**ORIGINAL ARTICLE**

**Cigdem Kader** [1]
**Mustafa Kara** [2]
**Ayse Yesim Gocmen** [3]
**Ayse Erbay**[4]
**Muhammet Fevzi Polat** [3]

[1]Bozok University, School of Medicine Department of Infectious Diseases and, Clinical Microbiology, Yozgat, Turkey.
[2]Bozok University, School of Medicine, Department of Obstetrics and Gynaecology, Yozgat, Turkey
[3]Bozok University, School of Medicine, Department of Biochemistry, Yozgat, Turkey
[4]Bozok University, School of Medicine, Department of Infectious Diseases and, Clinical Microbiology, Yozgat, Turkey.

***Corresponding Author:***
*Cigdem Kader, MD*
*Adnan Menderes Boulevard No:44,*
*Zip Code: 66200, Yozgat, Turkey*
*Phone: +90 354 212 70 60*
*E-mail: dr_cigdemtr@yahoo.com*

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konuralptipdergi@duzce.edu.tr
konuralptipdergisi@gmail.com
www.konuralptipdergi.duzce.edu.tr

# Antibodies Against Vaccine Preventable Diseases in Pregnant Women Measles, Mumps, Rubella, Varicella and Tetanus in Yozgat, Turkey

**ABSTRACT**

**Objective:** Vaccine preventable diseases (VPD) during pregnancy are risk not only for the mother, but also for the fetus. The aim of this study was to determine the seroprevalence of antibodies to tetanus and measles, mumps, rubella, varicella (MMRV) in pregnant women in Yozgat Turkey.

**Methods:** Blood samples were taken from 176 pregnant women who were admitted to obstetrics and gynecology outpatient department of our hospital. The specific IgG antibodies against tetanus and MMRV viruses were determined quantitatively by ELISA and a self-recorded questionnaire was filled including their demographics, disease and vaccination history.

**Results:** The mean age of the pregnant women participating in the study was 27.2±4.9 (18 -45). Tetanus and MMRV IgG seropositivity were respectively 90.3%, 97.2%, 98.3%, 99.4%, 100%. Immunity to tetanus was higher in pregnant who were vaccinated within last 5 years compared to non-vaccinated ones (93.7% vs 84.6 % respectively, p=0.049). Mean age of individuals having immunity for tetanus was 26.9±5 years while that of individuals without immunity 29.8±3.7 years (p=0.0195). There was a significant negative correlation between age and tetanus immunity rate. It was found that immunity to tetanus decreased as age increased (p=0.002).

**Conclusion:** In this study, high rate of tetanus and MMRV immunity was found among pregnant woman. However, considering of the risks due to the infection during pregnancy, antibodies to VPD should be scanned in childbearing women who have not vaccinated previously or have not the disease, and seronegative ones should be vaccinated before pregnancy.

**Keywords:** Pregnancy, Measles, Rubella, Mumps, Varicella, Tetanus, Vaccine-Preventable Diseases

# Türkiye, Yozgat'da Hamile Kadınlarda Aşı ile Önlenebilir Hastalıklara Karşı Antikorlar; Kızamık, Kabakulak, Kızamıkçık, Suçiçeği ve Tetanoz

**ÖZET**

**Amaç:** Hamilelik esnasında geçirilen aşı ile önlenebilen hastalıklar sadece anne için değil fetus için de risklidir. Bu çalışmanın amacı; Türkiye'nin Yozgat ilindeki hamile kadınlarda seçilmiş aşı ile önlenebilen hastalık etkenleri olan kızamık, kızamıkçık, kabakulak, suçiçeği (KKKS) ve tetanoz hastalığı antikorlarının seroprevalansını araştırmaktır.

**Yöntem:** Kan örnekleri, hastanemiz kadın hastalıkları ve doğum polikliniğine başvuran 176 hamile kadından alındı. KKKS virüsleri ve tetanoza karşı spesifik antikor titreleri ELISA ile kantitatif olarak saptandı. Hastalık, aşılama geçmişi ve demografik bilgilerinin dahil olduğu anket formu doldurulduu.

**Bulgular:** Katılımcıların yaş ortalaması 27.2±4.9 (18-45) idi. Tetanoz, KKKS IgG seropozitifliği sırasıyla % 90.34, % 97.16, % 98.30, % 99.4, % 100 idi. Son 5 yıl içerisinde tetanoz aşısı olanlarda bağışıklık oranı olmayanlara göre daha yüksek bulundu (93.7% ye 84.6 % , p= 0.049). Tetanoza karşı bağışık olanların yaş ortalaması 26.9±5, bağışık olmayanların yaş ortalaması 29.8±3.7 bulundu (p=0.0195). Yaş ile tetanoz bağışıklık oranları arasında ters korelasyon tespit edildi. Yaş artıkça tetanoz bağışıklık oranının azaldığı tespit edildi (p=0.002).

**Sonuç:** Çalışmamızda gebelerde yüksek oranda tetanoz ve KKKS bağışıklığı bulunmuştur. Ancak, hamilelik esnasında enfeksiyonun neden olduğu riskler göz önünde bulundurulduğunda, aşı ile önlenebilen hastalıklara karşı antikorlar; daha önce aşılanmamış ya da hastalık geçirmemiş doğurganlık çağında olan kadınlarda taranmalıdır ve seronegatif olanlar aşılanmalıdır.

**Anahtar Kelimeler:** Gebelik, Kızamık, Kızamıkçık, Kabakulak, Suçiçeği, Tetanoz, Aşı İle Önlenebilen Hastalıklar

## INTRODUCTION

Pregnancy is an especially vulnerable course for vaccine-preventable diseases (VPD) associated complications for both mother and newborn (1). Spectrum of perinatal outcomes from viral infections during pregnancy can be from no effect to pregnancy loss by spontaneous abortion to fetal infection with resulting congenital abnormalities (2). Hence, immunization before pregnancy would be ideal for the prevention of VPD (3). Passive immunity transplacental immune globulin G antibody transfer from vaccinated mother to fetus maintains infant protection for up to 6 months of life (3). Measles is a paramyxovirus that occurs usually with rash, diarrhea, and otitis media along with bronchopneumonia or encephalitis in serious cases. Infection during pregnancy enhances the risk of spontaneous abortion, preterm birth, and low birth weight.

Mumps is another paramyxovirus that occurs with flu-like symptoms and bilateral parotitis and is related with spontaneous abortion. Rubella is a togavirus that occurs with more nonspecific symptoms including lymphadenopathy, arthralgia, fever, and rash. Infection during pregnancy, particularly in the first trimester, can be destructive (3). Varicella zoster virus, a member of the herpes virus family, causes chicken pox. Illness particularly presents as a pruritic rash for 4–7 days, during which time the infected individual is highly contagious. Viral transmission occurs by direct contact with skin lesions or inhalation of aerosolized particles. Infection during pregnancy is related with neonatal varicella or herpes zoster and congenital varicella syndrome, which is characterized by skin scarring, limb hypoplasia, low birth weight, and numerous other anomalies. Congenital varicella syndrome appears in 1–2% of cases of maternal varicella infection with the highest risk of occurrence related with maternal infection from 13 to 20 weeks of gestation (3). *Clostridium tetani (C. tetani)* secretes the tetanospasmin neurotoxin, which leads tetanus infection (3). Tetanus developing in pregnancy or within postpartum 6 weeks is defined as maternal tetanus. Neonatal tetanus (NT) is classified as tetanus in the first month of life (4). Maternal and neonatal tetanus (MNT) are types of generalized tetanus (5). MNT cases are predominantly observed in isolated communities where obstetric and postnatal practices were unhygienic, and where vaccination coverage of tetanus toxoid was low (5). However tetanus can develop all over the world throughout the life, neonates, who become infected via contaminated umbilical stump, and their mothers are with highest risk, especially in case of childbirth under unhygienic conditions since the mothers don't have enough antitoxins due to lack of optimal immunization to guard themselves and their newborn babies (4). The aim of this study was to determine the seroprevalence of the causative agents of VPD antibodies against measles, mumps, rubella, varicella (MMRV) and tetanus in pregnant women in Yozgat, Turkey.

## MATERIAL AND METHODS

**Study design and study population:** This cross-sectional study was planned to determine the MMRV and tetanus antibody levels among adult pregnant women who admitted at the Bozok University, Faculty of Medicine Obstetrics and Gynecology outpatient clinic in Yozgat. Data and serum collection were conducted in 2013. After filling in the informed consent form, the respondents were subjected to face-to-face interviews using a questionnaire. A standard form was filled including age, pregnancy history, education level, occupation, presence of childhood vaccines, marital status, accident or injury history, and previous tetanus vaccination status. Each participant provided a 5 ml blood sample to determine their MMRV and tetanus antibody levels, and the serum was separated through centrifugation and kept at -80 ℃ until the day of study.

**Serologic study:** At enrollment, 5 ml of venous blood sample was collected from each pregnant women, and serum samples were stored at -80℃ until tested. The specific IgG antibodies against MMRV viruses and tetanus bacteria were determined quantitatively by immunosorbent enzyme- linked assay (ELISA) kits (Euroimmun, Lübeck, Germany). Immunity status was determined from the antibody concentration, according to the kits' manufacturer guidelines. The recommended cut-off values of antibody concentrations are shown in Table 1.

**Table 1.** The recommended cut-off values of antibody concentration

| MMRV IgG Titers | Cut-off Values | | |
|---|---|---|---|
| | Positive | Borderline | Negative |
| Euroimmun Anti-Measles Virus ELISA IgG | ≥275 IU/ml | 200-274 IU/ml | <200 IU/ml |
| Euroimmun Anti-Mumps Virus ELISA IgG | ≥22 RU/ml | 16-21 RU/ml | <16 RU/ml |
| Euroimmun Anti-Rubella Virus ELISA IgG | ≥11 IU/ml | 8-10 IU/ml | <8 IU/ml |
| Euroimmun Anti-Varicella Virus ELISA IgG | ≥110 IU/ml | 80-109 IU/ml | <80 IU/ml |

| Tetanus titers IU/ml | Recommendation |
|---|---|
| <0.01 IU/ml | No protection: basic immunisation or booster required, depending on anamnesis, serological control after 4 to 8 weeks |
| 0.01-0.1 IU/ml | Immunisation protection uncertain: booster required, serological control after 4 to 8 weeks |
| >0.1-0.5 IU/ml | Short-term immunisation protection present: booster recommended |
| >0.5-1.0 IU/ml | Immunisation protection present: booster or serological control recommended after 3 years minimum |
| >1.0-5.0 IU/ml | Long-term immunisation protection present: booster or serological control recommended after 5 years minimum |
| >5.0 IU/ml | Long-term immunisation protection present: booster or serological control recommended after 8 years minimum |

All borderline sera were re-tested to verify the results. Sera that were found repeatedly borderline were accepted negative.

**Statistical analysis:** Statistics were run with Software package STATA 11.0 (College station, Texas, USA). Continuous variables were expressed as mean ±standard deviation (SD) and categorical variables were expressed as percentage. The Chi-square test or Fisher's exact test was used, when appropriate to compare proportions. Continuous variables were compared using an independent-groups Student's t test if normality assumptions were met; otherwise, groups were compared using the Wilcoxon rank sum test. A p-value of <0.05 was considered statistically significant. Spearman's test was used for correlation analyses of age and tetanus IgG titer.

**Ethical considerations:** Before the study process, we obtained approval from the Ethics Committee of Bozok University, Yozgat, Turkey (Reference number: 14.02.2013/49). Purpose and procedures of the study was explained to each participant. After receiving oral and written information about the study procedures, the participants were asked to sign a written informed consent form for participation. Confidentiality was maintained throughout the study. There was no potential risk to participate, and they were informed that they could quit at any time in case they preferred not to complete the questionnaire.

## RESULTS

A total of 176 pregnant women were enrolled in our study. Mean age of the participants was 27.2±4.9 (ranging from 18 to 45 years old). Immunity rates to measles, mumps, rubella, varicella and tetanus were found 171 (97.2%), 173 (98.3%), 175 (99.4%), 176 (100%) and 159 (90.3%) respectively. Table-2 demonstrates some of the sociodemographical features of the evaluated pregnant women. Most of the pregnant women were high school graduates (36.4 %) and housewives (70.5 %). Among them, 1.7 % did not have social insurance.

The distribution of seropositivity rates against MMRV and tetanus according to the age groups are shown in Table-3.

**Table 3.** Distribution of MMRV and Tetanus seropositivity by age groups

| Ages (years) | Tetanus n (%) | Measles n (%) | Rubella n (%) | Mumps n (%) | Varicella n (%) |
|---|---|---|---|---|---|
| 18-19 (n=9) | 9 (100%) | 9 (100%) | 9 (100%) | 8 (88.9%) | 9 (100%) |
| 20-29 (n=107) | 98 (91.6%) | 103 (96.3%) | 106 (99.1%) | 105(98.1%) | 107 (100%) |
| 30-39 (n=58) | 50 (86.2%) | 57 (98.3%) | 58 (100%) | 58 (100%) | 58 (100%) |
| 40-49 (n=2) | 2 (100%) | 2 (100%) | 2 (100%) | 2 (100%) | 2 (100%) |
| Total (n=176) | 159 (90.3%) | 171 (97.2%) | 175 (100%) | 173 (98.3%) | 176 (100%) |

**Table 2.** Sociodemographic characteristics of the evaluated pregnant women ( n=176)

| Variable | No (%) |
|---|---|
| Age, years | 27.2±4.9 |
| **Education** | |
| Elementary school graduates | 33 (18.7 %) |
| Secondary school graduates | 32 (18.2 %) |
| High school graduates | 64 (36.4 %) |
| University graduates | 45 (25.6 %) |
| Illiterate | 2 (1.1 %) |
| **Occupation** | |
| Housewife | 124 (70.5 %) |
| Government employee | 33 (18.8 %) |
| Teacher | 10 (5.7 %) |
| Medical Doctor | 3 (1.7 %) |
| Employee | 6 (3.3 %) |
| **Marital satatus** | |
| Married | 176 (100 %) |
| **Social insurance** | |
| Present | 173 (98.3 %) |
| Absent | 3 (1.7 %) |
| **Residence** | |
| Urban | 109 (61.9 %) |
| Rural | 67 (38.1 %) |
| **Presence of childhood vaccines (MMRV)** | |
| Present | 176 (100 %) |
| **Accident or injury history** | |
| Absent | 131 (74.4 %) |
| Present | 45 (25.6 %) |

The highest seropositivity rate of %100 was found in the 40-49 years age group. There was no significant difference between the seropositivity rates with respect to marital status, education level, occupation, accident or injury history, childhood vaccines or coverage by social insurance. Ninety (51.1 %) of the evaluated pregnant women had a child history. The median value of number of children and pregnancies were found 1 and 2 respectively. It was found that 57.4% of the pregnant women had tetanus vaccine during their pregnancies.

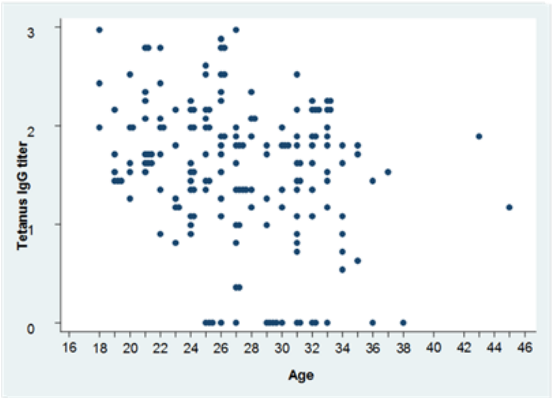


**Figure-1:** Tetanus antibody levels and age correlation graph (r= -0.231, p=0.002).

Pregnant women who were vaccinated against tetanus within last 5 years had higher immunity rates compared to non-vaccinated ones (93.7% vs.84.6 % respectively; p= 0.049). Mean age of individuals having immunity to tetanus was 26.9±5 years while that of individuals without immunity 29.8±3.7 years. Spearman's correlation analysis showed a significant negative correlation between age and antibody titers (r = -0.231, p =0.002) (figure-1). It was found that immunity to tetanus decreased as age increased.

## DISCUSSION

Considering the seroprevalence of VPD antibodies is essential not only for preventing infections regionally, but also so many infectious diseases that have potential of effecting the mother and fetus during pregnancy and the infant postnatal period. VPD during pregnancy can deteriorates the mother, and cause abortion, preterm delivery, growth retardation, malformation, and/or congenital fetal infection. Acquired immunity from the mother to the fetus via the placenta, continues to prevent the baby to be infected approximately 6 month following the delivery (6). Additionally, acquired immunity protects also the mother in case of child-to-mother infections during nursing and supplies postpartum protection and stability. Antibody titers against viruses are influenced by both natural infections and immunizations which the patient is experienced in her life and are country dependent (6). The antibody titers during pregnancy are influenced by the patient's history of immunizations, infections, and steady-state of the titers following the onset of the initial immune response. These evidences may be associated with applications such as voluntary vaccination campaigns and exposure to natural infections and national epidemics in accordance with birth year (6).

In the review of the studies related to antibody seroprevalance for preventable disease among pregnant women, Sauerbrei et al. found that in 209 maternal and chordal blood samples, mumps and tetanus immunity rates were 96% and 93% respectively. The immunity rates for poliomyelitis, diphtheria, measles and rubella were between 55% to 91% (7). Occurence of mumps, measles, rubella and chicken-pox during pregnancy were related to still birth, miscarriage, intrauterine infections (8). In our study, protective antibodies against MMRV were substantially high among pregnant subjects (97-100%). In the study with 804 pregnant women held in Yozgat province of Turkey, IgM/IgG seropositivity for rubella, cytomegalovirus (CMV) and toxoplasma were 0.1/94.0%, 0.1/99.8% and 0.2/36.9% respectively (9). In another study with 1926 pregnant women held in Istanbul province of Turkey, IgM/IgG seropositivity for rubella, toxoplasma and CMV were 0.15/95.7%, 0.9/31.2% and 0.7/99.2% respectively (9). A study from Japan revealed that antibody seroprevalance against MMRV among pregnant women was 72.4%, 76.2%, 83.9% ve 96.1% respectively (6). In our study, these ratios were 97.2% for measles, 98.3% for mumps and 99.4% for rubella, 100% for varicella. In England, tetanus seropositivity was found to be 79% among the pregnant women (11). In our study, tetanus immunity rate was found to be 90.3%. Ceylan et al. found 74.1% in the Diyarbakır province of Turkey (12). In other studies from Turkey, tetanus immunity rate among pregnant women were reported between 53-84% (13-14). Altıntaş et al. stated that among pregnant women who were vaccinated one or two times during their pregnancies had 98.8% immunity rates to tetanus (14). In our study, pregnant women who were vaccinated against tetanus within last 5 year had higher immunity compared to non-vaccinated ones (93.7% vs. 84.6 % respectively). Mean age of individuals having immunity for tetanus was 26.9±5 years while that of individuals without immunity 29.8±3.7 years. There was a significant negative correlation between age and tetanus immunity rate. It was found that immunity to tetanus decreased as age increased. Neonatal tetanus can develop in case of delivery under non-hygienic conditions due to infection of umbilical cord. Absence of maternal immunity increases the risk of neonatal tetanus. Although neonatal tetanus is a preventable disease via vaccination, it is still an important public health problem in the developing countries. Maternal immunity for severe infections via either vaccination or natural infection protects not only pregnant women but also newborns within the first month of their lives (4).

In Turkey, mumps and rubella vaccines were included in expanded vaccination program in addition to measles vaccination in 2006 and varicella vaccine was included in 2013. In 2007 children with age between 8 and 13 were vaccinated against MMR in a catch-up vaccination program (15,16). In our country, tetanus toxoid (TT) vaccinations began in 1935 and since 1957, it has been used in conjunction with the Diphtheria-Pertussis-Tetanus (DPT) vaccine. TT vaccinations were scheduled across the country by the Ministry of Health in 1985, and since then, tetanus vaccinations have been given as part of a childhood vaccination program. Vaccinations are given to children in the second, fourth and sixth months after birth, and at 12 months after the last vaccination. A further dose is given at the age of six, when the child has reached school age. This application is referred to as primary immunization, and a booster at the age of 10 years is recommended (16). Tetanus vaccine is administered after 20 weeks of pregnancy in Turkey (17). The primary vaccination Schedule is applied to unvaccinated or partially vaccinated pregnant women. Following the 20$^{th}$ of gestational week, 2 doses of vaccines separated by one-month interval

are applied. 6-12 months later, the third dose is provided (17). Tetanus vaccination is a part of the expanded vaccination program in Turkey. Tetanus immunization services are carried out free of charge at all primary health centers and hospitals (17). Advisory Committee on Immunization Practices (ACIP) recommends that one dose Tdap vaccine should be applied to pregnant women due to decrease in protection by previous immunization during pregnancy. The vaccine can be applied any time during pregnancy but to obtain sufficient immunization during delivery, it is better to apply the vaccine in third trimester (preferably between the $27^{th}$ and $36^{th}$ of gestational weeks) (18). ACIP recommends single dose Tdap in absence of rapel within the last 5-10 years, if Tdap is not available and 10 years passed over the rapel dose, then Td vaccination is recommended (in $2^{nd}$ or $3^{rd}$ trimesters). If Td has not been applied or not complete, preferably two consecutive doses of Td are applied between $20^{th}$ of gestational week and delivery, with four-week intervals, third dose is given 6 to 12 months later. One of the three dose is recommended to be of Tdab instead of Td (18).

In Turkey, Yozgat, the coverage rates of MMRV and tetanus vaccines have been comparatively high. On the other hand, because of the risks related to the infection in pregnancy, antibodies to VPD should be evaluated in women who were not vaccinated previously or did not experience the disease before, and the vaccination should be provided accordingly. In conclusion; the ongoing status of the antibody titers implies that immunity against these VPD should be proved, and the vaccination should be offered for patients with low antibody titers while childbearing period in Turkey. Screening immunity level against VPD in pregnancy is important to prevent infections especially for tetanus and this type of evaluation may be good oppurtunity to assess immunity and to supply vaccination; vaccinations should be advised for pregnant women with inadequate antibody titers following delivery. From a sociological perspective on prevention, a vaccination campaign for young children should be arranged and vaccinations should be applied to mothers with insufficient immunity to protect the baby from exposure to VPD. As potential immunization applications are directed only for mothers in the perinatal period, we should encourage mothers at this time to inform young children about immunization. We proposed that antibody against VPD should be measured before pregnancy, and woman who had inadequate level of immunization should be vaccinated accordingly.

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