$4H_2 + \frac{8}{5}NO_3^- \rightarrow \frac{4}{5}N_2 + \frac{8}{5}OH^- + \frac{16}{5}H_2O$$

Although heterotrophic denitrifiers exhibit a nearly infinite range for their organic substrates, a few simple organic substrates have been intensively studied.

**methanol, acetate, glucose, ethanol**

# Denitrification (Kinetic characteristics)

In summary, the heterotrophic denitrifiers have **kinetic characteristics similar to aerobic heterotrophs**. Because they are facultative aerobes, the shifts from $O_2$ respiration to $NO_3^-$ or $NO_2^-$ respiration causes only a small decrease in $f_s^0$ and Y, which gives only modest increases in $[\theta_x^{min}]_{lim}$. Thus, denitrification processes should perform similarly to aerobic processes used for $BOD_L$ removal.

On the other hand, the kinetic characteristics of **heterotrophic denitrifiers and autotrophic nitrifiers are very different**. The nitrifiers have lower $f_s^0$ values, are much slower growers, and require substantially longer solids retention times. Furthermore, maximum nitrification rates require a high D.O. concentration, while high D.O. concentration slows or stops denitrification. Because nitrification often is necessary to provide the $NO_3^-$ or $NO_2^-$ for denitrification, process design and operation must reconcile these conflicting physiological characteristics. It is the reconciliation of the needs of the nitrif1ers and heterotrophic denitrifiers that distinguishes the different approaches to denetrification in environmental biotechnology.

# Denitrification (Kinetic characteristics)

One important feature of the denitrifying bacteria is **that they often use $NO_3^-$ (or $NO_2^-$) as the N source for cell synthesis**. The added electron cost of reducing the N source to the - 3 oxidation state reduces $f_s^0$ and the true yield. For example, using $NO_3^-$] as the N source requires 8 extra electron equivalents per mole of biomass, represented as $C_5H_7O_2N$. $C_5H_7O_2N$ requires 20 electron equivalents to reduce the C to oxidation state zero in all cells, but it requires 28 electron equivalents to reduce the C and N when the N source is $NO_3^-$. Thus when $NO_3^-$ is the N source, the oxygen demand of biomass is (28 $e^-$ eq/mol cells) (1 mol cells/113 g cells) (8 g $0_2/e^-$ eq) = **1.98 g OD/g cells**, not 1.42 g OD/g cells.

# Tertiary denitrification

**Tertiary denitrification** is appropriate whenever the water to be treated contains $NO_3^-$ or $NO_2^-$, but little or no electron donor. This situation occurs naturally with agricultural runoff contaminated with nitrogen fertilizers. Tertiary denitrification also follows aerobic biological processes (i.e., secondary treatment) of wastewater. When the secondary treatment oxidizes all electron donors originally present, reduced nitrogen is oxidized to $NO_3^-$, while reduced carbon is mineralized. The outcome of secondary treatment is N converted to $NO_3^-$ , but almost no donors.

Almost any organic compound could be used as an exogenous electron donor. Historically, **methanol** was chosen for its economic benefits, not because it is a "better“ exogenous electron donor than any other choice. When available, concentrated organic wastes can be used as an inexpensive (or even free) electron donor.

# Tertiary denitrification

Inorganic electron donors also can be used and are gaining popularity. **Hydrogen** gas ($H_2$) is an excellent electron donor for autotrophic denitrification. Its advantages include lower cost per electron equivalent compared to organic compounds, less biomass production than with heterotrophs, and absolutely no reduced nitrogen added. The main disadvantage of $H_2$ in the past has been lack of a safe and efficient $H_2$-transfer system. The recent development of membrane-dissolution devices overcomes the explosion hazard of conventional gas transfer and makes $H_2$ a viable alternative.

Reduced sulfur also can drive autotrophic denitrification. The most common source of reduced S is elemental sulfur, S(s), which is oxidized to $SO_4^{2-}$. The S normally is embedded in a solid matrix that includes a solid base, such as $CaC0_3$, because the oxidation of S(s) generates strong acid.

$$S(s) + \frac{6}{5} NO_3^- + \frac{2}{5} H_2O \rightarrow SO_4^{2-} + \frac{3}{5} N_2 + \frac{4}{5} H^+$$

# Tertiary denitrification with activated sludge

**Tertiary denitrification with activated sludge** is a common approach for treating nitrate-bearing waters. The basic configuration is the same as for aerobic treatment using activated sludge: a mixed reactor, a quiescent settler, sludge recycle, and sludge wasting to control the solids retention time. The design SRT generally is around **5 d** when a heterotrophic electron donor is added. It is longer (e.g., **15 d**) when denitrification is autotrophic.

Although the liquid contents need to be well mixed, contact with the air needs to be minimized. This is achieved by subsurface mixing, usually with a submerged turbine.

Second, supplementation with electron donor is required. Although the exact dose of donor can be computed with stoichiometry, a rule of thumb is **4 g $BOD_L$/g $NO_3^-$-N** removed through denitrification. Extra electron donor must be supplied if $O_2$ enters the system.

# Tertiary denitrification with activated sludge

Settler design is based on the same principles as for aerobic activated sludge. It might seem that rising sludge should be a problem; however, successful denitrification drives the $NO_3^-$ concentration to a very low level in the reactor, which means that $N_2$ generation in the settler is minimal.

Sludge production can be computed from the SRT and stoichiometry. A rule of thumb is about **0.75 g VSS/g $NO_3^-$-N** removed by heterotrophic denitrification and an SRT near 5 d, but it is lower for autotrophs or if the SRT is increased.

# One-sludge denitrification

One-sludge denitrification, sometimes called single-sludge or combined denitrification, involves using the BOD in the influent of a wastewater to drive denitrification. Thus, one-sludge denitrification cannot work as an add-on process after secondary treatment, because secondary treatment removes the organic electron donors. Instead, denitrification must be fully integrated with the aerobic processes, BOD oxidation and nitrification. This integration must reserve organic electron donor for anoxic denitrification, while at the same time providing aerobic conditions that allow full nitrification, which generates the nitrate for denitrification.

Advantages

- Because no exogenous electron donor needs to be added, chemical costs are reduced over tertiary denitrification.
- Because some of the influent BOD is oxidized with nitrate as the electron acceptor, not $O_2$, aeration costs are reduced compared to alternative systems that nitrify the reduced-nitrogen forms in the influent.
- Full or nearly full N removal is achieved, thereby protecting receiving waters at risk from cultural eutrophication.

# One-sludge denitrification

Influent wastewater normally contains organic BOD and reduced nitrogen, often called total **Kjeldahl nitrogen**, or TKN. The TKN must be oxidized to $NO_3^-$-N without oxidizing all the BOD before denitrification takes place. The three basic strategies for reserving organic electron donor while nitrification takes place are:

- Biomass storage and decay
- Classical predenitrification
- Simultaneous nitrification with denitrification

# One-sludge denitrification

Although biomass storage and decay is a simple and effective way to reserve electrons for denitrification, it is not often employed as a stand-alone process. Two shortcomings explain why biomass storage and decay has limited applicability by itself. First, **endogenous respiration has slow kinetics**. A typical *b* value is 0.05/d for denitrification. Second, **the decay of biomass always releases $NH_4^+$-N**. Thus, the anoxic step returns N and nitrogenous BOD to the water, albeit at concentrations lower than in the process influent.

## Advantages/ disadvantages

**Classical predenitrification** has come into widespread use worldwide. Its **advantages** include the direct use of influent BOD for denitrification, which reduces aeration costs compared with strictly aerobic removal of BOD; faster kinetics than with biomass storage and decay; and no release of $NH_4^+$-N, as with biomass storage and decay. The main **disadvantage** of predenitrification is the large mixed-liquor recycle rate, which can substantially increase costs of piping and pumping.

# Denitrification-modelling

When the design is conservative, stoichiometry, not kinetics, controls the concentrations of substrates. Thus, we assume that $NO_3^-$-N is completely denitrified in the anoxic reactor, or $(NO_3^-)^1 = 0$. Likewise, $BOD_L$ and TKN are fully oxidized in the aerobic reactor, making $BOD_L^2 = TKN^2 = 0$, while the maximum amount of $NO_3^-$-N is generated in the aerobic reactor, $(NO_3)^2$.

This analysis does not compute the volumes required for the anoxic reactor, aerobic reactor, and settler. The distribution of volume between the anoxic reactor and aerobic reactor can be estimated from successful practice, in which $V_{aer} = 1.5V_{an}$. The settler volume should be based on the solids flux and overflow rate.

# Denitrification-modelling

In the anoxic reactor, denitrification occurs to the maximum degree possible. In other words, $NO_3^-$-N is driven to zero when BOD is in excess, or $BOD_L$ is driven to zero when $NO_3^-$-N is in excess. The electron donor is the input BOD, which we represent as complex organic matter, $C_{10}H_{19}O_3N$. When $C_{10}H_{19}O_3N$ is oxidized as the electron donor for anoxic heterotrophs, the donor half reaction is:

$$\frac{1}{50}C_{10}H_{19}O_3N + \frac{9}{25}H_2O = \frac{9}{50}CO_2 + \frac{1}{50}NH_4^+ + \frac{1}{50}HCO_3^- + H^+ + e^-$$

Which is written for 1 electron equivalent, or 8 g $BOD_L$. The acceptor half-reaction is that for reduction of $NO_3^-$ -N to $N_2$ :

$$f_e\left(\frac{1}{5}NO_3^- + \frac{6}{5}H^+ + e^- = \frac{1}{10}N_2 + \frac{3}{5}H_2O\right)$$

The acceptor half-reaction is multiplied by $f_e$ to account for the actual flow of electrons for energy generation under the operating SRT. The synthesis half-reaction, multiplied by $f_s$, is:

$$f_e\left(\frac{1}{20}NH_4^+ + \frac{1}{5}CO_2 + \frac{1}{20}HCO_3^- + H^+ + e^- = \frac{1}{20}C_5H_7O_2N + \frac{9}{20}H_2O\right)$$

# Denitrification-modelling

The three half-reactions can be combined in the usual manner ($R=-R_d + f_e R_a + f_s R_c$) if $f_s$ and $f_e$ are known. We can compute $f_s$ and $f_e$ if we choose an SRT for the predenitrification system using Equation:

$$f_s = f_s^0 \frac{1+(1-f_d)b\theta_x}{1+b\theta_x}$$

$$f_e = 1 - f_s$$

- As expected, $f_s$ and $Y_{n(den)}$ for denitrification decrease with increasing $\vartheta_x$, while $f_e$ increases.
- The ratio of $NO_3^-$-N consumed (as acceptor and N source), g $NO_3^-$-N/g $BOD_L$, systematically increases as $\vartheta_x$ goes up, since more biomass is oxidized by endogenous respiration.
- The ratio g $BOD_L$/g $NO_3^-$-N is the reciprocal of g $NO_3^-$N/g $BOD_L$ and is very useful for determining whether $BOD_L$ or $NO_3^-$-N is limiting denitrification.

# References

The images where their origin is not mentioned are derived from the book:

Environmental Biotechnology : Principles and Applications,

Bruce E. Rittmann and Perry L. McCarty,

McGraw-Hill Series in Water Resources and Environmental Engineering

# Τέλος Ενότητας

# Χρηματοδότηση

- Το παρόν εκπαιδευτικό υλικό έχει αναπτυχθεί στο πλαίσιο του εκπαιδευτικού έργου του διδάσκοντα.
- Το έργο «**Ανοικτά Ακαδημαϊκά Μαθήματα στο Πανεπιστήμιο Αθηνών**» έχει χρηματοδοτήσει μόνο την αναδιαμόρφωση του εκπαιδευτικού υλικού.
- Το έργο υλοποιείται στο πλαίσιο του Επιχειρησιακού Προγράμματος «Εκπαίδευση και Δια Βίου Μάθηση» και συγχρηματοδοτείται από την Ευρωπαϊκή Ένωση (Ευρωπαϊκό Κοινωνικό Ταμείο) και από εθνικούς πόρους.

# Σημειώματα

# Σημείωμα Ιστορικού Εκδόσεων Έργου

Το παρόν έργο αποτελεί την έκδοση 1.0.0.

# Σημείωμα Αναφοράς



# Σημείωμα Αδειοδότησης