THEORETICAL AND APPLIED APPROACHES IN HEALTH SCIENCES

Editors:
Prof. Dr. Eray YURTSEVEN
Dr. Latife UZUN

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Editors: Prof. Dr. Eray YURTSEVEN, Dr. Latife UZUN

Editor in chief: Berkan Balpetek

Cover and Page Design: Duvar Design

Printing : May -2025

Publisher Certificate No: 49837

ISBN: 978-625-5885-41-8

853 Sokak No:13 P.10 Kemeraltı-Konak/İzmir

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Chapter 1

Molecular Properties Of Sperm Capacitation

Gökhan KOÇAK[1], Nurdan COŞKUN[2]

SPERMATOGENESIS

Male and female gametes originate from primordial germ cells that develop from the yolk sac wall endoderm in the 4th week of pregnancy and progress towards the gonad primordia, and increase their numbers by undergoing a series of mitotic divisions in the gonads (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003). Just before puberty, the sex cords in the gonad primordium develop into seminiferous tubules, while the primordial germ cells (stem cells) differentiate into spermatogonia, which are diploid germ cells on the seminiferous tubule basement membrane. Spermatogonia that remain undivided until puberty, together with spermatogenesis that begins at puberty, form Type A dark spermatogonia, which are the stem cells of the seminiferous epithelium. While some of the Type A dark spermatogonia with dense basophilic and finely granular chromatin oval nuclei remain as stem cells, some differentiate into Type A light spermatogonia with pale-staining finely granular chromatin nuclei. Some of the Type A light spermatogonia undergo mitotic division and form Type B spermatogonia with spherical nuclei and centrally located nucleoli. As a result of mitotic division of Type B spermatogonia; The largest germ cells of the seminiferous tubule, spherical or ovoid-shaped primary spermatocytes (4n DNA) are formed. Each primary spermatocyte reaches the leptotene, pachytene, diplotene and diakinesis stages in the prophase stage, which lasts approximately 22 days, and enters metaphase with chromosome separation and crossing over. In the anaphase following metaphase, they complete the first meiosis with the chromosomes moving to opposite poles and form two secondary spermatocytes

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(2n DNA). Secondary spermatocytes also undergo the second meiosis and form spermatids. Spermatids located in the Sertoli cell depressions have haploid chromosomes and nuclei containing condensed chromatin regions (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

Spermatogonia and spermatids in the differentiation process are embedded in the recesses and protrusions of the Sertoli cells in their cytoplasm and are connected to each other by protoplasmic bridges. The degree of maturation increases from the base to the apex of the seminiferous tubules. In the final stage of spermatogenesis, which is completed with the formation of spermatids from spermatogonia, the differentiation process called spermiogenesis, spermatids become spermatozoa (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

SPERMIOGENESIS

The entirety of the changes that spermatids undergo to transform into spermatozoa as a result of spermatogenesis is called “spermiogenesis” and these changes occur in four stages (Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

1. Golgi Stage: The cytoplasm of the spermatid contains a distinct Golgi complex near the nucleus, mitochondria, a pair of centrioles, free ribosomes and smooth endoplasmic reticulum tubules. The hydrolytic enzymes produced in the endoplasmic reticulum of the spermatid are transmitted to the Golgi complex and after undergoing various changes, they are released from the trans side of the Golgi complex as PAS(+) granules called “proacrosomal granules”. The acrosomal vesicles formed by the fusion of these granules are attached to the nuclear membrane and determine the anterior pole of the same sperm. In this stage, the centrioles move away from the nuclear region. One of them is positioned at the opposite pole of the acrosomal region to form the axoneme of the flagellum (tail skeleton containing 9 pairs of microtubules in the periphery and 2 in the center).

2. Hat Stage: The acrosomal vesicle expands and grows, starting from where it contacts the nucleus and enveloping the anterior part of the nucleus halfway like a cap. When the acrosomal vesicle reaches its final size, it is called an “acrosome”, which contains hydrolytic enzymes.

3. Acrosome Stage: The acrosome, which is considered a special type of lysosome, contains hydrolytic enzymes such as hyaluronidase, acrosin, neuraminidase, acid phosphatase and trypsin-like proteases. Hydrolytic vesicles are released into the external environment as a result of exocytosis when the oocyte plasma membrane and the sperm outer acrosomal membrane fuse. While hyaluronidase causes the sperm to pass through the corona radiata layer, acrosin and trypsin-like proteases melt the zona pellucida, allowing the first step of fetilization, known as the acrosome reaction, to occur. Microtubules originating from the centriole distal to the dark, small, pear-shaped nucleus form the axoneme, which will form the flagellum. The sperm midpiece, which provides spermatozoon motility, develops as mitochondria surround the proximal part of the flagellum.

4. Maturation Stage: The excess cytoplasmic parts called “residual bodies” formed by the disappearance of the protoplasmic bridges between spermatids are phagocytosed by Sertoli cells. As a result of the changes in the spermatid; mature spermatozoa develop, which are ejected into the seminiferous tubule lumen but do not have the ability to move or fertilize, and carry the genetic characteristics specific to the species (Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

2.1. MATURE SPERM

As a result of spermiogenesis lasting sixty-five to seventy-two days, spermatozoa that separate from Sertoli cells through spermiation and pass into the seminiferous tubule lumen are morphologically mature germ cells, but they are not yet functionally mature. They gain their motility abilities through secretions from accessory glands in the ductus epididymis and their fertilization abilities through capacitation in the female genital tract.

A fully mature sperm consists of 3 parts: head, middle piece and tail. The head region is surrounded by an acrosome up to its anterior half and contains a flat-shaped nucleus containing dense chromatin. The axoneme, which starts from the neck region where there is a pair of centrioles connecting the head region to the tail region, extends along the tail and is surrounded by numerous mitochondria, especially in the neck region. At the end of the neck region, there is a thickening called the “annulus” that plays a role in the movement of the sperm. The tail, which is the longest part of the sperm that resembles a cilium in structure, consists of 3 parts; middle piece, principal piece, terminal piece. The middle piece consists of a layer of helical mitochondria, 9+2 microtubular axonemes, and nine filaments called outer dense fibers, which start from the neck of the sperm and extend along the tail. The longest part of the tail, the principal piece, consists of a central axoneme surrounded by seven outer dense fibers and a fibrous sheath. The outer dense fibers and fibrous sheath contain keratin proteins that form a rigid skeleton for microtubular sliding and coiling during the forward movement of the sperm. The shortest part of the tail, the terminal piece, contains only the axoneme because of the termination of the outer dense fibers and fibrous sheath (Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

3. SPERM MEMBRANE

Sperm membrane is composed of protein, lipid and carbohydrate structure. The main function of lipids is to provide stabilization by forming the membrane structure, and to play a role in capacitation, acrosome reaction and oocyte-sperm fusion. Human spermatozoa contain high levels of phosphatidylcholine, phosphatidylethanolamine and sphingomyelin (Gulaya and Margitich, 2001; Poulos and White, 1973; Niu and Wang, 2009). Cholesterol together with phospholipids provides the integrity and impermeability of the sperm membrane. Monosaccharides such as mannose and glucose and disaccharides are found in the structure of the sperm membrane. Tyrosine, tryptophan and histidine constitute the main amino acid structure. In the membrane of spermatozoa; Apart from specific antigens (tyrosine kinase sp 95, proacrosin, PH-20, PH-30, sp 56, galactosyl galactosyltransferase, spermadhesins, progesterone receptor), nonspecific proteins that carry out cell-cell or cell-matrix interactions, matrix proteins (collagen, fibronectin, laminin, adhesion molecules) as well as adhesion molecules such as immunoglobulins, cadherins, selectins and integrins have been shown to be involved (Katz 1991).

4. CAPACITATION

In the first studies on fertilization, it was observed that while fertilization occurred by bringing together the sperm and oocyte of lower class animals, fertilization did not occur in mammals when sperm and oocyte were brought together (Visconti et al., 1995a). In all mammalian species, when spermatozoa are first deposited into the female genital tract; they do not have sufficient motility and ability to fertilize the oocyte in metaphase 2. This situation suggested that mammalian sperm undergoes some special changes before fertilizing the mammalian oocyte and directed researchers to capacitation studies (Salicioni et al., 2007; Visconti et al., 1995a).

In order for sperm to fertilize the oocyte, a preparation process for fertilization called capacitation is necessary (Salicioni et al., 2007; Visconti et al., 1995a; Austin 1952; Chang 1951). Discovered half a century ago by Austin (1951-1952)-Chang (1951-1955) (Austin 1952; Chang 1951), capacitation encompasses all of the molecular and physiological events by which sperm gain the ability to fertilize in the female genital tract. It is known that glycosaminoglycans, chondroitin sulfates, heparin-like and some yet unidentified substances localized in the female genital tract are responsible for the changes in the plasma membrane of spermatozoa during capacitation. Capacitation is based on the typical ligand-receptor interaction mechanism; It is a calcium-dependent, cAMP-dependent, kinase-dependent, G-protein-dependent, redox-dependent event (Salicioni et al., 2007; Keith and Persaud, 2008).

Then, hyperactivation, acrosome reaction and oocyte-sperm fusion events occur (Salicioni et al., 2007; Visconti et al., 1995a). In a fertile male, approximately 60-500 million sperms ejaculated with one ejaculation are stored in the upper part of the vagina, traveling 3 mm per minute with uterine muscle movement and sperm motility, and only 500 of these sperms reach the fallopian tube. The sperms ejaculated into the fornix with ejaculation reach the ampulla via the cervix and uterus within 1-1.5 hours. If the ovum encounters sperm within 24-48 hours after being expelled from the follicle in the female reproductive system, fertilization occurs in the ampulla of the fallopian tube. The pH of fresh semen is around 7-7.4, and with its buffering properties, it protects the sperm from the antibacterial acidity of the vagina against the normal vaginal pH of 4-4.5 (Gordon and Lu, 1990; Keith and Persaud, 2008; Sadler and Langman, 2010; Moghissi 1984).. Although the sperm that enters the female genital tract immediately following ejaculation is motile, they do not have the capacity to fertilize due to decapacitating factors that prevent the onset of early fertilization events during their progression through the male genital tract and the female lower genital tract. Decapacitating factors, which are carbohydrates with a molecular weight of less than 2000 around the sperm; They are released from the epididymis and seminal vesicles, are incorporated into the semen during ejaculation, and are removed from the sperm in the cervix, uterus, or tubes (Mc Rorie and Williams, 1974; Rachel et al., 2005; Brewis et al., 2005). During capacitation, the high amount of zinc (Zn^+) content in the “outer dense fibers” structure surrounding the axoneme in the tail of spermatozoa protects the tail from premature oxidation and provides a solid structure. During epididymal passage, more than 60% of Zn is excreted from the sperm, resulting in an increase in disulfide bridges and a decrease in sulfhydryl groups, thus hardening and stabilizing the outer dense fiber structure. It has been shown that Zn^+ binding in the environment significantly increases sperm motility (Wroblewski et al., 2006; Andrews et al., 1994). In addition, serum albumin, calcium (Ca^{+2}), bicarbonate (HCO_3^-) and energy sources are also required for capacitation. Spermatozoa obtain their energy sources from glycolysis and, to a lesser extent, from oxidative phosphorylation in mitochondria. Capacitation is initiated by cholesterol from the cholesterol-rich acrosome of the spermatozoon and from the membrane structure covering the flagellum (Brewis et al., 2005). Cholesterol is bound to proteins called caveolins, which are inactive and attached to the membrane, within caveolae on the membrane upon its removal (Visconti et al., 1999).

Albumin, which is found especially in follicular fluid in female genital system secretions, binds cholesterol in the sperm membrane and removes it from the membrane. As cholesterol leaves the sperm membrane, membrane fluidity increases and signal formation occurs. As a result, inactive protein kinases, G-proteins and intermediary molecules such as phosphofructokinase become active and allow calcium to enter the cell, initiating capacitation (Cross 1998). Other compounds that bind cholesterol besides albumin are: High-density lipoprotein (HDL) and β-cyclodextrin (Visconti et al., 1999). The separation of cholesterol from the membrane changes the membrane composition, increases calcium and bicarbonate permeability, and regulates adenosine 3'-5' cyclic monophosphate (cAMP) metabolism by stimulating adenylyl cyclase (Brewis et al., 2005). During capacitation, the HCO3- concentration responsible for the increase in intracellular pH is low in the epididymis, while it is high in the seminal plasma and fallopian tubes (Lin and Kan, 1996). Membrane hyperpolarization activates calcium channels, voltage-sensitive Na+/H+ exchanger channels, and K+ channels. Activation of dynein arms occurs after intracellular alkalinization, which occurs with the activation of H+ channels and increased bicarbonate permeability. The presence of intracellular H+, Na+, and K+ also induces motility with capacitation. Intracellular enzyme activations such as adenylyl cyclase, phosphatase, and phosphodiesterase occur with the increase in intracellular Ca+2 due to hyperpolarization. Activated adenylyl cyclase stimulates cAMP production by transferring phosphate from ATP (Flesch et al., 2001). Protein kinase-A, which is stimulated by adenylyl cyclase activity and cAMP synthesis, activates tyrosine kinase, which transfers phosphate from ATP to the amino acid terminals of intracellular proteins and performs tyrosine phosphorylation (Gadella and Harrison, 2000). Serine, threonine and tyrosine amino acids are the main amino acids that act as receptors for the oocyte membrane protein ZP3 after phosphorylation. While calcium is the modulator of tyrosine phosphorylation in mammals, Talpha-1, which is found in seminal plasma and tubal fluid, and hydrogen peroxide (H2O2) and nitric oxide (NO), which are free radical (ROS) products that have a toxic effect on spermatozoa, can be considered as modulators of tyrosine phosphorylation in humans (Monks et al., 1986). ROS are reactive oxygen species that cause lipid peroxidation at high doses and negatively affect capacitation and motility (Gerbers et al., 1982).

During capacitation, along with the increase in phospholipid methylation in the sperm cell, phosphotidylcholine synthesis from phosphotidylethanolamine also occurs (Monks et al., 1986). Phospholipase-C, which is activated by phosphotyrosine formed as a result of tyrosine phosphorylation and dependent on calcium, cleaves phosphoinositol bisphosphate (PIP2) into two fragments: inositol triphosphate (IP3) and diacylglycerol (DAG) (Urata et al., 2001). Inositol triphosphate (IP3) binds to endoplasmic reticulum or acrosome receptors, releasing Ca2+, and the released Ca2+ is transported by binding to calmodulin. The Ca2+ released by inositol triphosphate (IP3) is also required for the activation of protein kinase C (PKC), which is stimulated by diacylglycerol (DAG) (Gerbers et al., 1982). Although extracellular calcium is required during capacitation, the acrosome is also used as an intracellular calcium store (Gerbers et al., 1982).

5. IN VITRO CAPACITATION

In vitro capacitation is performed by incubating ejaculated sperm instead of a medium with an electrolyte composition similar to tubal fluid. Exogenous energy sources (pyruvate, lactate, glucose) are used as energy sources for the seven-hour capacitation process. The fact that capacitation is a temporary event and a capacitated sperm cannot become capacitated again makes in vitro studies difficult (Roldan and Fragio, 1994). Human follicular fluid containing albumin, glycosaminoglycans and progesterone, which has a stimulating effect on sperm motility and acrosome reaction, is used as biological stimulants. Protasome factors, which come from the prostate and have a dose-dependent use and a protective effect against peroxidation, have a stimulating effect on sperm motility and sperm count in seminal plasma. Many pharmacological drugs have been tested in vitro to improve sperm motility and fertilization ability. Among these, phosphodiesterase inhibitors such as caffeine and pentoxifylline, when added to semen in the laboratory, increase the intracellular cAMP level, glycolysis and ATP production, thus increasing the motile sperm rate and also initiating motility in live but immotile spermatozoa. However, the in vitro use of caffeine has been largely abandoned due to its harmful effects on the acrosome reaction and sperm membrane. When used in vitro, pentoxifylline, which inhibits phosphodiesterase at the cellular level, increases lipid peroxidation and affects sperm membrane fluidity. High-dose ROS negatively affect sperm motility and viability, while on the other hand, it plays an important role in capacitation, hyperactivation and induction of the acrosome reaction. NO, a cytotoxic free radical, inactivates superoxide and increases intracellular cGMP (Travert et al., 1993).

6. ACROSOME REACTION

Acrosome is a structure originating from the Golgi complex during spermatogenesis and initiates the acrosome reaction as a result of sperm contact with the oocyte (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995). The superficial attachment of sperm and ovum, the interaction of fertilizin and antifertilizin receptors in sperm are species specific. While the contact of sperm zona receptors with the zona pellucida, which is an oocyte membrane protein, opens Ca+2 channels, Cl- channels open when progesterone released from oocyte cumulus cells contacts GABA receptors in the sperm membrane. In addition, pH increases with the removal of H+ from the sperm membrane, while phospholipase A2 (PLA2) increases arachidonic acid, lyso phosphatidylcholine and platelet-activating factor (Rorie and Williams, 1974). Acrosomal enzymes known as hyaluronidase, acrosin, neuraminidase and trypsin-like substances are found between the inner and outer acrosomal membranes of the sperm. With the fusion of the oocyte plasma membrane and the outer acrosomal membrane of the sperm, hydrolytic vesicles are released into the external environment as a result of exocytosis. The first enzyme released is hyaluronidase. Hyaluronidase allows the sperm to pass through the corona radiata layer, while acrosin and trypsin-like substances dissolve the zona pellucida, allowing the sperm to reach the vitellus. In addition to these enzymes, aryl sulfatase A and B, which play a role in fertilization but whose functions and fine structures are not fully known, are also among the acrosomal enzymes. Sperm entry occurs through the penetration gap formed in the oocyte membrane with the hyperactivation resulting from sperm capacitation. At this time, the vitellus cortical granules in the inner membrane of the oocyte are released into the perivitelline space, forming the "fertilization membrane" that prevents polyspermy (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995). After the sperm enters the oocyte, the oocyte, which is waiting in metaphase 2, completes the 2nd meiosis. With the expulsion of the second polar body, the sperm pronucleus and the oocyte pronucleus fuse, forming the zygote (Sadler and Langman, 2010; Gartner and Hiatt, 1997; Ross et al., 2003; Carlos et al., 1995).

REFERENCES

Andrews JC, Nolan JP, Hammerstedt RH, et al. Role of zinc during hamster sperm capacitation. Biol Reprod 51, 1238-1247 (1994).

Austin CR. The "capacitation" of the mammalian sperm. Nature. 1952;170:326. 14. Chang MC. Fertilizing capacity of spermatozoa deposited into the fallopian tubes. Nature.1951;168:697–698.

Brewis IA, Moore HD, Fraser LR, et al. Molecular mechanisms during sperm capacitation. Hum Fertil (Camb). 2005 Dec;8(4):253-61.

Carlos Junqueira L, Carneiro Jose, Kelley Robert O. Temel Histoloji: a Lange medical book, 1995, sf407-413

Cross NL. Role of cholesterol in sperm capacitation. Biol Reprod. 1998;59:7–11.

Flesch FM, et al. Bicarbonate stimulated phospholipid scrambling induces cholesterol redistribution and enables cholesterol depletion in the sperm plasma membrane. J Cell Sci.2001;114:3543–3555.

Gadella BM, Harrison RA. The capacitating agent bicarbonate induces protein kinase A dependent changes in phospholipid transbilayer behavior in the sperm plasma membrane.Development. 2000;127:2407–2rolda0.

Gartner LP, Hiatt JL. Color Textbook of Histology: Pennsylvania, W.B. Saunders Company, 1997, pp 406-412.

Gerbers DL, Tubb DJ, Hyne RV. Requirement of bicarbonate for Ca2+-induced elevations of cyclic AMP in guinea pig spermatozoa. J Biol Chem 257, 1982;8980-8984

Gordon, I, Lu KH. Production of embryos in vitro and its impact on livestock production. Theriogenology, 1990; 33, 77-87.

Gulaya NM, Margitich VM. Phospholipid composition of human sperm and seminal plasma in relation to sperm fertility. Arch Androl. 2001 May-Jun;46(3):169-75.

Katz AM, Rosenthal D, Sauder DN. Cell adhesion molecules. Structure, function, and implication in a variety of cutaneous and other pathologic conditions. Int J Dermatol. 1991 Mar;30(3):153-60.

Keith LM, Persaud TVN. Nobel kitapevi. İnsan embriyolojisine giriş, 2008.

Lin Y, Kan FW. Regionalization and redistribution of membrane phospholipids and cholesterol in mouse spermatozoa during in vitro capacitation. Biol Reprod. 1996;55:1133 1146.

Mc Rorie, Williams WL. Biochemistry of mammalian fertilization. Ann. Rev. Bidchem. 43 : 777, 1974.

Moghissi GS: The function of the cervix in human reproduction, Year Book Medical Publishers, Chicago, 1984.

N. J. Monks, D. M. Stein & L. R. Fraiser: Adenylate cyclase activity of mouse sperm during capacitation in vitro: effect of calcium and a GTP analogue. Int J Androl 9, 1986;67-76.

Niu DM, Wang JJ. Lipids in the sperm plasma membrane and their role in fertilization, Zhonghua Nan Ke Xue. 2009 Jul;15(7):651-5. Review. Chinese.

Poulos A, White IG. The phospholipid composition of human spermatozoa and seminal plasma. J Reprod Fertil. 1973 Nov;35(2):265-72.

Chapter 2

Resin Cements

Berna SADİOĞLU[1]

The development of adhesive resin cements began in the 1960s following Bowen's introduction of BIS-GMA (bisphenol A-glycidyl methacrylate)(Silva e Souza et al., 2010). Initially, their clinical adoption was limited due to concerns such as potential pulpal damage, insufficient mechanical strength, and marginal adaptation issues. However, advancements in material science have led to improvements in their mechanical properties, enhanced bonding to dentin, and optimized chemical formulations, making them widely used materials in contemporary dental practice(Manso et al., 2011).

Resin cements exhibit high compressive and tensile strength and demonstrate excellent adhesion to both tooth and porcelain surfaces. Compared to other types of dental cements, they possess significantly lower solubility. These advantageous properties have led to their recognition as one of the most optimal cement types in modern dentistry, with increasing use in clinical practice.(Stamatacos & Simon, 2013)

They adhere to the organic components of dentin via micromechanical interaction and to the inorganic components through chemical bonding. While they share similar characteristics with composite resins, resin cements typically contain a lower proportion of filler particles. Like composites, their bonding mechanism involves the formation of a hybrid layer.(Jivraj et al., 2006)

Resin cements are primarily composed of methyl methacrylate (MMA), Bis-GMA, or UDMA as their base monomers. They typically include 20–80% by weight of inorganic fillers such as colloidal silica or barium glass. Compared to composite resins, resin cements generally exhibit lower viscosity and contain a reduced amount of filler particles(Haddad et al., 2011).

[1]

Based on their polymerization mechanisms, resin cements can be categorized as:

1. Light-cure resin cements
2. Self-cure (chemically cured) resin cements
3. Dual-cure resin cements (both light and chemical curing)

According to the adhesive system used, they are further classified into:

1. Total-etch resin cements
2. Self-etch resin cements
3. Self-adhesive resin cements(Burgess et al., 2010; Miotti et al., 2020)

Classification of Resin Cements Based on Polymerization Mechanism a. Light-Cure Resin Cements:

These cements are formulated as a single paste containing a photosensitive initiator, camphorquinone, and an aliphatic amine as a co-initiator. Polymerization is initiated when visible light in the wavelength range of 420–450 nm activates the camphorquinone, which, in the presence of the aliphatic amine, generates free radicals to start the reaction. Light-cure resin cements are primarily used for the luting of translucent ceramic and composite restorations that are no thicker than 1.5–2 mm—allowing sufficient light penetration.

A major advantage of light-cure cements is their extended working time, which enables clinicians to effectively remove excess material before setting occurs. In addition, they demonstrate superior color stability compared to other types of resin cements, making them particularly suitable for esthetically demanding restorations (Peumans et al., 2000).

Manufacturer	**Cement Name**	**Adhesion Modality**	**Functional Monomer**	**Suggested Adhesive**
Bisco	Choice 2	Adhesive	//	//
Bisco	eCement	Adhesive	//	All bond Universal
Dentsply Sirona	Calibra Veneer	Adhesive	//	Prime&Bond active
DMG	Vitique	Adhesive	//	//
GC	G-Cem Veneer	Adhesive	Silane	G-Premio Bond
Ivoclar	Variolink Esthetic	Adhesive	//	Adhese universal
Kuraray Noritake	Panavia Veneer LC	Adhesive	//	//
Ultradent	PermaShade LC	Adhesive	//	Peak Universal Bond
3M ESPE	RelyX Veneer Cement	Adhesive	//	Adper Single Bond Plus

Table1: Currently available definitive light cure resin-based cements per each manufacturer. The present list is not exhaustive and is based predominantly on

the European market. Any comission of a product is unintentional. Product information was retrieved from currently available SDS sheets and manufacturers' websites.

b. Self-Cure Resin Cements (Chemically Cured):

These cements are supplied in a two-paste system. Polymerization is initiated by the reaction between an organic peroxide (initiator) and a tertiary amine (accelerator), resulting in the formation of free radicals. Despite their efficacy in clinical settings where light cannot penetrate, self-cure cements have several drawbacks. These include limited working time, difficulty in controlling the setting process, and the potential for long-term discoloration due to the presence of amine groups in their formulation. They are primarily used for the cementation of restorations that block light transmission, such as metal or metal-ceramic inlays/onlays, adhesive bridges, endodontic posts, and opaque ceramics or composites (Manso et al., 2011).

c. Dual-Cure Resin Cements (Light and Chemically Cured):

Also delivered as a two-paste system, dual-cure resin cements undergo polymerization via both light activation and chemical reaction. The base paste typically contains a light-sensitive initiator such as camphorquinone, while the catalyst paste includes a peroxide-amine system to ensure chemical curing. These cements are ideal for luting restorations thicker than 1.5–2 mm or those with limited translucency, where sufficient light penetration is not possible.

Polymerization begins with the exposure to visible light reaching the cement layer and is subsequently completed by the chemical reaction. The slower reaction rate of the peroxide-amine component helps delay premature polymerization. Most dual-cure resin cements are translucent, allowing them to mimic the color of surrounding tissues and underlying tooth structure, which enhances esthetic integration. They are widely used for cementing all-ceramic crowns, veneers, and both ceramic and composite inlays/onlays(Pegoraro et al., 2007).

Manufacturer	Cement Name	Adhesion Modality	Functional Monomer
Bisco	Duo-link Universal	Adhesive	//
Bisco	BisCem	Self-adhesive	//
Bisco	TheraCem	Self-adhesive	10-MDP
Coltene/Whaledent	Paracore	Adhesive and Core cement	//
Coltene/Whaledent	Duocem	Adhesive	//
Coltene/Whaledent	Solocem	Adhesive or Self-adhesive	10-MDP, 4-META
Dentsply Sirona	Calibra Ceram	Adhesive	PENTA
Dentsply Sirona	Calibra Universal	Self-adhesive	PENTA
DMG	PermaCem Universal	Adhesive	//
DMG	PermaCem	Adhesive	//
DMG	PermaCem 2.0	Self-adhesive	10-MDP
GC	G-Cem Linkforce	Adhesive	//
GC	G-Cem Capsule	Self-adhesive	4-META
GC	G-Cem ONE	Self-adhesive	10-MDP
Heraeus Kulzer	iCEM	Self-adhesive	//
Ivoclar	Variolink Esthetic	Adhesive	//
Ivoclar	Multilink Automix	Adhesive	//
Ivoclar	SpeedCEM Plus	Self-adhesive	10-MDP
Kerr	Nexus Universal	Adhesive	//
Kerr	Maxcem Elite	Self-adhesive	GPDMA
Kerr	Maxcem Elite Chroma	Self-adhesive	//
Kuraray Noritake	Panavia V5	Adhesive	//
Kuraray Noritake	Panavia F 2.0	Adhesive	10-MDP
Kuraray Noritake	Panavia 21	Adhesive	//
Kuraray Noritake	Panavia SA Universal	Adhesive or Self-adhesive	10-MDP LCSi
Shofu	ResiCem	Adhesive	//
Shofu	BeautyCem SA	Self-adhesive	Phosphonic acid monomers
Ultradent	PermaFlo DC	Adhesive	//
3M ESPE	RelyX Universal	Adhesive or Self-adhesive	Mixture of GPDMA, bisGPDMA and trisGPDMA
3M ESPE	RelyX Unicem 2	Self-adhesive	Mixture of GPDMA, bisGPDMA and trisGPDMA

Table2: Currently available definitive dual cure resin-based cements per each manufacturer.The present list is not exhaustive and is based predominantly on the European market. Any comission of a product is unintentional. Product

information was retrieved from currently available SDS sheets and manufacturers' websites.
Abbreviations:

- 10-MDP: 10-Methacryloyloxydecyl dihydrogen phosphate
- 4-META: 4-Methacryloxyethyl trimellitic anhydride
- bisGPDMA: Bis(glyceryldimethacrylate) phosphate
- GPDMA: Glycerol phosphate dimethacrylate
- LCSi: Long carbon-chain silane coupling agent
- PENTA: Dipentaerythritol penta-acrylate phosphate
- trisGPDMA: Tris(glyceryldimethacrylate) phosphate

Classification of Resin Cements According to Adhesive Systems

a. Total-Etch Resin Cements:

These are two- or three-step adhesive systems in which enamel and dentin are conditioned with 30–40% phosphoric acid for varying durations. Acid etching of the enamel creates microporosities approximately 5–50 µm in depth, facilitating mechanical retention. In dentin, acid application removes the smear layer and induces demineralization to a depth of 5–10 µm, thereby exposing the dentinal tubules. However, over-drying the dentin may lead to collapse of the collagen network, which can compromise bonding. To prevent this, the dentin surface is carefully dried and kept slightly moist.

The primer application step is critical, as it enhances the wettability of the dentin. Hydrophilic monomers in the primer displace water within the collagen network, creating a hydrophobic surface conducive to adhesive penetration. Following primer application, an adhesive resin containing hydrophobic monomers such as Bis-GMA, UDMA, or TEGDMA is applied. These systems are compatible with both light-cure and dual-cure resin cements.(*Ferracane et Al*, n.d.)

Total-etch resin cements generally provide the highest bond strength to tooth structures. However, due to their multiple steps and technique sensitivity, achieving optimal bonding can be challenging. As a response to these limitations, simplified alternatives such as self-etch and self-adhesive resin cements have been developed.(Burgess et al., 2010)

b. Self-Etch Resin Cements:

Postoperative sensitivity associated with total-etch systems is thought to result from the incomplete sealing of exposed dentinal tubules. To address this issue, self-etch adhesive systems were developed(Stangel et al., 2007). In self-etch systems, a primer is applied directly to the tooth surface to prepare it, followed by the application of the resin cement. Studies have shown that the bond strength achieved with self-etch resin cements is comparable to that of total-etch systems.(*Ferracane et Al*, n.d.)

c. Self-Adhesive Resin Cements:

Self-adhesive resin cements, a relatively recent development in the dental materials market, are typically dual-cure systems. They were designed to simplify the cementation process and eliminate some of the drawbacks associated with both conventional and resin-based cements.(Burgess et al., 2010)

These cements allow for a one-step application process, eliminating the need for separate tooth surface treatments such as microabrasion, etching, priming, or bonding. Their formulation includes phosphoric acid incorporated into the resin matrix, along with acrylic or diacrylic monomers and acidic adhesive monomers that enable intrinsic bonding to dental hard tissues. This allows chemical interaction between the cement and hydroxyapatite(Burgess et al., 2010).

Since phosphoric acid etching is not performed separately, the depth of demineralization and resin infiltration remains uniform, thereby reducing the risk of nanoleakage. Additionally, because the smear layer remains intact and dentinal tubules are not exposed, low molecular weight acids are less likely to penetrate the dentin. As a result, self-adhesive resin cements offer a fast, user-friendly, single-step application with minimal risk of postoperative sensitivity.

However, compared to total-etch systems, these cements generally exhibit lower bond strength to tooth structures. This is primarily due to the limited demineralization effect on the smear layer, which restricts micromechanical retention to enamel and dentin(Al-Assaf et al., 2007; *Ferracane et Al*, n.d.; Gerth et al., 2006).

Studies have shown that the bond strength of self-adhesive resin cements to enamel is generally weaker than to dentin. This phenomenon is attributed to the presence of phosphoric methacrylate within the cement, which reacts with water in the tooth structure to create a low-pH environment. Since dentin is more susceptible to acid etching than enamel, this acidic condition promotes greater demineralization in dentin, resulting in stronger bonding(Burgess et al., 2010).

To enhance bonding to enamel, selective etching of the enamel surface with phosphoric acid has been shown to nearly double the bond strength(Burgess et al., 2010). Other studies also report that pre-treatment with phosphoric acid followed by the application of a bonding agent significantly improves adhesion to enamel. (Burgess et al., 2010; Carvalho et al., 2004; De Munck et al., 2004; Peumans et al., 2007; Van Landuyt, Kanumilli, et al., 2006; Van Landuyt, Peumans, et al., 2006; Van Meerbeek et al., 2005)

Some self-adhesive resin cements exhibit fluoride release when they react with acidic environments. These cements typically maintain a near-neutral pH during the setting reaction. The presence of water in the cement initially imparts hydrophilic properties, which assist in neutralizing the low pH. As the water is gradually consumed, the cement forms a hydrophobic matrix with low solubility, minimal expansion, and long-term dimensional stability.

In addition to micromechanical retention, self-adhesive resin cements can also establish weak chemical bonds with calcium ions in the tooth structure, contributing to their overall adhesive performance.(Burgess et al., 2010)

Universal Cements

While self-adhesive cements were originally introduced to streamline clinical workflows without compromising the durability of restorations, their performance in bonding esthetic partial restorations has often been inconsistent. In most cases, surface pretreatment is still necessary—typically involving a silane coupling agent for ceramic or composite surfaces, or a 10-MDP primer for zirconia. To address the variety of clinical situations and the confusion caused by the wide array of products available, some manufacturers have developed resin cements that can be used either in a simplified self-adhesive mode or, when paired with compatible universal adhesives, in a conventional adhesive protocol. These cement-adhesive systems are designed for versatility, allowing their use with metal, composite, ceramic, and zirconia restorations without requiring additional primers. As a result, they can be considered truly "universal" in their application.

A truly universal cement should meet the following criteria:

1. It must be suitable for use in a variety of luting protocols, including self-adhesive, and adhesive techniques such as self-etch, selective enamel etch, or etch-and-rinse, based on clinical demands and operator preference.
2. It should be compatible with bonding to both dental hard tissues and a wide range of restorative materials, including metal, silica-based ceramics, and zirconia.
3. The cement should be paired with a universal adhesive resin that serves as a primer for both the tooth structure and restorative surfaces, eliminating the need for additional priming agents.
4. At least one component of the cement or adhesive system must include functional acidic monomers, and ideally silane coupling agents, to chemically link the tooth and restorative material surfaces.
5. The material should possess dual-cure capability to ensure adequate polymerization in all clinical scenarios.(Maravić et al., 2023)

Types of Definitive Resin-Based Cements: A Comparative Overview

Cement Type	Advantages	Disadvantages
Adhesive / Multi-Step Cements	✓ Proven long-term clinical success for full crowns, partial restorations, and veneers.	✗ Technique-sensitive application. ✗ Requires separate primers depending on the restorative material.
Self-Adhesive / One-Step Cements	✓ Easier to apply compared to multi-step systems. ✓ Clinically reliable for full crowns.	✗ Less effective for partial restorations and not suitable for veneers. ✗ Still requires primers for certain materials. ✗ Higher susceptibility to moisture-related degradation.
Universal Cements	✓ Highly versatile—can be used across various clinical conditions. ✓ Simplified handling in self-adhesive mode. ✓ When needed, only one primer (universal adhesive) is required.	✗ Lack of established long-term clinical data. ✗ Lower mechanical strength and more water sorption than other types. ✗ Limited number of supporting studies.

FIGURE 1 Novel classification and an overview of the properties of definitive resin-based cements proposed according to the current status perspective: adhesive cements (namely multi-steps), self-adhesive cements (traditionally one-step) and universal cements (a combination of the two luting modes according to the clinical requirements).

Universal cements represent an evolution of self-adhesive resin cements, building upon a similar underlying principle. They are typically provided as a two-paste system, which separates key chemical components—such as the oxidizing and reducing agents, acidic and nearly neutral elements, and hydrophilic and hydrophobic substances.

The updated formulations in these advanced cements are designed to work synergistically with their corresponding universal adhesives. This approach minimizes the risk of curing incompatibilities and enhances their effectiveness in bonding with both tooth structures and restorative materials.(Maravić et al., 2023)

To ensure reliable adhesion to dental tissues, universal cements incorporate functional acidic monomers into their formulations. These monomers are designed to both etch and penetrate the tooth structure, enabling chemical interaction with calcium ions (Ca^{2+}) in hydroxyapatite and with methacrylate-based monomers—essentially mimicking a “silanization” effect on the tooth surface.

Various functional monomers are utilized in current dental cement technologies, including 4-methacryloxyethyl trimellitic acid (4-MET), 4-methacryloxyethyl trimellitic anhydride (4-META), and dipentaerythritol penta-acrylate phosphate (PENTA).

However, the most widely recognized and effective monomer is 10-methacryloyloxydecyl dihydrogen phosphate (10-MDP). This compound, which features a phosphoric acid group, a spacer, and a methacrylate end, forms highly stable and strong bonds with hydroxyapatite by generating 10-MDP-Ca salts.(Yoshihara et al., 2018) It also creates hydrogen bonds with collagen fibrils.

On the other end of the molecule, the methacrylate group covalently bonds with the resin matrix. Furthermore, both 10-MDP and its calcium salts have been shown to suppress endogenous dentinal matrix metalloproteinases (MMPs), enzymes known to degrade the hybrid layer and compromise the durability of the resin-dentin interface.(Breschi et al., 2018; Mokeem et al., 2023)

REFERENCES

Al-Assaf, K., Chakmakchi, M., Palaghias, G., Karanika-Kouma, A., & Eliades, G. (2007). Interfacial characteristics of adhesive luting resins and composites with dentine. *Dental Materials : Official Publication of the Academy of Dental Materials*, *23*(7), 829–839.

Breschi, L., Maravic, T., Cunha, S. R., Comba, A., Cadenaro, M., Tjäderhane, L., Pashley, D. H., Tay, F. R., & Mazzoni, A. (2018). Dentin bonding systems: From dentin collagen structure to bond preservation and clinical applications. *Dental Materials : Official Publication of the Academy of Dental Materials*, *34*(1), 78–96.

Burgess, J. O., Ghuman, T., & Cakir, D. (2010). Self-adhesive resin cements. *Journal of Esthetic and Restorative Dentistry : Official Publication of the American Academy of Esthetic Dentistry ... [et Al.]*, *22*(6), 412–419.

Cadenaro, M., Maravic, T., Comba, A., Mazzoni, A., Fanfoni, L., Hilton, T., Ferracane, J., & Breschi, L. (2019). The role of polymerization in adhesive dentistry. *Dental Materials : Official Publication of the Academy of Dental Materials*, *35*(1), e1–e22.

Carvalho, R. M., Pegoraro, T. A., Tay, F. R., Pegoraro, L. F., Silva, N. R. F. A., & Pashley, D. H. (2004). Adhesive permeability affects coupling of resin cements that utilise self-etching primers to dentine. *Journal of Dentistry*, *32*(1), 55–65.

De Munck, J., Vargas, M., Van Landuyt, K., Hikita, K., Lambrechts, P., & Van Meerbeek, B. (2004). Bonding of an auto-adhesive luting material to enamel and dentin. *Dental Materials: Official Publication of the Academy of Dental Materials*, *20*(10), 963–971.

Dimitriadi, M., Petropoulou, A., Masouras, K., Zafiropoulou, M., Zinelis, S., & Eliades, G. (2021). The Effect of Touch-Cure Polymerization on the Conversion and Hardness of Core Build-Up Resin Composites: A Laboratory Study. *Materials*, *14*(20), 6025.

Ferracane et al. (n.d.). https://doi.org/10.1111/j.1365-2842.2010.02148.x

Gerth, H. U. V., Dammaschke, T., Züchner, H., & Schäfer, E. (2006). Chemical analysis and bonding reaction of RelyX Unicem and Bifix composites--a comparative study. *Dental Materials : Official Publication of the Academy of Dental Materials*, *22*(10), 934–941.

Haddad, M. F., Rocha, E. P., & Assunção, W. G. (2011). Cementation of prosthetic restorations: from conventional cementation to dental bonding concept. *The Journal of Craniofacial Surgery*, *22*(3), 952–958.

Jivraj, S. A., Kim, T. H., & Donovan, T. E. (2006). Selection of luting agents, part 1. *Journal of the California Dental Association*, *34*(2), 149–160.

Madruga, F. C., Ogliari, F. A., Ramos, T. S., Bueno, M., & Moraes, R. R. (2013). Calcium hydroxide, pH-neutralization and formulation of model self-adhesive resin cements. *Dental Materials : Official Publication of the Academy of Dental Materials*, *29*(4), 413–418.

Manso, A. P., & Carvalho, R. M. (2017). Dental Cements for Luting and Bonding Restorations: Self-Adhesive Resin Cements. *Dental Clinics of North America*, *61*(4), 821–834.

Manso, A. P., Silva, N. R. F. A., Bonfante, E. A., Pegoraro, T. A., Dias, R. A., & Carvalho, R. M. (2011). Cements and adhesives for all-ceramic restorations. *Dental Clinics of North America*, *55*(2), 311–332, ix.

Maravić, T., Mazzitelli, C., Mancuso, E., Del Bianco, F., Josić, U., Cadenaro, M., Breschi, L., & Mazzoni, A. (2023). Resin composite cements: Current status and a novel classification proposal. *Journal of Esthetic and Restorative Dentistry : Official Publication of the American Academy of Esthetic Dentistry ... [et Al.]*, *35*(7), 1085–1097.

Miotti, L. L., Follak, A. C., Montagner, A. F., Pozzobon, R. T., da Silveira, B. L., & Susin, A. H. (2020). Is Conventional Resin Cement Adhesive Performance to Dentin Better Than Self-adhesive? A Systematic Review and Meta-Analysis of Laboratory Studies. *Operative Dentistry*, *45*(5), 484–495.

Mokeem, L. S., Garcia, I. M., & Melo, M. A. (2023). Degradation and Failure Phenomena at the Dentin Bonding Interface. *Biomedicines*, *11*(5). https://doi.org/10.3390/biomedicines11051256

Pegoraro, T. A., da Silva, N. R. F. A., & Carvalho, R. M. (2007). Cements for use in esthetic dentistry. *Dental Clinics of North America*, *51*(2), 453–471, x.

Peumans, M., De Munck, J., Van Landuyt, K., Lambrechts, P., & Van Meerbeek, B. (2007). Five-year clinical effectiveness of a two-step self-etching adhesive. *The Journal of Adhesive Dentistry*, *9*(1), 7–10.

Peumans, M., Van Meerbeek, B., Lambrechts, P., & Vanherle, G. (2000). Porcelain veneers: a review of the literature. *Journal of Dentistry*, *28*(3), 163–177.

Radovic, I., Monticelli, F., Goracci, C., Vulicevic, Z. R., & Ferrari, M. (2008). Self-adhesive resin cements: a literature review. *The Journal of Adhesive Dentistry*, *10*(4), 251–258.

Roedel, L., Bednarzig, V., Belli, R., Petschelt, A., Lohbauer, U., & Zorzin, J. (2017). Self-adhesive resin cements: pH-neutralization, hydrophilicity, and hygroscopic expansion stress. *Clinical Oral Investigations*, *21*(5), 1735–1741.

Silva e Souza, M. H., Jr, Carneiro, K. G. K., Lobato, M. F., Silva e Souza, P. de A. R., & de Góes, M. F. (2010). Adhesive systems: important aspects related to their composition and clinical use. *Journal of Applied Oral Science : Revista FOB*, *18*(3), 207–214.

Stamatacos, C., & Simon, J. F. (2013). Cementation of indirect restorations: an overview of resin cements. *Compendium of Continuing Education in Dentistry (Jamesburg, N.J. : 1995)*, *34*(1), 42–44, 46.

Stangel, I., Ellis, T. H., & Sacher, E. (2007). Adhesion to tooth structure mediated by contemporary bonding systems. *Dental Clinics of North America*, *51*(3), 677–694, vii.

Suh, B. I., Feng, L., Pashley, D. H., & Tay, F. R. (2003). Factors contributing to the incompatibility between simplified-step adhesives and chemically-cured or dual-cured composites. Part III. Effect of acidic resin monomers. *The Journal of Adhesive Dentistry*, *5*(4), 267–282.

Van Landuyt, K. L., Kanumilli, P., De Munck, J., Peumans, M., Lambrechts, P., & Van Meerbeek, B. (2006). Bond strength of a mild self-etch adhesive with and without prior acid-etching. *Journal of Dentistry*, *34*(1), 77–85.

Van Landuyt, K. L., Peumans, M., De Munck, J., Lambrechts, P., & Van Meerbeek, B. (2006). Extension of a one-step self-etch adhesive into a multi-step adhesive. *Dental Materials : Official Publication of the Academy of Dental Materials*, *22*(6), 533–544.

Van Meerbeek, B., Kanumilli, P., De Munck, J., Van Landuyt, K., Lambrechts, P., & Peumans, M. (2005). A randomized controlled study evaluating the effectiveness of a two-step self-etch adhesive with and without selective phosphoric-acid etching of enamel. *Dental Materials : Official Publication of the Academy of Dental Materials*, *21*(4), 375–383.

Vrochari, A. D., Eliades, G., Hellwig, E., & Wrbas, K. T. (2010). Water sorption and solubility of four self-etching, self-adhesive resin luting agents. *The Journal of Adhesive Dentistry*, *12*(1), 39–43.

Yoshihara, K., Hayakawa, S., Nagaoka, N., Okihara, T., Yoshida, Y., & Van Meerbeek, B. (2018). Etching Efficacy of Self-Etching Functional Monomers. *Journal of Dental Research*, *97*(9), 1010–1016.

Chapter 3

The Role of *Demodex* spp. Mites on Dermatological Diseases and Their Effects on Human Health

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Demodex spp. is known as an ectoparasite that usually settles on the facial area of the body such as forehead, nose, cheeks and chin. The disease caused by this parasitic agent is called demodicosis. *Demodex* species cause infestation by spreading to large geographical areas worldwide. Information on 288 articles on *Demodex* spp. mites was obtained from WOS databases and it was determined that the countries publishing the most articles on this subject were USA (24%), China (13%) and Türkiye (11%). The most articles were published in Ophthalmology (63%), Dermatology (7.74%), General Internal Medicine (7%) and Parasitology (4%), respectively (Lacey et al., 2016; Yıldız et al., 2024).

Demodicosis, which has a cosmopolitan distribution, can also be seen in cold climates and desert regions. To date, 70 different *Demodex* species have been identified and it has been determined that these species can cause infestation and pathogenic effect on many animal species such as human, horse, sheep, cattle, goat, pig, rabbit, cat and dog (Ütük and Dumanlı, 2015). *Demodex folliculorum* and *Demodex brevis* cause this infestation in humans. *D. folliculorum* is known to have a longer life span and to live in groups. This species settles in the infundibulum of the hair follicles and feeds on fatty substances. *D. brevis* feeds on sebaceous and meibomian glands (Liu et al., 2010, Koo et al., 2012, Lacey et

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al., 2016, Schear ve al., 2016, Sedzikowska et al., 2016, Litwin et al., 2017; Afşar et al., 2024).

These mites are usually found in the hair follicles and at the base of the sebaceous glands in the skin, especially around the nose, nose wings, cheeks and mouth rastlanır (Akilov & Mumcuoglu, 2003; Liu et al., 2010; Chen and Plewig, 2015). Less commonly, it can be found on the forehead, chin, eyebrows, eyelashes, eyelashes, ears and scalp, and rarely on the outside of the face, especially on the perineum. The severity of this parasitosis can vary with age. According to many researchers, this mite, which can be transmitted due to factors such as the use of common items, close contact and lack of attention to hygiene rules, can cause the emergence of some clinical symptoms with the increase in its number in the regions where it is found (Akilov and Mumcuoglu, 2003; Liu et al., 2010). Bu akarların direkt yolla hastalık yapıcı etkisi olmamasına rağmen bağışıklık sistemi zayıf bireylerde akne rozasea, akne vulgaris ve seboreik dermatit gibi bazı cilt problemlerine neden olabileceği bildirilmiştir (Özcel et al., 2007a; Özcel et al., 2007b; Chen and Plewig, 2015).

These mites can parasitise many animal species other than humans. Especially domestic animals such as dogs and cats, farm animals such as horses, cattle, sheep, goats, goats, pigs and rabbits, wild animals such as deer, rodents such as hamsters, rats and mice, and bats are among the creatures affected by these parasites. Studies have shown that *Demodex* species have a pathogenic effect on the development of some skin and eye diseases (Erbagci and Özgoztası, 1998; Akilov and Mumcuoglu, 2003; Czepita et al., 2007; Kim et al., 2011; Lindsley et al., 2012; Szkaradkiewicz et al., 2012; Chen and Plewig, 2015; Cheng et al., 2015; Tarkowski et al., 2015; Afşar et al., 2024).

Some studies show that insufficient cleaning of these products after the use of cosmetic products, not paying attention to facial cleansing, increased sebum secretion with sweating in hot weather and skin lubrication may cause *Demodex* species to become pathogenic. In addition, factors such as steroid use, ageing and weakening of the immune system may increase the pathogenic effects of these mites. For these reasons, *Demodex* species play an important role in the development of skin diseases such as acne vulgaris, rosacea, perioral dermatitis, seborrhoeic dermatitis and, rarely, blepharitis with an increase in follicles in the eye. Public awareness is very important in the effective control of these mites (Oytun, 1969; Bonnar et al., 1993; Unat et al., 1995; Özcel et al., 2007a; Tanrıverdi et al., 2018; Çelik and Kaya, 2020; Cheng et al., 2021; Afşar et al., 2024).

History

Demodex, which means 'fat woodworm' in Latin, was first identified in history by Jakob Henle in 1841. Gustav Simon described these mites as *Acarus folliculorum* in 1842, and Richard Owen changed the genus name to *Demodex* in 1843. The pathogenic effects of *Demodex* species were first reported by Borrel in 1908, their biological properties were described by Gmeiner in the same period and the parasitosis they cause was named 'demodicosis'. Lawrence demonstrated the association of demodicosis with facial diseases in 1916, and three years later Hirst made a taxonomic classification of *D. folliculorum*. Chamber and Somerset reported the association of these mites with cancer in 1925, and Fuss analysed their life cycle and incidence according to skin conditions in 1933. Beerman and Stokes studied the pathogenesis of *D. folliculorum* in 1934, especially its relationship with rosacea, and Spickett studied the behaviour and biology of the mite in 1961. Nutting studied the host-parasite relationship in 1965 and Akbulatova described two subspecies of *Demodex* in humans in 1966. Desch and Nutting studied the characteristics of these two species in 1972 and found that *D. brevis* survives in the sebaceous glands of human skin (Coston, 1967; Nutting, 1976; Rufli et al., 1981; Özcel et al., 2007a; Czepita et al., 2007, Tarkowski et al., 2015, Lacey et al., 2016).

Taxonomy

The taxonomy of *D. folliculorum* and *D. brevis* parasitising humans is as follows (Sahn and Sheridan, 1992; Forton and Seys, 1993; Murube, 2015). To date, 70 species in the genus *Demodex* and more than 50 families in the Prostigmata group have been described. Among these, there are important ectoparasite species especially in the Demodicidae family (Ütük and Dumanlı, 2015).

Division: Arthropoda
- Subdivision: Chelicerata
  - Class: Arachnida
    - Subclass: Acari
      - Order: Trombidiformes
        - Suborder: Prostigmata
          - Family: Demodicidae
            - Genus: *Demodex*
              - Species: *Demodex folliculorum*
              - Species: *Demodex brevis*

Morphology

D. folliculorum and *D. brevis* are microscopic mites that inhabit the hair follicles and sebaceous glands of humans. Males of *D. folliculorum* have a mean length of 279.8 ± 52.0 µm, with a gnathosoma length of 19.5 ± 0.9 µm and a width of 24.2 ± 0.9 µm. Podosoma length 67.6 ± 2.7 µm, width 45.1 ± 2.0 µm; opisthosoma length 190 ± 49.9 µm, width 32.6 ± 1.6 µm. Females have a mean length of 294.2 ± 58.8 µm, gnathosoma length 21.3 ± 0.7 µm, width 26.5 ± 1.0 µm. Podosoma length 75.0 ± 2.8 µm, width 51.9 ± 3.0 µm; opisthosoma length 197.9 ± 55.7 µm, width 40.5 ± 3.4 µm. The head of these mites has two clawed dagger-shaped chelicerae and 2-3 jointed pedipalps. Each leg terminates in a caecum and two claws. The genital opening of female mites is on the abdomen between the fourth pair of legs, while the genital opening of males is on the back at the level of the second pair of legs. The body structures of the developmental stages of *Demodex* species differ markedly. The triangular egg is approximately 104.8 ± 6.3 µm long, 41.7 ± 1.9 µm wide and 37.7 ± 2.5 µm deep. The larvae have three pairs of legs and average 282.7 ± 45.1 µm in length and 33.5 ± 2.6 µm in width. The next developmental stage, protonymphs, reach a length of approximately 364.8 ± 36.3 µm and a width of 36.4 ± 4.4 µm. The nymphal stage, characterised by four pairs of legs and a body surface resembling a chestnut shell, is on average 392.1 ± 46.9 µm long and 41.8 ± 6.2 µm wide (Desch and Nutting, 1972; Mumcuoğlu, 2004; Aksakal, 2014; Tarkowski et al., 2015; Lacey et al., 2016; Fromstein et al., 2018).

D. brevis, another species parasitising humans, has a shorter and wider body structure compared to *D. folliculorum*. The average body length is 165.7 µm in males and 208.6 µm in females. The gnathosoma of the head is 14.5 ± 0.5 µm long and 17.1 ± 1.4 µm wide in males and 16.3 ± 1.1 µm long and 19.2 ± 1.1 µm wide in females. The podosoma, the body part, is 54.4 ± 2.9 µm long and 46.0 ± 4.2 µm wide in males and 65.2 ± 2.3 µm long and 50.2 ± 3.4 µm wide in females. The pointed opisthosoma, which is located on the posterior part and constitutes about one third of the body length, is 97.1 ± 17.2 µm long and 39.8 ± 4.6 µm wide in males and 126.8 ± 25.0 µm long and 44.6 ± 7.7 µm wide in females. In males, the penis, located posteriorly on the dorsum, is approximately 17.6 ± 1.0 µm long, while in females the vulva on the abdomen is 6.9 ± 0.4 µm long. The immature developmental stages of *D. brevis* have distinct structural differences. When the morphometric characteristics of the developmental stages were analysed, the oval-shaped egg was approximately 60.1 ± 3.4 µm long and 34.4 ± 2.2 µm wide. The larvae with three pairs of legs are approximately 105.4 ± 11.5 µm long and 33.8 ± 4.0 µm wide. The subsequent protonymph is 147.6 ± 14.1 µm long and 34.4 ± 3.5 µm wide. The nymphs with four pairs of legs are approximately 165.0 ± 26.3 μm long and 41.2 ± 5.4 μm wide (Desch and Nutting 1972; Mumcuoğlu, 2004; Aksakal, 2014; Tarkowski et al., 2015; Lacey et al., 2016; Fromstein et al., 2018).

When the two species parasitising humans are compared, it is observed that *D. folliculorum* is larger than *D. brevis* at all stages of development. The opisthosoma of *D. folliculorum* has a rounded tip, whereas the same region of *D. brevis* is more pointed. The length of the opisthosoma accounts for about 70 per cent of the whole body length in *D. folliculorum*, but only one third in *D. brevis*. The eggs of *D. folliculorum* are large, triangular, plate-like, while those of *D. brevis* are smaller and oval. *D. folliculorum* has prominent chestnut-shaped structures on the legs, while these structures are absent in *D. brevis*. In *D. folliculorum*, the mouthparts are more developed and prominent, whereas this structure is less prominent in *D. brevis*. Identification of *Demodex* spp. according to their morphological characteristics has been reported to be important in dermatological diseases (Desch and Nutting, 1972; Saygı et al.,1984; Markell et al., 1992; Unat et al., 1995; Özcel et al., 2007a).

Life Cycle

Male and female *Demodex* spp. survive by utilising sebum and epithelial debris as food. These parasites mate on the skin surface and move towards the hair follicles. Larvae emerge from the eggs laid here and turn into protonymphs in 2-3 days. The deuteronymph form then develops and progresses towards the follicle mouth. The parasites that complete their development continue their life on the same host for about 10 days (Czepita et al., 2007; Rusiecka-Ziolkowska et al., 2014; Murube, 2015; Clanner-Engelshofen et al., 2018). The hair follicles and sebum glands of human skin, especially on the face, provide a suitable temperature and nutrient environment for the proliferation of *Demodex* spp. These mites penetrating on the skin disrupt the integrity of the skin and cause the entry of some bacterial agents. In this process, dermatological symptoms such as allergic reactions, rashes, acne formation and erythema may occur. Ophthalmological disorders such as blepharitis, itching and inflammation of the base of the eyelashes can also occur due to this infestation. In addition, damage to capillaries and tissue hyperplasia can lead to the formation of red, tumour-like swellings in the nasal area (Markell et al., 1992; Unat et al., 1995; Czepita et al., 2007; Rusiecka-Ziolkowska et al., 2014; Murube, 2015).

Epidemiology

Demodex spp., which is short-lived and survives on the host, is usually transmitted between humans through close physical contact. It can be seen in individuals of all races and in various parts of the world, and the prevalence of infestation increases with increasing age (Budak et Yolasığmaz, 1995; Wesolowska et al., 2014; Ütük and Dumanlı, 2015). *Demodex* spp. are thought to be rarely seen in paediatric age group due to low levels of sebum production. However, these mites have been found in rosacea-like lesions, especially in immunosuppression in children aged 2-6 years. In studies, it has been reported that the prevalence of infestations caused by *Demodex* spp. is quite low in people under 20 years of age, but this rate increases significantly with age. In a study investigating the relationship between the frequency of this mite and age, Demodex spp. was detected in approximately 80% of people aged 51-80 years and 90-100% of people aged 60-89 years (Akdeniz et al., 2002; Kulac et al., 2008, Kaya et al., 2012; Lacey et al., 2016; Lopez-Ponce et al., 2017; Sedzikowska et al., 2018; Afşar et al., 2024).

Demodex spp., which are sensitive to heat and dryness, can move approximately 8-16 cm per hour, showing a limited mobility in unlit environments. These mites especially prefer the regions where sebum production is intense and settle in the nasal skin follicles where sebaceous glands are abundant, the tip of the nose and the wings of the nose. In some cases, parasites were also found in the eyelash follicles, temporal region, eyebrows, scalp, external ear canal, nipple, pubic and gluteal areas, frontal region, thorax, back and arms. On the other hand, in the lower extremities and axillary regions, it is stated that the mites are not able to attach in these regions due to the greater distance between the hair follicles and intensive sweat production, and therefore infestation is rarely encountered in these areas (Sahn and Sheridan, 1992; Farina et al., 1998; Dolenc Voljc et al., 2005; Aycan, 2008; Tarkowski et al., 2015; Litwin et al., 2017; Zeytun et al., 2017).

Intensive use of cosmetic products and insufficient purification of the skin from these products, increased skin oil and sebum production with environmental temperature, and immunodeficiency as a result of steroid use are among the main factors that increase the pathogenicity of *Demodex*. However, a suppressed immune system, whether congenital or acquired, is an important risk factor that increases the severity of the infestation (Bonnar et al., 1993; Olt et al., 2013; Ütük and Dumanlı, 2015; Yazar et al., 2016, Litwin et al., 2017; Kaya et al., 2019).

Samples were taken by eyelash epilation method from clinically healthy individuals without any history of eye infection. As a result of the evaluation, *D. folliculorum* was detected in 12.97% and *D. brevis* in 3.05% of the participants (Kaya et al., 2012). Although *Demodex* infestation is quite common, it is often overlooked as a causative agent of ocular surface inflammation. However, in recent years, there has been a significant increase in the number of studies on ocular demodicosis and this has contributed to an increased awareness of the role of the parasite in ophthalmological pathologies (Cheng et al., 2015, Fromstein et al., 2018; Tanriverdi et al., 2018; Zeytun and Karakurt, 2019; Yıldız et al., 2024).

Immunology

It is known that immunocompromised individuals become vulnerable to obligate parasites such as *D. folliculorum* and *D. brevis* (Olt ve ark., 2013, Kaya ve ark., 2019). Demodicosis has been reported to be more common in adults and elderly people. In infestations caused by this mite, more prominent clinical findings are usually observed in immunocompromised individuals. Disorders in immune response mechanisms lead to pathological proliferation of *Demodex* mites and cause various skin lesions. In studies, it has been reported that findings such as rosacea and perioral dermatitis have been observed in children diagnosed with acute lymphoblastic leukaemia due to immunosuppression (Akilov and Mumcuoglu, 2004; Ütük and Dumanlı, 2015).

Suppression of T cells and dysfunction of the sebaceous glands play an important role in the development of demodicosis. This condition is rarely seen in paediatric patients and frequently in patients with AIDS and lymphoproliferative disorders (Chen ve ark., 2020). It has been reported that the infestation is more severe in immunocompromised persons receiving steroid treatment and in middle-aged and older persons with weakened immune systems. It has been reported that IgE antibody levels, which is an indicator of immune response, are statistically significantly higher in individuals with inadequate immune system due to colonisation and infections compared to healthy individuals and that the agents are observed more intensely in samples taken from the cheek area of these individuals (Akilov and Mumcuoglu, 2004; Lacey et al., 2009; Forton and De Maertelaer, 2017).

Studies have scientifically demonstrated the relationship between demodicosis and certain HLA Cw2 and HLA Cw4 haplotypes. It has been reported that the risk of developing clinical symptoms of the disease is 5 times higher in people with HLA Cw2 haplotype and 3.1 times higher in people with HLA Cw4 haplotype. People with HLA-A2 phenotype were reported to be 2.9 times more resistant to the mite. In the analyses performed on membrane ranges to evaluate the immune response, $CD95^+$ cell levels were found to be significantly higher in all patients. When leukocyte activities were analysed, it was found that there was a significant decrease in $CD3^+$, $CD4^+$, $CD8^+$ and $CD16^+$ cell levels and $CD3^+/CD20^+$ ratios, while there was no significant change in $CD20^+$ cell number and percentage, $CD4^+/CD8^+$ ratio, phagocytosis function, CH50 levels and serum

IgM, IgG and IgA antibody levels (Akilov and Mumcuoglu, 2003; Akilov and Mumcuoglu, 2004).

Infection

Demodex spp. infestation can be transmitted by direct skin contact as well as by the use of common objects contaminated with mites. In addition, dust particles containing mite eggs in the environment can also play an active role in the transmission of infestation (Czepita ve ark., 2007; Sedzikowska ve ark., 2016). Since *Demodex* spp. are host specific, species found in animals do not live in humans. However, prolonged contact with cats and dogs heavily infected with these mites may cause transient dermatitis in humans (Ütük and Dumanlı, 2015).

Pathogenesis and Clinical Findings

There are still different opinions about the pathogenesis of *D. folliculorum* and *D. brevis* in the literature (Kaya ve ark., 2012). It has been reported that people infected with *Demodex* spp. are usually asymptomatic, but in some cases, symptoms occur at different levels. The occurrence and severity of infestation may vary depending on the characteristics of the host (Murube, 2015).

The localisation of *D. folliculorum* and *D. brevis* on human skin differs. *D. folliculorum* is located in the infundibulum of the hair follicle, while *D. brevis* is located in the deep parts of the sebaceous glands. *D. folliculorum* causes superficial skin lesions and *D. brevis* causes papulopustular eruptions and crusting. Primary demodicosis occurs due to excessive mite density without pre-existing or concurrent inflammatory dermatoses. Inflammation of the sebaceous glands is characterised by follicular or macular inflammation of the skin around the lips, nostrils and eyes. The lesions usually start with erythematous rashes covering 10-15% of the facial area and itching, but they decrease with treatment. However, relapses may be seen with seasonal exacerbations. Secondary demodicosis is usually associated with systemic or local immune suppression. It presents with symptoms such as diffuse papulopustular rashes and burning sensation affecting 25-40% of the facial area, and the symptoms are persistent even if treated. Increased exacerbations in the summer months are thought to be associated with *D. brevis*. *Demodex* spp. has been reported to aggravate the course of dermatological diseases such as rosacea, blepharitis, pityriasis folliculorum, perioral dermatitis, eosinophilic folliculitis and pustular folliculitis. It has been reported that *Demodex* spp. are more frequently found on the skin of immunosuppressed, AIDS and diabetic patients (Forton and Seys, 1993; Forton et al.,2005; Chen and Plewig, 2015; Forton and Maertelaer, 2019).

These mites cause tissue damage by piercing the epithelial cells lining the hair follicles and sebaceous glands with their chisel-like chelicerae in order to feed on the cell contents. This process can result in mild to moderate hypertrophy of the affected epithelial cells. In addition, blockage of hair follicles or gland ducts can lead to the formation of papules and nodules (Ütük and Dumanlı, 2015). *D. folliculorum*, especially in clusters, forms plugs in the eyelash roots, causing follicular stretching and eyelash deformation. Microscopic abrasions caused by mites result in epithelial hyperplasia and reactive hyperkeratinization. Enfestasyonun patognomonik bulgusu olan kirpik diplerindeki silindirik kepeklenme ise akarların oluşturduğu yapısal bozukluklar ve metabolik atıkların burada birikmesiyle ilişkilidir. The cylindrical dandruff at the base of the eyelashes, which is the pathognomonic sign of infestation, is associated with structural defects caused by mites and accumulation of metabolic wastes there (Liu et al., 2010; Cheng et al., 2015; Zeytun and Karakurt, 2019).

Pathogenic mites in the follicles trigger a giant cell reaction in the host. This immune response causes epithelial hyperplasia, leading to mechanical blockage and obstruction of the follicle and sebaceous duct. The host organism develops an immune response against the residual products of the mite at both humoral and cellular levels. In addition, a foreign body-type granulomatous reaction occurs against the mite's chitin layer. In clinically asymptomatic carriers, structural skin deformities such as slowly developing raised and hard nodules, comedones, rotated hairs and enlarged hair follicles may be observed (Özcel et al., 2007a; Özcel et al., 2007b).

These mites are known to cause abscesses, skin reactions and pigmentation by creating an environment favourable to the growth of bacteria. They have also been reported to cause keratosis, hyperplasia, tension sensation and melanocyte increase in the eyelid follicles. In a study, it was reported that pityriasis folliculorum was observed in women who preferred cosmetic products instead of soap and water for facial cleansing. This mite causes inflammation in seborrhoeic and contaminated skin, leading to hair loss and secondary bacterial infections. It plays a role in the pathogenesis of acne rosacea, especially by facilitating the colonisation of microorganisms such as *Staphylococcus albus*. *D. folliculorum* concentrated at the base of the eyelashes may cause blepharitis and structural disruptions in the epithelial layer (Budak and Yolasığmaz, 1995; Forton et al., 2005; Chen and Plewig, 2015; Lacey et al., 2016; Forton and Maertelaer, 2019).

Diagnostic Methods

Various methods have been used in the past for the diagnosis of Demodex mites. Although samples can be taken and mites can be isolated by squeezing or comedone methods, it is usually difficult to obtain standardised results from these methods. In addition, these methods may cause pain in the patient and may lead to bruising. For the clinical diagnosis of demodicosis without specific symptoms, cellophane tape, skin scraping and punch biopsy methods as well as the non-invasive standard superficial skin biopsy (SSTB) method described by Marks and Dawber are used (Lacey et al., 2016). Mites circulating on the surface can be identified by skin scraping and cellophane tape. For definitive diagnosis, sebum lesions from hair follicles and seborrhoeic areas on the skin and purulent discharge squeezed out from lesioned areas such as acne and acne rosacea are sampled. These specimens are examined under a light microscope with a 100x objective with lacto-phenol or 10-30% KOH solution. In addition, Hoyer's solution or glycerin is added for detailed examination and diagnosis is made (Özcel et ark., 2007a; Özcel et al., 2007b). The cellophane tape method allows migrating and developing mites to be captured with tapes left on the skin for several hours. However, this technique can only provide superficial samples of pilosebaceous contents (Lacey et al., 2016). Therefore, the SYDB method, in which the corneum layer of the skin and follicle content are examined together, is frequently preferred. In the SYDB method, a drop of cyanoacrylate is placed on the slide and the adhesive surface of the slide is pressed against the suspected skin where the lesion is seen for a few minutes. Then, Hoyer's solution is dripped onto the slide and the coverslip is closed and examined in detail under a microscope with a 10x and 40x objective. The egg, nymph and adult stages of these mites are examined with a 100x objective. *Demodex* spp. infestation of the patient is considered positive if the mite density is five or more than five per cm^2. In diagnosis, the experience of the person performing the examination increases the rate of positive detection and facilitates species determination (Forton and Seys, 1993; Aycan et al., 2007; Lacey et al., 2016; Aytekin, 2018).

Eyelash epilation is a reproducible and convenient method for the detection of mites in the eyelid. In this method, eyelashes taken from the upper and lower eyelids of the patients are placed between the slide and coverslip in a drop of glycerine and evaluated under a light microscope with 40x and 100x magnification (Kaya et al., 2012; Lacey et al., 2016).

Mites can be difficult to visualise because they are translucent organisms. While fluorescein dye facilitates the identification of mites in epilated eyelashes, dyes such as rose bengal, merbromine, iodine, congo red, trypan blue, acridine orange, iodonitrotetrazolium, safranin and methylene blue can be successful in staining mites at different rates. In addition, dark field microscopy was used to visualise the living mites by autofluorescence. Methylene blue staining is recommended as a simple and convenient method for detecting and counting the presence of mites (Lacey et al., 2016; Kiuchi, 2018).

It is not yet clear whether the morphologically different *Demodex* species are really separate species or phenotypic variants. Molecular studies on *Demodex* species parasitising humans and animals are very limited. In a molecular analysis of *D. folliculorum* and *D. brevis*, it was determined that the transfer RNA (tRNA) gene sequences of both species were different. It was also found that these species have multiple gene arrangements between tRNA genes and carry many truncated tRNA genes. In the identification of *D. folliculorum*, it was revealed that one of the most appropriate molecular barcodes is the mitochondrial 16S rDNA gene and this gene can be used to distinguish the species (Palopoli et al., 2014; Ütük and Dumanlı, 2015; Daneshparvar et al., 2017).

Treatment

Drugs such as permethrin, lindane and crotamiton used in the treatment of scabies can also be effective in *D. folliculorum* infestation. One of the most reliable treatment methods is the application of 5% permethrin cream to the face as a mask twice a day for about 30 minutes. This treatment should be continued for 10 days. However, in severe infestations, reactions such as facial redness, itching, oedema may occur due to the death of the mites and the risk of re-infestation may occur. 2-3% sulphur ointments, Danish ointment and crotamiton were also found to be effective in treatment. Gamma benzene hexachloride (gamma-BHC) 0.5-1% applied after thorough washing of the skin lesions with hot water and soap is also known to reduce mite density and provide clinical improvement. Although it has no acaricidal effect, improvement has been observed in some cases where tetracycline and metronidazole were used orally together (Czepita et al., 2007; Zorbozan et al., 2016).

Oral ivermectin is recommended when the lesions are extensive. In *Demodex* spp. infestations, especially in cases of blepharitis, it has been statistically shown that 4% pilocarpine gel not only provides regression of symptoms but also significantly reduces the number of mites in pre- and post-treatment evaluations (Fulk et al., 1996; Czepita et al., 2007, Inceboz et al., 2009; Salem et al., 2013, Hirsch-Hoffmann et al., 2015).

Studies conducted in our country have shown that the number of *Demodex* mites decreases in the prognosis of diseases such as rosacea, perioral dermatitis, seborrheic dermatitis, blepharitis and micropapular pruritic dermatitis in patients who received specific permethrin treatment. In another study, the relationship between *D. folliculorum* and microbial flora was examined in patients presenting with acne complaints, and 5% permethrin was applied to eight patients with *D.*

folliculorum for two months. As a result of the checks performed every two months, a significant decrease in the number of mites was observed. Two of the seven patients whose lesions healed and no parasites were found re-applied with acne complaints after their second month follow-up, but *D. folliculorum* was not detected in these patients. These results show that permethrin treatment both reduces mite density and provides clinical improvement (Hirsch-Hoffmann et al., 2015; Zorbozan et al., 2016; Hecht et al., 2019; Navel et al., 2019).

Protection

Demodex mites can usually be transmitted from infested individuals to other individuals through direct contact. It should be noted that these mites cannot survive in dry environments. Skin cleanliness is of great importance to prevent infestation. Regular cleaning of the skin with mild soap and water is effective in reducing the density of mites (Chioveanu et al., 2024). For protection and control purposes, hand and face towels should not be shared. Using paper towels for a long time by patients during the treatment process reduces the risk of reinfection. In addition, considering the possibility of transmission from pets such as cats and dogs to humans, it is important to report skin lesions to a veterinarian for animals within the scope of protection (Özcel et al., 2007a; Özcel et al., 2007b; İnceboz et al., 2009).

Resources

Afşar, M., Hazrati-Tappeh, K., & Emre-Beyhan, Y. (2024). *Prevalence and severity of Demodex folliculorum and Demodex brevis infestation and their association with common skin diseases in Northwest Iran*. *Journal of Arthropod-Borne Diseases, 18*(2), 113.

Akdeniz, S., Bahceci, M., Tuzcu, A. K., Harman, M., Alp, S., & Bahceci, S. (2002). Is *Demodex folliculorum* larger in diabetic patients? *Journal of the European Academy of Dermatology and Venereology, 16*, 539–541.

Akilov, O. E., & Mumcuoglu, K. Y. (2003). Association between human demodicosis and HLA class I. *Clinical and Experimental Dermatology, 28*(1), 70–73.

Akilov, O. E., & Mumcuoglu, K. Y. (2004). Immune response in demodicosis. *Journal of the European Academy of Dermatology and Venereology, 18*(4), 440–444.

Aksakal, M. (2014). *Postadölesan akneli hastalarda Demodex spp.'nin araştırılması* (Uzmanlık tezi, Mustafa Kemal Üniversitesi Tayfur Ata Sökmen Tıp Fakültesi, Dermatoloji Anabilim Dalı, Hatay).

Aycan, Ö. M. (2008). *Demodex folliculorum ve D. brevis enfestasyonlarını etkileyen faktörler* (Doktora tezi, İnönü Üniversitesi Sağlık Bilimleri Enstitüsü, Malatya).

Aycan, Ö. M., Otlu, G. H., Karaman, U., Daldal, N., & Atambay, M. (2007). Frequency of demodicosis in various patient and age groups. *Türkiye Parazitoloji Dergisi, 31*(2), 115–118.

Aytekin, S. (2018). Yüzeysel deri biyopsi tekniği. *Güncel Dermatoloji Dergisi, 3*(1), 30–32.

Bonnar, E., Eustace, P., & Powell, F. C. (1993). The *Demodex* mite population in rosacea. *Journal of the American Academy of Dermatology, 28*, 443–448.

Budak, S., & Yolasığmaz, A. (1995). Demodicosis folliculorum. In M. A. Özcel (Ed.), *İmmun yetmezlikte önemi artan parazit hastalıkları* (pp. 169–171). Türkiye Parazitoloji Derneği Yayını No: 12.

Burns, D. A. (1992, Mayıs). Folikül akarları ve hastalıktaki rolleri. *Klinik ve Deneysel Dermatoloji, 17*(3), 152–155.

Chen, W., & Plewig, G. (2015). Are *Demodex* mites principal, conspirator, accomplice, witness or bystander in the cause of rosacea? *American Journal of Clinical Dermatology, 16*(2), 67–72.

Chen, W., Yang, S., & Li, W. (2020). *Demodex*: The worst enemy of human skin. *International Journal of Dermatology, 59*(12), 1415–1424.

Cheng, A. M., Hwang, J., Dermer, H., & Galor, A. (2021). Prevalence of ocular demodicosis and its association with dry eye symptoms and signs in an elderly population. *Cornea, 40*(8), 995–1001.

Cheng, A. M., Sheha, H., & Tseng, S. C. (2015). Recent advances on ocular *Demodex* infestation. *Current Opinion in Ophthalmology, 26*(4), 295–300.

Coston, T. O. (1967). *Demodex folliculorum* blepharitis. *Transactions of the American Ophthalmological Society, 65*, 361–392.

Czepita, D., Kuzna-Grygiel, W., Czepita, M., & Grobelny, A. (2007). *Demodex folliculorum* and *Demodex brevis* as a cause of chronic marginal blepharitis. *Annales Academiae Medicae Stetinensis, 53*(1), 63–67.

Çelik, E., & Kaya, Ö. M. A. (2020). Fototerapi alan hastalarda *Demodex folliculorum* ve *Demodex brevis* türlerinin araştırılması. *Bozok Tıp Dergisi, 10*(2), 164–170.

Daneshparvar, A., Mowlavi, G., Mirjalali, H., Hajjaran, H., Mobedi, I., et al. (2017). Molecular characterization and analysis of 16S ribosomal DNA in some isolates of *Demodex folliculorum*. *Iranian Journal of Parasitology, 12*(2), 224–229.

Desch, C., & Nutting, W. B. (1972). *Demodex folliculorum* (Simon) and *D. brevis* Akbulatova of man: Redescription and reevaluation. *Journal of Parasitology, 58*(1), 169–177.

Dolenc-Voljc, M., Pohar, M., & Lunder, T. (2005). Density of *Demodex folliculorum* in perioral dermatitis. *Acta Dermato-Venereologica, 85*(3), 211–215.

Erbagci, Z., & Ozgoztasi, O. (1998). The significance of *Demodex folliculorum* density in rosacea. *International Journal of Dermatology, 37*(6), 421–425.

Farina, M. C., Requena, L., Sarasa, J. L., Martin, L., Escalonilla, P., et al. (1998). Spinulosis of the face as a manifestation of demodicidosis. *British Journal of Dermatology, 138*(5), 901–903.

Forton, F., Germaux, M. A., Brasseur, T., De Liever, A., Laporte, M., Mathys, C., ... & Seys, B. (2005). Demodicosis and rosacea: Epidemiology and significance in daily dermatologic practice. *Journal of the American Academy of Dermatology, 52*(1), 74–87.

Forton, F., & Seys, B. (1993). Density of *Demodex folliculorum* in rosacea: A case control study using standardized skin surface biopsy. *British Journal of Dermatology, 128*, 650–659.

Forton, F. M. N., & De Maertelaer, V. (2019). Rosacea and demodicosis: Little-known diagnostic signs and symptoms. *Acta Dermato-Venereologica, 99*(1), 47–52.

Fromstein, S. R., Harthan, J. S., Patel, J., & Opitz, D. L. (2018). Demodex blepharitis: Clinical perspectives. *Clinical Optometry (Auckland, N.Z.), 10*, 57–63.

Fulk, G. W., Murphy, B., & Robins, M. D. (1996). Pilocarpine gel for the treatment of demodicosis: A case series. *Optometry and Vision Science, 73*(12), 742–745.

Hecht, I., Melzer-Golik, A., Sadi Szyper, N., & Kaiserman, I. (2019). Permethrin cream for the treatment of Demodex blepharitis. *Cornea*. https://doi.org/10.1097/ICO.0000000000002013

Hirsch-Hoffmann, S., Kaufmann, C., Banninger, P. B., & Thiel, M. A. (2015). Treatment options for Demodex blepharitis: Patient choice and efficacy. *Klinische Monatsblätter für Augenheilkunde, 232*(4), 384–387.

İnceboz, T., Yaman, A., Över, L., Öztürk, A. T., & Akisu, C. (2009). Diagnosis and treatment of demodectic blepharitis. *Türkiye Parazitoloji Dergisi, 33*(1), 32–36.

Kaya, O. A., Akkucuk, S., Ilhan, G., Guneri, C. O., & Mumcuoglu, K. (2019). The importance of *Demodex* mites (Acari: Demodicidae) in patients with sickle cell anemia. *Journal of Medical Entomology, 56*(3), 599–602.

Kaya, O. A., Atambay, M., & Daldal, N. (2012). Sağlıklı kişilerin kirpiklerinde *Demodex folliculorum* ve *Demodex brevis* görülme sıklığı. *Kafkas Üniversitesi Veteriner Fakültesi Dergisi, 18*, 57–60.

Kemal, M., Sumer, Z., Toker, M. I., Erdogan, H., Topalkara, A., & Ark. (2005). The prevalence of *Demodex folliculorum* in blepharitis patients and the normal population. *Ophthalmic Epidemiology, 12*(4), 287–290.

Kim, J. H., Chun, Y. S., & Kim, J. C. (2011). Clinical and immunological responses in ocular demodecosis. *Journal of Korean Medical Science, 26*(9), 1231–1237.

Kiuchi, K. (2018). Better detection of *Demodex* mites by Löffler's alkaline methylene blue staining in patients with blepharitis. *Clinical Ophthalmology, 12*, 727–731.

Koo, H., Kim, T. H., Kim, K. W., Wee, S. W., Chun, Y. S., & Ark. (2012). Ocular surface discomfort and *Demodex*: Effect of tea tree oil eyelid scrub in *Demodex* blepharitis. *Journal of Korean Medical Science, 27*(12), 1574–1579.

Kulaç, M., Ciftci, I. H., Karaca, S., & Çetinkaya, Z. (2008). Clinical importance of *Demodex folliculorum* in patients receiving phototherapy. *International Journal of Dermatology, 47*(1), 72–77.

Lacey, N., Delaney, S., & Powell, F. C. (2009). The *Demodex* mite population in rosacea. *Journal of the American Academy of Dermatology, 60*(3), 574–576.

Lacey, N., Russell-Hallinan, A., & Powell, F. C. (2016). Study of *Demodex* mites: Challenges and solutions. *Journal of the European Academy of Dermatology and Venereology, 30*(5), 764–775.

Liu, J., Sheha, H., & Tseng, S. C. (2010). Pathogenic role of *Demodex* mites in blepharitis. *Current Opinion in Allergy and Clinical Immunology, 10*(5), 505–510.

Litwin, D., Chen, W., Dzika, E., & Korycińska, J. (2017). Human permanent ectoparasites: Recent advances on biology and clinical significance of *Demodex* mites: Narrative review article. *Iranian Journal of Parasitology, 12*(1), 12–21.

Lindsley, K., Matsumura, S., Hatef, E., & Akpek, E. K. (2012). Interventions for chronic blepharitis. *Cochrane Database of Systematic Reviews*. https://doi.org/10.1002/14651858.CD005556.pub2

Lopez-Ponce, D., Zuazo, F., Cartes, C., Salinas-Toro, D., Perez-Valenzuela, C., & Ark. (2017). High prevalence of *Demodex* spp. infestation among patients with posterior blepharitis: Correlation with age and cylindrical dandruff. *Archivos de la Sociedad Española de Oftalmología, 92*(9), 412–418.

Markell, E. K., Voge, M., & John, D. T. (1992). *Medical parasitology* (7th ed.). Philadelphia: WB Saunders Company.

Meinking, T., Taplin, D., Vicaria, M., Schachner, L. A., & Hansen, R. C. (Eds.). (2011). *Pediatrik dermatoloji* (s. 1575). Elsevier. ISBN: 9780723435402

Monsel, G., Delaunay, P., & Chosidow, O. (2016). Bölüm 34.1: Eklembacaklılar - Folikül akarları (*Demodicidae*). In C. Griffiths, J. Barker, T. Bleiker, R. Chalmers, & D. Creamer (Eds.), *Rook's textbook of dermatology* (9th ed., Vol. 1, pp. 34.52–34.54). John Wiley & Sons.

Mumcuoğlu, K. (2004). *Tıbbi önemi olan eklembacaklılar kursu ders notları* (s. 115–117). Cumhuriyet Üniversitesi Tıp Fakültesi Parazitoloji – Hebrew Üniversitesi Haddassah Tıp Fakültesi Parazitoloji ABD.

Murube, J. (2015). *Demodex hominis*. *The Ocular Surface, 13*(3), 181–186.

Navel, V., Mulliez, A., Benoist d'Azy, C., Baker, J. S., Malecaze, J., & Ark. (2019). Efficacy of treatments for *Demodex* blepharitis: A systematic review and meta-analysis. *The Ocular Surface*. https://doi.org/10.1016/j.jtos.2019.06.004

Nutting, W. B. (1965). Hair follicle mites (Acari: Demodicidae) of man. *International Journal of Dermatology, 15*(2), 79–98.

Nutting, W. B., Firda, K. E., & Desch, C. E. Jr. (1989). İnsan kıl folikülü akarlarının (*Demodicidae*) topolojisi ve histopatolojisi. In G. P. Channabasavanna & C. A. Viraktamath (Eds.), *Akarolojide ilerleme* (Vol. 1, pp. 113–121). Leiden, Hollanda: E. J. Brill.

Olt, S., Yalçın, G. G., Uysal, O. S., Karakece, E., & Çiftci, I. H. (2013). *Demodex spp. infestation in a breast-cancer patient: A case report*. *Nigerian Medical Journal, 54*(5), 349–350.

Özcel, M. A. (1995). *İmmun yetmezlikte önemi artan parazit hastalıkları*. Ege Üniversitesi Basımevi, Bornova, İzmir.

Özcel, M. A., İnci, A., Turgay, N., & Köroğlu, E. (2007b). *Tıbbi ve veteriner immunoparazitoloji*. Türkiye Parazitol Derg Yayınları, İzmir.

Özcel, M. A., Özbel, Y., & Ak, M. (2007a). *Özcel'in tıbbi parazit hastalıkları*. Türkiye Parazitol Derg Yayınları, İzmir.

Özcel, M. A., Özbel, Y., & Ak, M. (2007b). *Özcel'in tıbbi parazit hastalıkları*. Türkiye Parazitol Derg Yayınları, İzmir.

Oytun, Ş. (1969). *Tıbbi entomoloji* (3. baskı). Ankara Üniversitesi Tıp Fakültesi Yayın No: 218, Ankara.

Palopoli, M. F., Minot, S., Pei, D., Satterly, A., & Endrizzi, J. (2014). Complete mitochondrial genomes of the human follicle mites *Demodex brevis* and *D. folliculorum*: Novel gene arrangement, truncated tRNA genes, and ancient divergence between species. *BMC Genomics, 15*(1124), 1–15. https://doi.org/10.1186/s12864-014-1124-4

Ruhl, S., Moser, B., & Niebler, A. (2016). Investigation of *Demodex* mite population in human hosts and its relationship with clinical manifestations of blepharitis. *Dermatology, 44*(5), 535–541.

Salem, D. A., El-Shazly, A., Nabih, N., El-Bayoumy, Y., & Saleh, S. (2013). Evaluation of the efficacy of oral ivermectin in comparison with ivermectin-metronidazole combined therapy in the treatment of ocular and skin lesions of *Demodex folliculorum*. *International Journal of Infectious Diseases, 17*(5), 343–347. https://doi.org/10.1016/j.ijid.2013.01.009

Saygı, G., Marufi, M., & Köylüoğlu, Z. (1984). Biri selofanlı bant preparatı ile saptanan üç *Demodex folliculorum* olgusu. *Türkiye Parazitol Derg, 7*, 137–144.

Sahn, E. E., & Sheridan, D. M. (1992). *Demodicidosis* in a child with leukemia. *Journal of the American Academy of Dermatology, 27*, 799–801.

Sedzikowska, A., Oseka, M., & Grytner-Ziecina, B. (2016). Ocular symptoms reported by patients infested with *Demodex* mites. *Acta Parasitol, 61*(4), 808–814.

Sedzikowska, A., Oseka, M., & Skopinski, P. (2018). The impact of age, sex, blepharitis, rosacea, and rheumatoid arthritis on *Demodex* mite infection. *Archives of Medical Science, 14*(2), 353–356.

Simon, G. (1842). *"Ueber eine in den kranken und normalen Haarsäcken des Menschen lebende Milbe"* [İnsanların hastalıklı ve normal saç foliküllerinde yaşayan bir akar üzerine]. *Archiv für Anatomie, Physiologie und Wissenschaftliche Medicin*, Berlin: Verlag von Veit & Comp., 221.

Spickett, G. (1961). Akarın davranış ve biyolojisi üzerine çalışmalar. *Journal of the American Academy of Dermatology, 9*(3), 133–145.

Tarkowski, W., Owczynska, M., Blaszczyk-Tyszka, A., & Mlocicki, D. (2015). Demodex mites as potential etiological factor in chalazion - A study in Poland. *Acta Parasitol, 60*(4), 777–783. https://doi.org/10.1515/ap-2015-0105

Tanriverdi, C., Demirci, G., Balci, O., Odabasi, M., & Ozsutcu, M. (2018). Investigation of *Demodex* parasite existence in treatment-resistant chronic blepharitis cases. *Turkiye Parazitol Derg, 42*(2), 130–133. https://doi.org/10.5152/tpd.2018.2785

Ütük, A. E., & Dumanlı, N. (2015). Prostigmata. In *Arthropodoloji* (1st ed., pp. 95–103). Medisan Yayınevi, Ankara.

Unat, E. K., Yücel, A., Altaş, K., & Samastı, M. (1995). *Unat'ın tıp parazitolojisi*. İstanbul Üniv. Cerrahpaşa Tıp Fak. Vakfı Yayınları No: 15, İstanbul.

Yildiz, R., Afşar, M., & Elasan, S. (2024). Global trends in studies on the presence of *Demodex* spp. in patients diagnosed with blepharitis. *Journal of Arthropod-Borne Diseases, 18*(2), 84. https://doi.org/10.18502/jabd.v18i2.2116

Yazar, S., Kuk, S., Miman, Ö., & Saygı, G. (2016). *Saygı'nın Temel Tıbbi Parazitoloji'si*. Erciyes Üniversitesi Yayınları, Kayseri, 261–262.

Zeytun, E., Karakurt, Y. (2019). Prevalence and load of *Demodex folliculorum* and *Demodex brevis* (Acari: Demodicidae) in patients with chronic blepharitis in the province of Erzincan, Turkey. *Journal of Medical Entomology, 56*(1), 2–9. https://doi.org/10.1093/jme/tjy177

Zeytun, E., Tilki, E., Dogan, S., & Mumcuoglu, K. Y. (2017). The effect of skin moisture, pH, and temperature on the density of *Demodex folliculorum* and *Demodex brevis* (Acari: Demodicidae) in students and staff of Erzincan University, Turkey. *International Journal of Dermatology, 56*(7), 762–766. https://doi.org/10.1111/ijd.13427

Zorbozan, O., Okut, E. B., Unver, A., & Turgay, N. (2016). *Demodex folliculorum* ile ilişkili blefarit olgusu. *Türk Mikrobiyol Cemiyeti Dergisi, 46*(1), 47–50.

Chapter 4

The Impact of Assisted Reproductive Techniques on Maternal Health Outcomes

Dilay GÖK KORUCU[1]

Introduction

The first successful human pregnancy achieved through in vitro fertilization was documented in 1976, followed by the landmark birth of the first IVF-conceived infant in 1978 (Steptoe & Edwards, 1976, 1978). Assisted reproductive technology (ART), including in vitro fertilization (IVF) and intracytoplasmic sperm injection (ICSI), has achieved significant advancements in success rates. Consequently, its clinical applications have broadened, yet the rising prevalence of ART-conceived pregnancies has also heightened concerns regarding associated maternal and perinatal outcomes. While the majority of individuals undergoing IVF and their offspring exhibit no health complications, ART has been consistently linked to elevated risks of adverse maternal and perinatal outcomes.

Severe matrernal morbidity and mortality: Advanced maternal age, a key risk factor for pregnancy-related mortality, simultaneously represents a predominant indication for ART treatment. Clinical evidence suggests that ART-mediated pregnancies, especially those achieved through IVF, demonstrate significantly higher rates of severe maternal morbidity relative to natural conceptions. This disparity may be partially explained by the demographic characteristics of ART populations, who frequently present with advanced maternal age and greater baseline comorbidity burdens - known contributors to adverse maternal outcomes (Pelikh, Smith, Myrskylä, Debbink, & Goisis, 2025; Wu et al., 2022). After rigorous adjustment for maternal age, plurality, and other potential confounders using propensity score matching, IVF-conceived pregnancies showed a 1.38-fold increased risk (95% CI [X-X]) of severe maternal

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morbidity compared to spontaneous conceptions, with absolute rates of 30.8 and 22.2 per 1000 births, respectively (Dayan et al., 2019). Severe maternal morbidity was predominantly characterized by postpartum hemorrhage, ICU admission, and sepsis. Unlike IVF, non-invasive fertility interventions (e.g., ovulation induction, intrauterine insemination) demonstrated no increased risk of adverse maternal outcomes.

Stroke: Evidence from observational studies demonstrates that ART-conceived pregnancies carry an elevated risk of postpartum stroke relative to spontaneous conceptions (Dicpinigaitis et al., 2024; Sachdev, Yamada, Lee, Sauer, & Ananth, 2023). While the pathophysiology remains uncertain, potential contributors include: baseline infertility-related risk profiles (advanced age, comorbidities, higher BMI); and ART procedural factors—particularly FET, which elevates preeclampsia risk and thereby stroke susceptibility (Bortoletto, Prabhu, & Baker, 2022; Brohan et al., 2023).

Venous thrombosis: IVF-conceived pregnancies demonstrate elevated risks of both pulmonary and venous thromboembolism during gestation, with particular vulnerability observed in the first trimester. Notably, this increased thromboembolic risk persists even in the absence of clinically apparent ovarian hyperstimulation syndrome (OHSS). In one study, for example, the risk of pulmonary embolism in the first trimester in individuals after IVF versus in individuals with natural pregnancies was 3 in 10,000 versus 0.4 in 10,000 (HR 6.97, 95% CI 2.21-21.96)(Henriksson et al., 2013; Rova, Passmark, & Lindqvist, 2012). While these data identify a potential safety signal, clinical application awaits confirmation from large prospective cohorts. The risk-benefit ratio of thromboprophylaxis in IVF pregnancies remains uncertain. Enhanced patient awareness of thromboembolic symptoms represents a prudent, low-risk intervention pending further evidence.

Risk of cancer: Current evidence suggests that fertility medications and IVF treatment are not associated with increased overall cancer risk or elevated incidence of breast, cervical, or ovarian malignancies. However, the observational design (frequently retrospective) of available studies limits the ability to draw definitive conclusions regarding long-term oncologic safety.

Ovarian cancer: While ovarian cancer rates are elevated in ART-treated women versus the general population, this excess risk is principally explained by their infertile status rather than therapeutic interventions. A 2020 study comparing 30,000 ART-exposed and 10,000 untreated subfertile women found that ovarian cancer incidence was elevated versus the general population (SIR 1.43, 95% CI 1.18–1.71) but not versus untreated subfertile controls (aHR 1.02, 95% CI 0.70–1.50), supporting infertility—rather than ovarian stimulation—as the primary risk factor (Spaan et al., 2021). With a median follow-up of 24 years, the study demonstrated an inverse relationship between ovarian cancer risk and both parity (risk decreasing with higher live birth numbers) and cumulative number of successful ART cycles. A 2018 population-based cohort study (N=255,786 IVF-treated individuals) identified a modest but statistically significant increase in ovarian cancer incidence (standardized incidence ratio [SIR] 1.39, 95% CI 1.25-1.53). However, the absolute risk difference remained clinically small (5 additional cases per 100,000 person-years) (Williams et al., 2018).

Borderline ovarian tumors: Observational data indicate a potential association between ovarian-stimulating medications (clomiphene, gonadotropins) and increased risk of borderline ovarian tumors among infertility patients, including those undergoing IVF. However, residual confounding and the need for further research preclude definitive causal conclusions at this time (Rizzuto, Behrens, & Smith, 2019). In the 2020 cohort study, ART-treated individuals showed elevated risk of borderline ovarian tumors compared to both the general population and untreated subfertile controls. However, the absence of a dose-response relationship (with increasing ART cycles) calls into question the biological plausibility of this association (Spaan et al., 2021).

Breast cancer: No elevated breast cancer risk has been demonstrated with ART, even in high-risk populations (BRCA1/2+). Therefore, ART remains an appropriate option for patients with hereditary breast cancer risk (Beebeejaun et al., 2021; Derks-Smeets et al., 2018). Systematic analysis of three decades of data (1990-2020) revealed no significant increase in breast cancer risk among women exposed to fertility medications versus unexposed controls, including both population-based and infertility comparison groups (Beebeejaun et al., 2021).

Long term risk of cardiovascular disease : While IVF pregnancies may increase thromboembolic risk, longitudinal data show no persistent cardiovascular disease risk post-treatment. A pooled analysis of 41,000 patients revealed comparable cardiac event rates in fertility-treated and untreated groups (Dayan et al., 2017).

In conclusion, while ART pregnancies show similar overall morbidity to spontaneous conceptions, clinicians should be aware of slightly higher risks for venous thromboembolism and gestational hypertension. Importantly, no excess risk of cardiovascular disease or cancer has been documented beyond baseline infertility-associated risks.

References

Beebeejaun, Y., Athithan, A., Copeland, T. P., Kamath, M. S., Sarris, I., & Sunkara, S. K. (2021). Risk of breast cancer in women treated with ovarian stimulation drugs for infertility: a systematic review and meta-analysis. *Fertil Steril, 116*(1), 198-207. doi:10.1016/j.fertnstert.2021.01.044

Bortoletto, P., Prabhu, M., & Baker, V. L. (2022). Association between programmed frozen embryo transfer and hypertensive disorders of pregnancy. *Fertil Steril, 118*(5), 839-848. doi:10.1016/j.fertnstert.2022.07.025

Brohan, M. P., Daly, F. P., Kelly, L., McCarthy, F. P., Khashan, A. S., Kublickiene, K., & Barrett, P. M. (2023). Hypertensive disorders of pregnancy and long-term risk of maternal stroke-a systematic review and meta-analysis. *Am J Obstet Gynecol, 229*(3), 248-268. doi:10.1016/j.ajog.2023.03.034

Dayan, N., Filion, K. B., Okano, M., Kilmartin, C., Reinblatt, S., Landry, T., . . . Udell, J. A. (2017). Cardiovascular Risk Following Fertility Therapy: Systematic Review and Meta-Analysis. *J Am Coll Cardiol, 70*(10), 1203-1213. doi:10.1016/j.jacc.2017.07.753

Dayan, N., Joseph, K. S., Fell, D. B., Laskin, C. A., Basso, O., Park, A. L., . . . Ray, J. G. (2019). Infertility treatment and risk of severe maternal morbidity: a propensity score-matched cohort study. *Cmaj, 191*(5), E118-e127. doi:10.1503/cmaj.181124

Derks-Smeets, I. A. P., Schrijver, L. H., de Die-Smulders, C. E. M., Tjan-Heijnen, V. C. G., van Golde, R. J. T., Smits, L. J., . . . Gómez García, E. B. (2018). Ovarian stimulation for IVF and risk of primary breast cancer in BRCA1/2 mutation carriers. *Br J Cancer, 119*(3), 357-363. doi:10.1038/s41416-018-0139-1

Dicpinigaitis, A. J., Seitz, A., Berkin, J., Al-Mufti, F., Kamel, H., Navi, B. B., . . . Liberman, A. L. (2024). Association of Assisted Reproductive Technology and Stroke During Hospitalization for Delivery in the United States. *Stroke, 55*(4), 999-1005. doi:10.1161/strokeaha.124.046419

Henriksson, P., Westerlund, E., Wallén, H., Brandt, L., Hovatta, O., & Ekbom, A. (2013). Incidence of pulmonary and venous thromboembolism in pregnancies after in vitro fertilisation: cross sectional study. *Bmj, 346*, e8632. doi:10.1136/bmj.e8632

Pelikh, A., Smith, K. R., Myrskylä, M., Debbink, M. P., & Goisis, A. (2025). Maternal Morbidity and Medically Assisted Reproduction Treatment Types. *Obstet Gynecol, 145*(2), 220-230. doi:10.1097/aog.0000000000005808

Rizzuto, I., Behrens, R. F., & Smith, L. A. (2019). Risk of ovarian cancer in women treated with ovarian stimulating drugs for infertility. *Cochrane Database Syst Rev, 6*(6), Cd008215. doi:10.1002/14651858.CD008215.pub3

Rova, K., Passmark, H., & Lindqvist, P. G. (2012). Venous thromboembolism in relation to in vitro fertilization: an approach to determining the incidence and increase in risk in successful cycles. *Fertil Steril, 97*(1), 95-100. doi:10.1016/j.fertnstert.2011.10.038

Sachdev, D., Yamada, R., Lee, R., Sauer, M. V., & Ananth, C. V. (2023). Risk of Stroke Hospitalization After Infertility Treatment. *JAMA Netw Open, 6*(8), e2331470. doi:10.1001/jamanetworkopen.2023.31470

Spaan, M., van den Belt-Dusebout, A. W., Lambalk, C. B., van Boven, H. H., Schats, R., Kortman, M., . . . van Leeuwen, F. E. (2021). Long-Term Risk of Ovarian Cancer and Borderline Tumors After Assisted Reproductive Technology. *J Natl Cancer Inst, 113*(6), 699-709. doi:10.1093/jnci/djaa163

Steptoe, P. C., & Edwards, R. G. (1976). Reimplantation of a human embryo with subsequent tubal pregnancy. *Lancet, 1*(7965), 880-882. doi:10.1016/s0140-6736(76)92096-1

Steptoe, P. C., & Edwards, R. G. (1978). Birth after the reimplantation of a human embryo. *Lancet, 2*(8085), 366. doi:10.1016/s0140-6736(78)92957-4

Williams, C. L., Jones, M. E., Swerdlow, A. J., Botting, B. J., Davies, M. C., Jacobs, I., . . . Sutcliffe, A. G. (2018). Risks of ovarian, breast, and corpus uteri cancer in women treated with assisted reproductive technology in Great Britain, 1991-2010: data linkage study including 2.2 million person years of observation. *Bmj, 362*, k2644. doi:10.1136/bmj.k2644

Wu, P., Sharma, G. V., Mehta, L. S., Chew-Graham, C. A., Lundberg, G. P., Nerenberg, K. A., . . . Mamas, M. A. (2022). In-Hospital Complications in Pregnancies Conceived by Assisted Reproductive Technology. *J Am Heart Assoc, 11*(5), e022658. doi:10.1161/jaha.121.022658

Chapter 5

Single Fiber Electromyography, Concentric Needle Electromyography and Myastenia Gravis

Hasan YAŞAR[1]

General information

The neuromuscular junction (NMJ) is the specialized region where motor nerves send signals to cause skeletal muscle to contract. Disturbances in neuromuscular transmission cause clinical weakness that may fluctuate and lead to fatigue. When the defect in neuromuscular transmission is mild, the reduction may not be observed with standard methods of repetitive stimulation. Signaling at the NMJ is affected in many diseases, including congenital myasthenic syndromes, myasthenia gravis (MG), Lambert-Eaton myasthenic syndrome, Isaacs syndrome, Schwartz-Jampel syndrome, Fukuyama type congenital muscular dystrophy, amyotrophic lateral sclerosis, and sarcopenia (1). In MG, a neuromuscular disease, autoantibodies are directed against acetylcholine receptors or, in rarer variants, other postsynaptic NMJ proteins (2). MG is the most common acquired autoimmune disease of the NMJ, affecting 400–600 patients per million individuals (3). Diplopia and ptosis are the most common complaints of patients with ocular MG. The differential diagnosis of MG is of great importance in patients with these symptoms (4). Single-fiber EMG (SFEMG) examination is highly sensitive in the differential diagnosis of MG (5).

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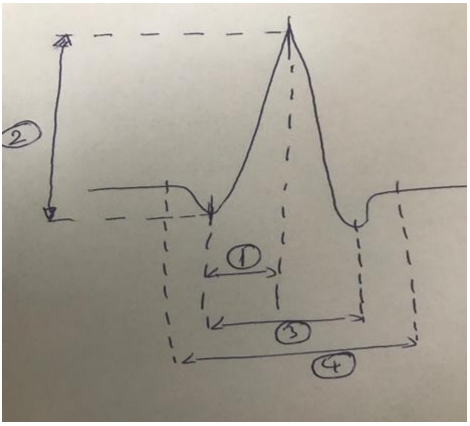
Single-fiber electromyography examination

Structure of trigger potential in SFEMG examination



1. Rise time
2. Amplitude height
3. Duration of potential
4. Total duration

Amplifier settings of 0.2–0.5 mV/DIVISION are ideal for recording signals. An oscilloscope sweep speed of 0.5 ms/DIVISION makes it easy to evaluate the temporal variability of signals (6). Neuromuscular jitter represents the time variability of the interval between firing of action potentials in single muscle fibers in successive discharges (7). Jitter is elicited by a slight voluntary muscle contraction (VAT) of 50 to 100 pairs of single fiber action potentials (SFAP) from the same motor unit. The mean consecutive difference (MCD) between these pairs represents the jitter. The needle is rotated, advanced, or withdrawn until the clear sound of a SFAP is heard. The examiner triggers one SFAP and moves the needle slightly, trying to find another SFAP from one motor unit that is time-locked to the first. Jitter is the interpotential interval variability caused by the temporal variation of the two endplates. Jitter is abnormal when the average jitter of 20 pairs is above the reference limit. Jitter is also abnormal if more than 10% of 20 individual pairs exceed the reference limit (8). Skin temperature should be kept above 32°C (9). The extensor digitorum longus (EDC) is abnormal in most patients with generalized MG and is the first muscle to be examined. If there is significant weakness in the muscle and jitter is normal, the weakness is not due to MG. If symptoms or weakness are limited to the eye muscles only, the orbicularis oculi or frontalis muscle may be examined first. Needle EMG is also a useful indicator in the diagnosis of botulism, especially infantile botulism. Fibrillation potentials and positive sharp waves are frequently seen on EMG due to chemical denervation. Motor unit action potentials (MUAP) may be normal or have a myopathy-like pattern like other NMJ disorders. Compound muscle action potentials is usually absent, making motor nerve conduction studies assessment difficult (10). In SFEMG, the waves should be smooth, biphasic, and the shape of consecutive action potentials should be the same. Needle electrode is inserted through the muscle 1 to 1.5 cm from the endplate region and SFAPs in the vicinity of the NMJ are recorded. SFAPs with rise times of less than 300 microseconds and amplitudes greater than 200 microVolts are considered for analysis (6). SFEMG in voluntary muscle is a powerful electrophysiological method to assess impaired neuromuscular transmission in MG and is much more sensitive than repetitive nerve stimulation (RNS) testing in detecting abnormalities in MG. This is because SFEMG can detect subclinical impairment of neuromuscular transmission (increased jitter) that does not produce neuromuscular block. In contrast, RNS is positive only when there is significant neuromuscular blockade (11). When the jitter value is below 80µs, impulse blocking does not occur, which brings some technical disadvantages. The maximum jitter value should be 150 µs. Jitter values lower than 5 µs should not be considered as they represent recordings from the same muscle fiber (8). The mean sorted difference (MSD) should be chosen when it is smaller than the MCD. If the MCD/MSD ratio is greater than 1.25, the correct measure of jitter is MSD. Below 1,25 the preferred method for expressing jitter is MCD. The specific level of abnormality in MCD varies with muscle and age, but no normal muscle should have jitter greater than 55 microseconds or blockage in more than one fiber (9). If the MCD of jitter in any fiber pair is greater than 55 µs or more than 10% of the fiber pairs are blocked, the result is considered abnormal (12).

Concentric needle electromyography examination

Due to concerns about virus or prion infection, reusable single fiber electrodes are no longer used. Instead, single-use concentric needle electrodes (CNEs) are preferred. However, as the recording area expands, selectivity decreases. To get the most out of CNE, filter settings should be set between 1 KHz and 10 KHz. Unlike SFEMG, CNE jitter values do not increase with age (8). Although the signals recorded with CNE appear to be a single potential, they are more likely a combination of several potentials. Therefore, fiber density measurements cannot be made using CNE. Acceptable potentials for jitter analysis recorded with concentric needles should have a positive and a negative peak, and the initial rising phase should show a stable shape with no shoulders or notches. The signal peaks should have a constant shape and should not have large amplitude changes. Because of the effect of filtering on amplitude, potentials with signal amplitudes >50 microvolts are acceptable in concentric jitter recordings. The rise time should be less than 300µs. Concentric needle examination loses significant selectivity and some sensitivity in jitter assessment (5, 13). During the voluntary jitter analysis, at least 60 records should be taken. It should not fall below 60. For jitter studies, the CNE with the smallest recording area is recommended (14). There must be a gap of at least 150 µs between the triggering signal and the other signal.

After each jitter measurement, the recording electrode should be shifted sufficiently. The CNE sometimes records more than one potential. The one with the most abnormal jitter should not be used as the triggered spike. Increased Jitter in CNE Suggests Junctional Pathology, Not Reinnervation. If there is more than 100 microseconds of extension but no block, it should check the quality of the records (13).

Results

The patient should first be asked whether he or she has used botulinum neurotoxin (BoNT). In muscles directly injected with BoNT, jitter is abnormal in all cases, with a high rate of blockade usually observed in the first weeks. These muscles can never be used to measure jitter. The muscle can be used for jitter measurement 11 months after the last injection, and the reference limit should be increased by 33%.

Although not mandatory, the patient should discontinue acetylcholinesterase(AChE) inhibitors 12 hours before the test. Jitter is abnormal in actively innervated and chronically innervated muscles. Jitter cannot be used as an electrodiagnostic test in muscles that show MUAPs suggestive of myopathy. Drugs that may interfere with neuromuscular transmission should be questioned, such as calcium channel blockers (verapamil and amlodipine) or AChE inhibitors (8). Neuromuscular transmission disorders are improved at lower temperatures. They are impaired at higher temperatures. Neuromuscular junction physiology is also sensitive to heat (15). Myasthenia gravis is diagnosed by history, clinical findings, electrophysiological examination, response to anticholinesterase drugs, and abnormal acetylcholine receptor antibody titer. Increased jitter is not sufficient for the diagnosis of myasthenia gravis (16). The specificity and sensitivity of SFEMG in ocular MG is not the gold standard and its contribution to clinical diagnosis is limited (17). If SFEMG is normal in a weak muscle, the patient does not have MG. However, this is only true for SFEMG. It is not true for CNE. If the jitter measurement with CNE is normal, this reduces the possibility of myasthenia gravis but does not exclude it. It may give false-negative results especially in cases of ocular MG or mild generalized MG (18).

References

1. Ohkawara B, Ito M, Ohno K. Secreted signaling molecules at the neuromuscular junction in physiology and pathology. International Journal of Molecular Sciences. 2021;22(5):2455.
2. Plomp JJ, Huijbers MG, Verschuuren JJ. Neuromuscular synapse electrophysiology in myasthenia gravis animal models. Annals of the New York Academy of Sciences. 2018;1412(1):146-53.
3. Li L, Xiong W-C, Mei L. Neuromuscular junction formation, aging, and disorders. Annual review of physiology. 2018;80(1):159-88.
4. Oey PL, Wieneke GH, Hoogenraad TU, Van Huffelen AC. Ocular myastyenia gravis: The diagnostic yield of repetitive nerve stimulation and stimulated single fiber EMG of orbicularis oculi muscle and infrared reflection oculography. Muscle & Nerve: Official Journal of the American Association of Electrodiagnostic Medicine. 1993;16(2):142-9.
5. Tanhehco JL. Single-fiber electromyography. Physical Medicine and Rehabilitation Clinics. 2003;14(2):207-29.
6. Sirin NG, Mehdikhanova L, Sirinocak PB, Arkali NB, Baslo MB, Orhan EK. Near-tendon single-fiber electromyography in patients with myopathy. Neurophysiologie Clinique. 2022;52(2):174-7.
7. Lundh H, Schiller HH, Elmqvist D. Correlation between single fibre EMG jitter and endplate potentials studied in mild experimental botulinum poisoning. Acta Neurologica Scandinavica. 1977;56(2):141-52.
8. Kouyoumdjian JA, de Paula Estephan E. Electrophysiological evaluation of the neuromuscular junction: a brief review. Arquivos de Neuro-psiquiatria. 2023;81(12):1040-52.
9. Lange DJ. Electrophysiologic testing of neuromuscular transmission. Neurology. 1997;48(suppl_5):18S-22S.
10. Cherian A, Baheti NN, Iype T. Electrophysiological study in neuromuscular junction disorders. Annals of Indian Academy of Neurology. 2013;16(1):34-41.
11. Sonoo M, Uesugi H, Mochizuki A, Hatanaka Y, Shimizu T. Single fiber EMG and repetitive nerve stimulation of the same extensor digitorum communis muscle in myasthenia gravis. Clinical neurophysiology. 2001;112(2):300-3.
12. Chiou-Tan FY, Tim RW, Gilchrist JM, Weber CF, Wilson JR, Benstead TJ, et al. Practice parameter for repetitive nerve stimulation and single fiber EMG evaluation of adults with suspected myasthenia gravis or Lambert-Eaton myasthenic syndrome: summary statement. Muscle and Nerve. 2001;24(9):1236-8.

13. Sanders DB, Arimura K, Cui L, Ertaş M, Farrugia ME, Gilchrist J, et al. Guidelines for single fiber EMG. Clinical Neurophysiology. 2019;130(8):1417-39.
14. Juel VC. Single fiber electromyography. Handbook of clinical neurology. 2019;160:303-10.
15. Rutkove SB, Kothari MJ, Shefner JM. Nerve, muscle, and neuromuscular junction electrophysiology at high temperature. Muscle & Nerve: Official Journal of the American Association of Electrodiagnostic Medicine. 1997;20(4):431-6.
16. Valls-Canals J, Povedano M, Montero J, Pradas J. Stimulated single-fiber EMG of the frontalis and orbicularis oculi muscles in ocular myasthenia gravis. Muscle & Nerve: Official Journal of the American Association of Electrodiagnostic Medicine. 2003;28(4):501-3.
17. Rouseev R, Ashby P, Basinski A, Sharpe J. Single fiber EMG in the frontalis muscle in ocular myasthenia: specificity and sensitivity. Muscle & Nerve: Official Journal of the American Association of Electrodiagnostic Medicine. 1992;15(3):399-403.
18. CN Machado F, A. Kouyoumdjian J, E. Marchiori P. Diagnostic accuracy of concentric needle jitter in myasthenia: prospective study. Muscle & nerve. 2017;55(2):190-4.

Chapter 6

The Pivotal Role of Ticks in Zoonotic and Veterinary Disease Transmission

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Significance of the Disease

Tick infestations play a central role in the epidemiology of Crimean-Congo Hemorrhagic Fever (CCHF), an acute viral zoonosis of major public health significance. The principal vectors of the disease are *Hyalomma* spp. ticks, whose biological and ecological characteristics directly influence the transmission dynamics and geographic spread of the virus. Endemic in regions such as Eastern Europe, the Middle East, and parts of Asia including Türkiye CCHF is primarily transmitted through the bite of infected ticks or via direct contact with the blood and bodily fluids of viremic animals and humans. The persistence and expansion of tick populations, driven by environmental factors and livestock movement, are critical determinants of disease prevalence. As such, effective surveillance and control of tick infestations are essential components of CCHF prevention strategies, particularly in rural and agricultural settings where human exposure to ticks is heightened (Belobo et al., 2021; Ozdarendeli, 2023).

Ticks are among the most important ectoparasites affecting both human and animal health, primarily due to their hematophagous feeding behavior and exceptional capacity as vectors of a wide array of pathogens. Present on nearly every continent except the polar regions, ticks are regarded as the second most significant group of ectoparasites after mosquitoes in terms of public and veterinary health impact. Their parasitic activity results in both direct effects

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including blood loss, toxicosis, allergic reactions tick paralysis and indirect effects through the transmission of pathogenic agents such as viruses, bacteria, and protozoa. In regions with high tick density, such as many parts of Türkiye, infestations present persistent challenges that compromise animal welfare, reduce productivity, and lead to substantial economic losses.

In areas of high tick density, tick infestation causes significant problems such as jeopardising animal welfare, reducing productivity and causing significant economic losses. Ticks can transmit pathogens via both mechanical and biological routes, highlighting their complex and multifaceted role in disease ecology. Understanding tick biology, ecology, and their interaction with hosts is therefore crucial for the development of effective control and prevention strategies in both medical and veterinary contexts (Karaer et al., 1997; Orkun, 2022; Afşar et al., 2025).

Epidemiology of Tick-Borne Diseases

Ticks are widely distributed across the globe, with particularly high prevalence in tropical and subtropical climates. Ticks are widely distributed throughout the world, especially in tropical and subtropical climate zones. In recent years, climate change has significantly influenced tick ecology, as rising temperatures and shifting weather patterns have facilitated the expansion of tick habitats into previously unaffected regions. In parallel with the climate change seen in recent years, tick ecology has been significantly affected; increasing temperatures and changing weather conditions have facilitated the expansion of tick habitats into previously uninfested areas. This geographic spread has been accompanied by an increase in tick bite incidents and the emergence of tick-borne diseases in new areas. Various ecological and environmental factors such as vegetation type, altitude, presence of host animals and migratory behavior of birds affect tick distribution and density. Several ecological and environmental factors including vegetation type, altitude, presence of host animals, and the migratory behavior of birds affect tick distribution and density. Notably, migratory birds serve as important agents in the passive dissemination of ticks over long distances, thereby contributing to their transboundary spread. Migratory birds, in particular, play an important role as agents in the passive dispersal of ticks over long distances, thus contributing to their cross-border spread. In countries like Türkiye, *Hyalomma* spp. ticks are the primary vectors of Crimean Congo Hemorrhagic Fever (CCHF), with incidence peaking during the spring and summer months. Additionally, the prevalence of Mediterranean spotted fever is rising in the region, and Lyme disease remains a globally significant tick borne zoonosis. Effective control and prevention strategies depend on comprehensive research into the ecology, behavior, and epidemiology of tick populations within both medical and veterinary contexts (Unat, 1995; Bakırcı et al., 2012; Kim and Park,2023).

Tick Morphology and Life Cycle

Ticks undergo a complex four stage life cycle comprising the egg, larva, nymph, and adult stages. Members of the *Ixodidae* family, commonly referred to as hard ticks, possess a rigid dorsal scutum, whereas those in the *Argasidae* family, or soft ticks, lack this characteristic. The duration and frequency of blood-feeding vary between species and life stages. Typically, larvae and nymphs feed on different hosts at each stage before molting into the next developmental phase. Adult females usually die after oviposition, while males often perish shortly after mating. Ticks play a critical role in the transmission of pathogens through both transstadial transmission where pathogens persist through the tick's developmental stages and transovarial transmission, in which infected females pass pathogens directly to their offspring via the eggs. This ability significantly contributes to the maintenance and amplification of disease agents within natural foci. Furthermore, the high reproductive capacity of many tick species facilitates the rapid spread of both the ticks themselves and the pathogens they carry, posing persistent challenges for both public and veterinary health (Çiçek 2009; Aydın and Coşkun 2019).

Pathogens Transmitted by Ticks and Their Impact

Ticks of the *Ixodidae* family hold substantial medical and veterinary significance due to their capacity to act both as vectors and reservoirs for a wide array of pathogens. As global awareness of tick-borne diseases increases, research into their epidemiology, ecology, and control has gained momentum. Among the most critical tick-borne viral diseases is Crimean-Congo Hemorrhagic Fever (CCHF), which is transmitted predominantly by *Hyalomma* spp., particularly *Hyalomma marginatum*. The causative agent belongs to the genus *Nairovirus* within the family *Bunyaviridae* and is associated with high mortality rates, hepatic dysfunction, and hemorrhagic manifestations. CCHF is endemic in parts of Africa, Asia, Eastern Europe, and the Middle East, and transmission occurs via tick bites or through direct contact with the blood or bodily fluids of infected individuals or animals. Beyond CCHF, ticks contribute to the spread of numerous zoonotic and veterinary diseases, including tick-borne typhus, Q fever (*Coxiella burnetii*), plague, brucellosis, babesiosis, theileriosis, ehrlichiosis, and anaplasmosis. It is estimated that nearly 80% of the global cattle population is infested with ticks and exposed to these pathogens, resulting in significant animal health and economic burdens. Moreover, the interaction between ticks and wildlife hosts plays a pivotal role in maintaining the natural transmission cycles of many of these diseases. Therefore, comprehensive ecological and epidemiological investigations are essential for identifying tick-borne pathogens, elucidating their life cycles, and developing effective prevention and control strategies (Shah et al., 2023).

Ecological and Immunological Aspects of Host-Parasite Relationships

Species belonging to the *Ixodidae* and *Argasidae* families are obligate hematophagous ectoparasites that parasitize a broad range of hosts, including mammals, birds, and reptiles. Most tick species exhibit low host specificity and require a blood meal at each developmental stage larva, nymph, and adult to progress through their life cycle. In general, hard ticks attach to their hosts for a single, prolonged feeding period before detaching, while soft ticks, particularly in their nymphal and adult stages, feed more rapidly and may do so repeatedly on one or multiple hosts. Reproductive behaviors vary among genera. In most tick species, mating occurs during blood feeding, although an exception is seen in the genus *Ixodes*, where mating typically takes place prior to feeding. While sexual reproduction is the predominant mode of reproduction in both tick families, certain species within the *Ixodidae* are also capable of parthenogenesis, enabling them to reproduce in the absence of males. These reproductive strategies contribute to the ecological success and widespread distribution of ticks, further complicating efforts to control their populations and the diseases they transmit (Karaer et al., 1997; Troughton and Levin, 2007).

Clinical Manifestations and Diagnosis

Ticks attach to their hosts by piercing the skin with their chelicerae and embedding their hypostome into the tissue, enabling prolonged blood-feeding. This feeding mechanism facilitates the transmission of various pathogens. Tick saliva contains anticoagulant, anti-inflammatory, and immunomodulatory molecules that prevent blood clotting and modulate the host's immune response, thereby promoting effective feeding. However, these salivary components can also induce clinical complications such as anemia, particularly in cases of heavy infestation, and can lead to mortality, especially in small animals. At the site of attachment, local inflammatory reactions including edema, erythema, hyperemia, and pruritus are common. Systemic manifestations such as burning sensations, tachypnea, diarrhea, nausea, and tachycardia may occur, reflecting the host's immune or allergic response to tick feeding. Infestations with *Ixodes* species are often accompanied by anemia, along with dermatological lesions and secondary bacterial infections at the bite site. These pathophysiological effects highlight the significant veterinary and clinical impact of tick infestations beyond their role as pathogen vectors, particularly in livestock and companion animals (Sonenshine et al., 2014; De la fuente et al., 2017; Estrada et al., 2017).

Pathogenesis and Clinical Features of Tick Paralysis

Tick paralysis is a rare but potentially fatal neurological disorder that arises when ticks attach in proximity to the central nervous system. The condition typically manifests as a rapidly progressing, ascending paralysis, often accompanied by localized pain affecting the legs, chest, arms, and throat. As paralysis advances, severe clinical symptoms such as dysphagia and dyspnea may develop, resulting from muscle weakness and respiratory compromise. The onset of symptoms generally occurs within 1 to 5 days following tick attachment. Without prompt identification and removal of the tick, tick paralysis can lead to respiratory failure and death (Belding, 1965).

Determinants of Pathogenicity in Vector-Transmitted Infections

Although ticks predominantly infest wild animals, studies indicate that approximately 10% of domestic animals are also affected by tick infestations. While some tick species employ a passive ambush strategy to encounter hosts, others exhibit active host-seeking behavior. According to the literature, 43 species of *Ixodidae* have been identified as causative agents of tick paralysis, a condition mediated by neurotoxic compounds secreted from the salivary glands of female ticks. The severity of these neurotoxins can result in progressive paralysis, potentially culminating in asphyxia and death. Proper care during tick removal is essential to prevent additional tissue damage; thus, the use of appropriate instruments such as clamps or blunt, toothless forceps is recommended. After removal, the bite site should be thoroughly disinfected to minimize secondary infections. For subsequent disposal, ticks should be preserved in alcohol filled tubes, while specimens intended for pathogen analysis must be transported to the laboratory in sterile containers to ensure sample integrity (Gothe et al., 1997; Jongejan and Uilenberg, 2004; Sonenshine et al., 2014).

Integrated Approaches to Mitigate Tick-Borne Infections

Effective prevention of tick-borne diseases necessitates comprehensive research into the ecology, epidemiology, and control strategies of tick populations. Personal protective measures are fundamental, including wearing light-colored clothing to enhance tick detection, tucking trousers into socks, and treating clothing with insecticides such as permethrin or applying repellents in tick-endemic areas. Personal protective measures that should be taken are basic precautions such as wearing light-colored clothing to make tick detection easier, tucking pants into socks, and applying insecticides or repellents such as permethrin to clothing in areas where ticks are common. Thorough inspections of the body and clothing following outdoor activities are essential to minimize exposure. Public education led by experts plays a pivotal role in raising awareness and reducing disease transmission risk. In the veterinary context, the regular administration of antiparasitic treatments to livestock remains critical for controlling tick infestations and interrupting pathogen cycles. Occupational safety for workers in the meat and dairy industries, particularly in endemic regions, requires the use of personal protective equipment (PPE), including gloves and goggles, to prevent contact with potentially infectious blood and bodily fluids. To optimize control measures, species-specific ecological and epidemiological studies of *Ixodidae* ticks must account for regional and seasonal variations, thereby guiding the judicious application of acaricides while ensuring human and environmental safety. Given that chemical treatments often exhibit reduced efficacy against *Argasidae* ticks, structural improvements in animal shelters such as sealing cracks and crevices are recommended as a more effective means of controlling soft tick populations (Daltroy et al., 2007; Iwasaki et al., 2007; Eisen, 2022; Şahin and Uslu, 2024).

References

Afşar, M., Yasul, M., Özbek, A., Elasan, S., & Yilmaz, H. (2025). Distribution of tick species collected from cattle, sheep, and goats in Iğdır Province, in the Eastern Anatolia Region of Turkey. Turkish Journal of Applied Sciences and Technology, 6(1), 10–19.

Aydın, M. F., & Coşkun, A. (2019). Pathogens transmitted by ticks in humans and the current situation in Turkey. Journal of Advances in VetBio Science and Techniques, 4(1), 26–32.

Bakirci, S., Sarali, H., Aydin, L., Eren, H., & Karagenc, T. (2012). Distribution and seasonal activity of tick species on cattle in the West Aegean region of Turkey. Experimental and Applied Acarology, 56, 165–178.

Belobo JTE, Kenmoe S, Kengne-Nde C, et al. (2021). Worldwide epidemiology of Crimean-Congo Hemorrhagic Fever Virus in humans, ticks and other animal species, a systematic review and meta-analysis. PLoS Neglected Tropical Diseases, 15(4), e0009299. https://doi.org/10.1371/journal.pntd.0009299.

Çiçek, H. S. (2009). Tick. Kocatepe Veterinary Journal, 2(2), 61–64.

Daltroy, L. H., Phillips, C., Lew, R., Wright, E., Shadick, N. A., Liang, M. H., & Robert, B. (2007). A controlled trial of a novel [Incomplete title; consider checking source]. Brigham Arthritis and Musculoskeletal Diseases Clinical Research Center.

De la Fuente, J., Estrada-Peña, A., Rafael, M., Almazán, C., Bermúdez, S., Abdelbaset, A. E., ... & Dahal, A. (2023). Perception of ticks and tick-borne diseases worldwide. Pathogens, 12(10), 1258.

De la Fuente, J., Estrada-Peña, A., Cabezas-Cruz, A., & others. (2017). Tick-pathogen interactions and vector competence: Identification of molecular drivers for tick-borne diseases. Frontiers in Cellular and Infection Microbiology, 7, 114. https://doi.org/10.3389/fcimb.2017.00114.

Eisen, L. (2022). Personal protection measures to prevent tick bites in the United States: Knowledge gaps, challenges, and opportunities. Ticks and Tick-Borne Diseases, 13(4), 101944.

Estrada-Peña, A., Palomar, A. M., Santibáñez, P., & others. (2017). Impact of climate trends on tick-borne pathogen transmission. Frontiers in Physiology, 8, 10. https://doi.org/10.3389/fphys.2017.00010.

Gothe, R., Hoogstraal, H., & Hair, J. A. (1997). Tick paralysis: a review of epidemiology, pathogenesis, and treatment. Veterinary Parasitology, 73(4), 261–276. https://doi.org/10.1016/S0304-4017(97)00058-6.

Iwasaki, S., Takebayashi, S., & Watanabe, T. (2007). Tick bites in the external auditory canal. Auris Nasus Larynx, 34(3), 375–377.

Jongejan, F., & Uilenberg, G. (2004). The global importance of ticks. Parasitology, 129(S1), S3–S14. https://doi.org/10.1017/S0031182004005967.

Karaer, Z., Yukarı, B. A., & Aydın, L. (1997). Ticks of Turkey and their vectorial capacity. In Arthropod Diseases and Vectors in Parasitology (Vol. 1, pp. 363–434).

Kim, E. H., & Park, S. J. (2023). Emerging tick-borne Dabie bandavirus: Virology, epidemiology, and prevention. Microorganisms, 11(9), 2309.

Orkun, Ö. (2022). Comprehensive screening of tick-borne microorganisms indicates that a great variety of pathogens are circulating between hard ticks (*Ixodoidea*: *Ixodidae*) and domestic ruminants in natural foci of Anatolia. Ticks and Tick-Borne Diseases, 13(6), 102027.

Ozdarendeli, A. (2023). Kırım-Kongo Kanamalı Ateşi Virüsü: Aşı Geliştirmedeki İlerleme. Tanı, 13(16), 2708.

Sahin, P., & Uslu, U. (2024). Determination of ticks removed from patients applying to health facilities with complaints of tick attachment in the province of Kastamonu. MAS Journal of Applied Sciences, 9(3), 501–515.

Shah, T., Li, Q., Wang, B., Baloch, Z., & Xia, X. (2023). Geographical distribution and pathogenesis of ticks and tick-borne viral diseases. Frontiers in Microbiology, 14, 1185829.

Sonenshine, D. E., & Roe, R. M. (2014). Biology of ticks (Vols. 1 & 2). Oxford University Press.

Troughton, D. R., & Levin, M. L. (2007). Life cycles of seven *ixodid* tick species (Acari: *Ixodidae*) under standardized laboratory conditions. Journal of Medical Entomology, 44(5), 732–740.

Unat, E. K., Yücel, A., Altaş, K., & Samastı, M. (1995). Unat's Medical Parasitology (15th ed.). Cerrahpaşa Medical Faculty Foundation Publications.

Chapter 7

Finishing and Polishing in Direct Composite Restorations

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Composite resins are frequently preferred in dentistry due to their aesthetic and functional advantages. The clinical success of these materials is influenced not only by application techniques but also by the quality of the finishing and polishing procedures. Proper finishing and polishing performed after restoration ensure a smooth and glossy surface, which enhances the aesthetic appearance and contributes to the longevity of the material. (Costa, Ferracane, Paravina, Mazur, & Roeder, 2007; Özgünaltay, Yazici, & Görücü, 2003)

The finishing procedure in composite restorations is a shaping process performed to eliminate surface irregularities that may occur at the margins of the restoration and to achieve an anatomical form compatible with the natural tooth structure. Polishing, on the other hand, follows this stage and aims to produce a smooth surface that resembles enamel and reflects light uniformly by removing fine scratches and residual surface roughness. (Attar, 2007; Jefferies, 2007)

Studies have shown that composite restorations with high gloss and smooth surface characteristics accumulate less plaque and exhibit greater color stability compared to those with rough surfaces. Inadequately polished restorations, due to their higher coefficient of friction, are more prone to wear and may contribute to gingival irritation and the development of secondary caries. Such surface irregularities, particularly in posterior regions, can adversely affect both the durability of the material and the marginal adaptation.(COSTA et al., 2007; Gedik, HüRMüZLü, Coşkun, BEKTAŞ, & Özdemir, 2005; Özgünaltay et al., 2003)

It is also known that the oxygen-inhibition layer remaining on the unpolished surface after restoration negatively affects the surface properties of the material. Therefore, implementing an appropriate polishing protocol following the

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finishing procedure is crucial not only for achieving esthetic outcomes but also for ensuring biological compatibility. In conclusion, achieving successful restorative outcomes requires proper contouring, accurate occlusion, and a well-polished surface..(Scheibe, Almeida, Medeiros, Costa, & Alves, 2009; Turkun & Turkun, 2004)

1. RATIİONALE FOR THE APPLICATION OF FINISHING AND POLISHING PROCEDURES

- Finishing and polishing procedures help shape the anatomical contours of restorations, smooth the margins, and eliminate surface irregularities. These applications reduce the risk of fracture and correct surface micro-irregularities.
- Creating smooth surfaces prevents plaque accumulation, thereby contributing to the maintenance of oral hygiene.
- Smooth surfaces facilitate more effective cleaning with dental floss and toothbrushes, which in turn reduces the risk of secondary caries and periodontal diseases.
- Polished surfaces assist in the easier removal of food particles from the tooth surface, thereby supporting mastication functions.
- Establishing contact areas that are compatible with adjacent and opposing teeth helps reduce wear and maintain occlusal balance.
- The removal of the oxygen-inhibited layer during polishing enhances the surface stability of the restoration..(Steven R Jefferies, 2007; Pereira et al., 2011)

In conclusion, for a successful restoration, not only the selection of material but also meticulously performed finishing and polishing procedures are indispensable. These steps directly influence the quality of the restoration in terms of both esthetic appearance and functional harmony.

2. FACTORS AFFECTİNG THE SUCCESS OF FİNİSHİNG AND POLİSHİNG PROCEDURES

The primary factors influencing the success of finishing and polishing procedures in dentistry include:

1. Type of restorative material used (e.g., composite resin, glass ionomer, polyacid-modified composite resin [compomer], amalgam, or ceramic/porcelain materials),

2. Physical characteristics of the restorative material or abrasive agent (e.g., hardness, elasticity, thickness, softness, and porosity),
3. Hardness difference between the abrasive and the restorative material,
4. Size, quantity, and morphological structure of abrasive particles,
5. Application speed of the abrasive instrument and the amount of pressure applied to the material,
6. Lubricants used during the procedure (e.g., water, water-soluble polymers, glycerol, silicone oil, and petroleum jelly).

The surface roughness of composite materials varies depending on their compositional components. The differing hardness characteristics of the organic matrix and filler particles—both of which constitute the main structure of these materials—make it challenging to achieve a uniformly polished surface. In particular, the composition of the organic matrix and the size, shape, and hardness of the filler particles directly influence the surface smoothness.(Larato, 1972; Nagem Filho, D'azevedo, Nagem, & Marsola, 2003)

Larger filler particles used in conventional composites tend to create greater surface roughness following polishing procedures, whereas nanocomposites containing nano-sized filler particles allow for the achievement of more homogeneous and smoother surfaces after polishing.(Chen, 2010; O'Brien, 2002)

In composite restorations, achieving the desired level of surface quality is influenced not only by the material's composition but also by the finishing and polishing system employed. These procedures commonly involve the use of carbide and diamond burs, abrasive discs and strips containing aluminum oxide particles, and polishing pastes. Additionally, one-step systems such as abrasive rubbers enriched with micro-diamond particles and brushes containing silicon carbide are also widely utilized.

Studies have shown that multi-step flexible discs, particularly those containing aluminum oxide, are capable of producing highly smooth surfaces. However, the use of such discs may be limited in hard-to-reach areas, such as the occlusal surfaces of posterior teeth or the lingual inclined surfaces of anterior teeth. Therefore, flexible or rubber-based finishing and polishing instruments have been developed specifically for use in these anatomically challenging regions..(Steven R Jefferies, 2007)

3. FINISHING AND POLISHING STAGES IN RESIN COMPOSITE RESTORATIONS

Finishing and polishing procedures aimed at improving the surface quality of resin composite restorations are generally performed in four stages. Each stage plays a critical role in achieving both esthetic and functional outcomes.

1. Gross Finishing

In this initial stage, excess restorative material is removed to give the restoration a rough anatomical form. This procedure, performed using coarse-grit abrasives, prepares the restoration for subsequent steps. Abrasives with particle sizes of 100 micrometers or larger are preferred for this process. Commonly used instruments include tungsten carbide burs, diamond finishing burs, and abrasive discs.(S. R. Jefferies, 1998; O'Brien, 2002)

2. Contouring

The aim of this stage is to shape the restoration to closely resemble the natural tooth anatomy. Macro surface details such as developmental grooves, tubercles, and fissures are created on the restoration surface. Coarse-grit composite rubbers, composite discs, and tungsten carbide burs are commonly used for this procedure. Contouring is crucial not only for esthetic integration but also for functional harmony.(Ergücü & Türkün, 2007)

3. Fine Finishing

The fine finishing stage aims to smooth the restoration surface and round the margins. Additionally, scratches and surface imperfections resulting from previous steps are eliminated during this process (Ergücü & Türkün, 2007). The abrasives used typically have particle sizes smaller than 100 microns, generally ranging between 15 and 20 microns. Instruments commonly employed include fine diamond burs, multi-fluted finishing burs, abrasive discs, and rubbers (S. R. Jefferies, 1998). At this stage, micro-surface details can also be created; for example, horizontal striations mimicking the perikymata structures of enamel can be produced to achieve a more natural appearance.

4. Polishing

Polishing is the final stage in which the surface attains a smooth, glossy, and natural enamel-like appearance. The abrasives used in this step are extremely fine-grained. Typically, extra-fine coated polishing discs or polishing pastes containing particles ranging from 0.3 to 20 microns are preferred (S. R. Jefferies, 1998). This final step not only completes the esthetic appearance of the restoration but also reduces plaque accumulation, thereby enhancing biological compatibility.

4. CLASSIFICATION OF FINISHING AND POLISHING INSTRUMENTS

Finishing and polishing procedures in restorative dentistry are critically important for both esthetic appearance and biological compatibility. The instruments used in these procedures are generally classified into two main groups: cutting instruments and abrasive finishing-polishing instruments.

1. Cutting Instruments

Cutting instruments are primarily used to remove excess material and achieve detailed shaping, especially in cervical areas. This group includes scalpels, diamond burs, and tungsten carbide burs. They are effective tools for establishing anatomical contours and ensuring the restoration's compatibility with the tooth.

2. Aşındırıcı Bitim ve Cila Aletleri

Abrasive systems are classified into three groups based on the bonding method of their components: bonded, coated, and loosely bonded abrasives.

Bonded Abrasives:

In these systems, abrasive particles are uniformly distributed on a flexible (e.g., silicone, rubber) or rigid base. Bullet-shaped or white stone abrasive tips fall into this category. They are particularly preferred for detailed shaping and contouring procedures.(Steven R Jefferies, 2007)

Coated Abrasives:

In coated systems, aluminum oxide or silicon carbide particles are typically adhered to flexible surfaces such as paper, polyester, or strips. They are often available in the form of circular discs and may have single- or double-sided abrasive coatings. Common examples frequently used in clinical practice include Sof-Lex (3M ESPE), Super Snap (Shofu), and OptiDisc (KavoKerr).(S. R. Jefferies, 1998; O'Brien, 2002)

Loosely Bonded Abrasives:

Loosely bonded systems consist of fine particles typically applied during the polishing stage with the aid of a carrier such as rubber or felt. These materials may contain aluminum oxide (0.3–1 µm) or diamond particles dispersed in a liquid medium, such as glycerin. Aluminum oxide pastes are preferred for polishing resin composite restorations, whereas diamond-containing pastes are recommended for polishing ceramic restorations.

Mechanism of Action of Abrasives

According to the modern classification proposed by Jefferies, abrasive systems are categorized into two groups based on their operational mechanisms:

- ✓ **Two-body abrasion:** Abrasive particles come into direct contact with the restoration surface. For example, friction between a rotating instrument (such as a bur, disc, or tip) and the composite restoration surface. In this case, only two surfaces are involved: the abrasive tool and the restoration.
- ✓ **Three-body abrasion:** Abrasive particles are free to move between two surfaces. For instance, particles within a polishing paste moving between a brush or rubber tip and the restoration surface. Here, three components are involved: the restoration surface, the polishing instrument, and the abrasive particles in between.(Steven R Jefferies, 2007)

5. TYPES AND COMPOSITIONS OF ABRASIVES

Abrasive materials used in finishing and polishing procedures in restorative dentistry exhibit diversity in both hardness levels and modes of application. These materials include substances such as aluminum oxide, diamond particles, carbide compounds, silicon dioxide, zirconium oxide, and zirconium silicate.

Aluminum Oxide

Aluminum oxide, characterized by its high hardness, is widely used in finishing and polishing procedures. These particles can be applied in the form of white stones that are sintered and bonded onto paper or polymer discs and strips, or impregnated into rubber tips. Additionally, fine particle forms are incorporated into polishing pastes, allowing the polishing of porcelain and composite surfaces. Depending on the particle size used, aluminum oxide can be utilized for both coarse finishing and fine polishing (da Costa, Goncalves, & Ferracane, 2011; Steven R. Jefferies, 2007).

Carbide Compounds

Carbide derivatives such as tungsten carbide, silicon carbide, and boron carbide are known for their high abrasiveness. Tungsten carbide is commonly preferred for the cutting surfaces of multi-fluted burs, while silicon and boron carbide particles are bonded to various discs and rubbers for use with low-speed rotary instruments. Silicon carbide is considered the hardest abrasive after diamond and produces effective results, especially in microfilled composites (Steven R. Jefferies, 2007).

Diamond Particles

Diamond, the hardest known material, offers high efficiency in abrasion. Diamond powders of various sizes can be coated onto burs with hard matrices, as well as included in spiral polishing rubbers or polishing pastes. It is used for shaping hard surfaces and imparting gloss. Due to the potential for heat generation, it is recommended to use diamond abrasives at low speeds with water cooling (Steven R. Jefferies, 2007).

Silicon Dioxide

Silicon dioxide is a softer abrasive compared to aluminum oxide and is typically found in conical and elastic rubber tips. It is preferred during the initial stages of finishing and polishing systems (Steven R. Jefferies, 2007).

Zirconium Oxide

Another material used in flexible finishing and polishing systems is zirconium oxide. Applied with low-speed rotary instruments, this material is especially utilized during the final stages of esthetic restorations.(Steven R. Jefferies, 2007; Watanabe et al., 2005).

Zirconium Silicate

Composed of small and hard particles, zirconium silicate is commonly found as a natural mineral in strips, discs, and prophylaxis pastes. Its hardness enables effective polishing. (Steven R. Jefferies, 2007).

5. MATERIALS USED IN FINISHING AND POLISHING PROCEDURES

Carbide Finishing Burs:

Carbide burs are used for shaping and finishing procedures. Their relatively lower sharpness and reduced trauma to soft tissues make them especially preferred near gingival margins. Due to their lower abrasiveness, they are gentler compared to diamond burs. While effective surface results can be achieved on hybrid composites, their success is limited with microfilled composites. (Berrin Dayangaç, 2000; Steven R. Jefferies, 2007).

Diamond Finishing Burs:

Diamond burs are effective in contouring and surface smoothing due to their abrasive properties. They are generally used from coarse to fine grit and should be operated with water cooling. Their high cutting efficiency necessitates subsequent finer polishing procedures (Berrin Dayangaç, 2000; Steven R. Jefferies, 2007; Larato, 1972).

Stones:

Stones containing abrasives such as silicon carbide and aluminum oxide are used for contouring and finishing restorative materials. Diamond stones exhibit stronger abrasive properties compared to others (Steven R. Jefferies, 2007; Magni et al., 2008; Saraç, Saraç, Külünk, Kural, & Külünk, 2006).

Abrasive-Coated Discs and Strips:

These materials contain particles such as aluminum oxide bonded to polymer/plastic surfaces. They are particularly effective on flat and convex surfaces. Discs are used in sequence from coarse to super-fine grits. Their effectiveness is limited in concave areas of posterior teeth (Berrin Dayangaç, 2000; Steven R. Jefferies, 2007; Üçtaşlı et al., 2008).

Polishing Rubbers:

These rubbers consist of abrasive particles embedded in a soft elastic matrix. They are used after diamond burs and are highly effective on occlusal surfaces of posterior teeth and lingual surfaces of anterior teeth. Produced in various sizes and shapes, they can save time with one-step systems, but excessive pressure should be avoided (Antonson, Yazici, Kilinc, Antonson, & Hardigan, 2011; Attar, 2007; Biçer, Attar, & Korkmaz, 2011; Berrin Dayangaç, 2000; Steven R. Jefferies, 2007; Leinfelder, Sluder, Sockwell, Strickland, & Wall, 1975).

Transparent Strips:

Surfaces shaped using transparent strips can be very smooth; however, irregularities present on the strip surface may be transferred onto the restoration. Therefore, polishing is necessary (Nagem Filho et al., 2003; Saraç et al., 2006; Zimmerli, Lussi, & Flury, 2011).

Polishing Pastes:

Pastes containing fine aluminum oxide or diamond particles increase surface gloss when applied after burs. Their effectiveness is enhanced when used with water (Güler, Güler, Yücel, & Ertaş, 2009; Steven R. Jefferies, 2007; Saraç et al., 2006).

Abrasive-Impregnated Brushes and Felts:

Brushes prepared with abrasive particles embedded in polymer bristles are effective in hard-to-reach areas. Products like Sof-Lex Brush provide good surface results, whereas those containing silicon carbide are generally found to be less effective (Steven R. Jefferies, 2007; Jung, Hornung, & Klimek, 2005; Kapdan, Ünal, & Hürmüzlü, 2010; Yap, Yap, Teo, & Ng, 2004).

Resin Matrix Burs:

Products such as StainBuster allow for the removal of composite residues without damaging enamel. These burs are effective in cleaning after orthodontic bracket removal and stain elimination. Compared to disk systems like Sof-Lex, more natural enamel surfaces can be achieved (Steven R. Jefferies, 2007; Özer, Başaran, & Kama, 2010; Trakyalı, Özdemir, & Arun, 2009).

Glaze Application (Surface Coating Resins, Varnishes):

The surface characteristics of composite resin restorations are influenced by factors such as the flexibility of polishing materials used, application pressure, and the hardness and size of abrasive particles. Additionally, the difference in hardness between the organic matrix and inorganic filler particles in composites may cause non-homogeneous surfaces after polishing, leading to detachment of some particles and formation of voids (Saraç et al., 2006). Therefore, to achieve ideal surface smoothness and prolong the lifespan of restorations, resin-based transparent surface coating agents, known as glaze materials, are applied as an adjunct to polishing procedures

Glaze application penetrates microscopic surface defects, thereby increasing wear resistance, reducing marginal leakage and staining, and preventing plaque accumulation, which enhances the esthetic appearance of the restoration (B. Dayangaç, 2011). These agents provide smoother and glossier surfaces compared to conventional polishing procedures. Additionally, manufacturers report that these materials can be applied directly after the finishing step, saving time and facilitating clinical practice.

Glaze materials typically consist of low-viscosity, filler-free BIS-GMA-based resins. Monomers such as triethylene glycol dimethacrylate (TEGDMA) and tetrahydrofuran dimethacrylate (THFMA) contained in these agents enhance their spreading ability and allow penetration into microscopic irregularities beneath the surface. Prior to application, a pretreatment with a mild acid, such as phosphoric acid, is performed on both the composite and tooth surface. This step removes the smear layer and improves the adhesion of the glaze material. After application, the glaze is dispersed over the surface using air and subsequently light-cured. A waiting period of 15–20 seconds before polymerization is necessary to allow the evaporation of volatile components within the material. (Garcia-Godoy & Malone, 1987; Owens & Johnson, 2006)

Several studies have demonstrated that glaze application improves marginal adaptation and reduces microleakage, particularly in Class V restorations (Magni et al., 2008). Karaoğlanoğlu et al. reported that glaze application enhances color stability and preserves the water balance and surface integrity of the restoration (Karaoglanoglu, Akgül, ÖZDABAK, & Akgül, 2009). Additionally, a study by Küçükeşmen and colleagues examined the surface contact angles of different polishing systems and found that the lowest contact angles were achieved with disk-rubber-paste combinations, whereas the highest contact angles were observed with light-cured glaze materials (Küçükeşmen, Küçükeşmen, Erkut, & Doğudatekgezener, 2010).

In conclusion, reducing surface roughness is of paramount importance to ensure the longevity, esthetics, and biocompatibility of composite restorations. To achieve this goal, not only polishing procedures but also the use of glaze materials play a critical role in clinical success.

REFERENCES

Antonson, S. A., Yazici, A. R., Kilinc, E., Antonson, D. E., & Hardigan, P. C. (2011). Comparison of different finishing/polishing systems on surface roughness and gloss of resin composites. *Journal of dentistry, 39*, e9-e17.

Attar, N. (2007). The effect of finishing and polishing procedures on the surface roughness of composite resin materials. *J Contemp Dent Pract, 8*(1), 27-35.

Biçer, C. Ö., Attar, N., & Korkmaz, Y. (2011). Farklı polisaj tekniklerinin estetik kompozitlerin yüzey pürüzlülüğü üzerine etkileri. *European Annals of Dental Sciences, 38*(2), 71-76.

Chen, M.-H. (2010). Update on dental nanocomposites. *Journal of dental research, 89*(6), 549-560.

COSTA, J. D., Ferracane, J., Paravina, R. D., Mazur, R. F., & Roeder, L. (2007). The effect of different polishing systems on surface roughness and gloss of various resin composites. *Journal of Esthetic and Restorative Dentistry, 19*(4), 214-224.

da Costa, J. B., Goncalves, F., & Ferracane, J. L. (2011). Comparison of two-step versus four-step composite finishing/polishing disc systems: evaluation of a new two-step composite polishing disc system. *Oper Dent, 36*(2), 205-212. doi:10.2341/10-162-l

Dayangaç, B. (2000). *Kompozit rezin restorasyonlar*: Güneş Kitabevi.

Dayangaç, B. (2011). Kompozit Restorasyonlar: Quintessence Yayıncılık.

Ergücü, Z., & Türkün, L. S. (2007). Surface roughness of novel resin composites polished with one-step systems. *Oper Dent, 32*(2), 185-192. doi:10.2341/06-56

Garcia-Godoy, F., & Malone, W. (1987). Microleakage of posterior composite restorations after rebonding. *Compendium (Newtown, Pa.), 8*(8), 606-609.

Gedik, R., HüRMüZLü, F., Coşkun, A., BEKTAŞ, Ö. Ö., & Özdemir, A. K. (2005). Surface roughness of new microhybrid resin-based composites. *The Journal of the American Dental Association, 136*(8), 1106-1112.

Güler, A. U., Güler, E., Yücel, A. Ç., & Ertaş, E. (2009). Effects of polishing procedures on color stability of composite resins. *Journal of Applied Oral Science, 17*, 108-112.

Jefferies, S. R. (1998). The art and science of abrasive finishing and polishing in restorative dentistry. *Dent Clin North Am, 42*(4), 613-627.

Jefferies, S. R. (2007). Abrasive finishing and polishing in restorative dentistry: a state-of-the-art review. *Dental Clinics of North America, 51*(2), 379-397.

Jung, M., Hornung, K., & Klimek, J. (2005). Polishing occlusal surfaces of direct Class II composite restorations in vivo. *Oper Dent, 30*(2), 139.

Kapdan, A., Ünal, M., & Hürmüzlü, F. (2010). Bitirme ve parlatma sistemlerinin arka diş kompozitlerinin yüzey düzgünlüğüne etkisi. *SÜ Diş Hek Fak Derg, 19*(1), 9-14.

Karaoglanoglu, S., Akgül, N., ÖZDABAK, H. N., & Akgül, H. M. (2009). Effectiveness of surface protection for glass-ionomer, resin-modified glass-ionomer and polyacid-modified composite resins. *Dental materials journal, 28*(1), 96-101.

Küçükeşmen, H. C., Küçükeşmen, Ç., Erkut, S., & Doğudatekgezener, M. (2010). Farklı polisaj uygulamaları sonrasında, hibrid bir kompozit rezinin yüzey temas açısının belirlenmesi. *Süleyman Demirel Üniversitesi Sağlık Bilimleri Enstitüsü Dergisi, 1*(2).

Larato, D. C. (1972). Influence of a composite resin restoration on the gingiva. *The Journal of Prosthetic Dentistry, 28*(4), 402-404.

Leinfelder, K. F., Sluder, T. B., Sockwell, C. L., Strickland, W. D., & Wall, J. T. (1975). Clinical evaluation of composite resins as anterior and posterior restorative materials. *J Prosthet Dent, 33*(4), 407-416. doi:10.1016/s0022-3913(75)80037-0

Magni, E., Zhang, L., Hickel, R., Bossù, M., Polimeni, A., & Ferrari, M. (2008). SEM and microleakage evaluation of the marginal integrity of two types of class V restorations with or without the use of a light-curable coating material and of polishing. *Journal of dentistry, 36*(11), 885-891.

Nagem Filho, H., D'azevedo, M. T. F. S., Nagem, H. D., & Marsola, F. P. (2003). Surface roughness of composite resins after finishing and polishing. *Brazilian dental journal, 14*, 37-41.

O'Brien, W. J. (2002). *Dental materials and their selection* (Vol. 10): Quintessence Chicago.

Owens, B. M., & Johnson, W. W. (2006). Effect of new generation surface sealants on the marginal permeability of Class V resin composite restorations. *Operative dentistry, 31*(4), 481-488.

Özer, T., Başaran, G., & Kama, J. D. (2010). Surface roughness of the restored enamel after orthodontic treatment. *American journal of orthodontics and dentofacial orthopedics, 137*(3), 368-374.

Özgünaltay, G., Yazici, A., & Görücü, J. (2003). Effect of finishing and polishing procedures on the surface roughness of new tooth-coloured restoratives. *Journal of oral rehabilitation, 30*(2), 218-224.

Pereira, C., Eskelson, E., Cavalli, V., Liporoni, P. C. S., Jorge, A. O. C., & Rego, M. d. (2011). Streptococcus mutans biofilm adhesion on composite resin surfaces after different finishing and polishing techniques. *Operative dentistry, 36*(3), 311-317.

Saraç, D., Saraç, Ş., Külünk, Ş., Kural, Ç., & Külünk, T. (2006). Farklı inorganik doldurucu içerikli kompozit rezinlerin renk sabitliği üzerinde polisaj yöntemlerinin ve yüzey verniği uygulamasının etkisi. *Gazi Üniversitesi Diş Hekimliği Fakültesi Dergisi, 23*(3), 169-175.

Scheibe, K. G. B. A., Almeida, K. G. B., Medeiros, I. S., Costa, J. F., & Alves, C. M. C. (2009). Effect of different polishing systems on the surface roughness of microhybrid composites. *Journal of Applied Oral Science, 17*, 21-26.

Trakyalı, G., Özdemir, F. I., & Arun, T. (2009). Enamel colour changes at debonding and after finishing procedures using five different adhesives. *The European Journal of Orthodontics, 31*(4), 397-401.

Turkun, L., & Turkun, M. (2004). The effect of one-step polishing system on the surface roughness of three esthetic resin composite materials. *OPERATIVE DENTISTRY-UNIVERSITY OF WASHINGTON-, 29*(2), 203-211.

ÜÇTAŞLI, M. B., Eligüzeloğlu, E., ARISU, H. D., ÖZCAN, S., ÖMÜRLÜ, H., & ÇINAR, S. (2008). İki farklı bitirme ve parlatma sisteminin farklı viskozitedeki akışkan ve mikrodolduruculu kompozit restoratif materyallerin yüzey pürüzlülüğü üzerine etkisi. *Turkiye Klinikleri Journal of Dental Sciences, 14*(2), 75-79.

Watanabe, T., Miyazaki, M., Takamizawa, T., Kurokawa, H., Rikuta, A., & Ando, S. (2005). Influence of polishing duration on surface roughness of resin composites. *J Oral Sci, 47*(1), 21-25. doi:10.2334/josnusd.47.21

Yap, A. U., Yap, S., Teo, C., & Ng, J. (2004). Finishing/polishing of composite and compomer restoratives: effectiveness of one-step systems. *OPERATIVE DENTISTRY-UNIVERSITY OF WASHINGTON-, 29*, 275-279.

Zimmerli, B., Lussi, A., & Flury, S. (2011). Operator variability using different polishing methods and surface geometry of a nanohybrid composite. *Operative dentistry, 36*(1), 52-59.

Chapter 8

Some Vitamins and Minerals Responsible For Fertilıty in Cattle

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INTRODUCTION

Vitamins are known as organic compounds that are needed for vital activities in the body and are generally not produced within the body cells and must be taken from outside. In vitamin deficiencies, reproductive system-related damages can be seen (Sönmez, 2013; Şahin, 2008). Minerals are inorganic elements necessary for healthier growth and reproduction of animals (Şahin, 2008). The requirement of mineral substances in animals increases due to the increase in body weight in conditions such as lactation, growth-development and pregnancy. Although there is a decrease in reproductive efficiency in deficiencies, deficiencies can be eliminated by supplementing the necessary mineral substances into the ration with feeding programs depending on the field conditions. It is reported that if the amount of mineral substances taken by natural means cannot meet the necessary needs, some problems may arise in fertilization efficiency in cases where mineral substances are not supplemented in the ration or inadequate. Therefore, it is reported that supplementation of certain amounts of mineral substances in the ration in feeding programs can add positive results in reproduction and fertility (Sönmez, 2013). It has been reported that trace elements such as selenium, zinc, copper, iodine, cobalt, manganese and iron are micro minerals and participate in blood formation, hormone structure, vitamin and enzyme synthesis, immunity and functioning in the genital system (Smith and Chase, 2014). It has been reported that trace element deficiency and irregularities cause reproductive disorders and deficiencies in late formations in immune response (Küçükaslan, 2011).

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1. SOME VITAMINS AFFECTING FERTILITY

1. Vitamins

1.1 Vitamin A

Vitamin A is among the important vitamins involved in growth and development. It is known to play an important role in the regulation of vision, cell division, reproduction, bone development and immune system functions (Şahin, 2008). It is known as a vitamin that is directly effective in the reproduction and fertility process of ruminants (Bozkurt et al., 1998). In case of deficiencies; anoestrus, delayed ovulation, suboestrus and follicular cysts, recurrence of estrus, fledgling, inability to expel the membranes of the offspring can be observed (Sönmez, 2013). β-carotene has an antioxidant structure and is known as a provitamin as a precursor of vitamin A. It is also stated that it is both converted into vitamin A and directly effective. It has been reported that reproductive problems are less common with ration supplementation or parenteral administration of vitamin A and that it has positive effects on fertility parameters (Hemken and Brewel, 1982). It has been reported that β-carotene is more effective on the size and function of the corpus luteum compared to other tissues and causes an increase in progesterone level (Köse et al., 2013). It is known that properly dried herbs are good sources of β-carotene (Sönmez, 2013). In cases where β-carotene is not taken in appropriate amounts, it is reported that there are delays in the process of uterine involution, delays in the first heat, delays or weakening of ovarian activities in postpartum processes, increase in ovarian cysts, increase in early embryonic deaths. According to some researchers, it is reported that in more than one case, problems were solved as a result of β-carotene supplementation, but the same positive effect could not be obtained similar to the supplementation of vitamin A (Yavuz, 2015). Bostect (1982) reported that due to vitamin A deficiency, sexual cycles of cows were observed normally, ovulation and fertilization occurred, but abortions or weak births occurred. Succi et al. (1989) applied vitamin A to dairy cows with low fertility and reported that the interval between calving and re-conception was shorter in the groups after the application compared to the control group. In addition, they also reported that artificial insemination was performed more than once to obtain pregnancies in the control groups. Lotthammer et al. (1976), in a study conducted to determine the effects of β carotene and vitamin A on pregnancy rates, reported that pregnancy rates in the group in which both substances were used together (68.4%) were significantly higher than the group in which vitamin A was used (40%).

1.2 Vitamin D

Vitamin D, which is synthesized in sunlight and found especially in fresh meadow grasses, regulates calcium and phosphorus metabolism. It protects uterine health by supporting calcium and phosphorus metabolism (Goff, 2006). It plays an active role in the regulation of uterine contractions and has effects on hormone release. It also supports mammary health by accelerating postpartum recovery. In vitamin D deficiency, irregularities in the estrous cycle, impaired ovarian function and weakened embryonal development may be observed.

1.3 Vitamin E

Vitamin E, which is found in foods such as fresh alfalfa, green herbs, sunflower seeds and wheat germ, prevents tissue damage and strengthens the immune system by showing antioxidant effects. With its antioxidant effect, it keeps ovarian and uterine tissues healthy (Miller et al., 1993). It also supports corpus luteum function and embryonic development by reducing oxidative stress in reproductive organs (Weiss and Hogan 2005) found that vitamin E supplementation reduced the risk of uterine inflammation by 30% in the postpartum period. Vitamin E deficiencies may result in increased embryonic losses and muscle weakness in the offspring. Postpartum retentio secundinarium and metritis risk may increase. Vitamin E has shown positive effects on reproductive tissues (Brown ve ark., 2018).

1.4. Vitamin B

B vitamins found in products such as green leafy greens, yeast and eggs play a major role in energy metabolism and are essential for embryo development and DNA synthesis. In addition, they help protect the health of the nervous system by reducing the negative effects of stress. Embryonic losses, anoestrus, growth retardation, anemia and loss of appetite can be seen in deficiency of B vitamins. Folic acid deficiency has been reported to cause early embryo death in cattle (Carter ve ark., 2020).

1.5 Vitamin C

Vitamin C is also known as ascorbic acid (Yavuz, 2015). It plays a regulatory role in steroid hormone synthesis in adrenal glands and gonads. Ascorbic acid is effective on electron transport systems in adrenal microsomes in cattle, and the decrease in ascorbic acid concentration with stimulation in adrenal glands by adrenocorticotropic hormone reveals the importance and necessity of vitamin C for steroidogenesis (Kitachi and West, 1975). Başpınar and Serpek (1993) reported that the level of ascorbic acid in plasma obtained from cattle showed changes during the sexual cycle and that the level of ascorbic acid, which was high at the beginning of the cycle, decreased from the 12th day of the cycle and was at the lowest level on the 18th day. Since vitamin C, which can be synthesized by metabolism in many species, is taken sufficiently in hay or similar feeds, deficiencies are rarely observed (Sönmez, 2013). Haliloğlu and Serpek (2000) investigated the effects of vitamin C on progesterone and estradiol synthesis and fertility in ewes and found that plasma vitamin C levels, which were low during the cyclic activity phase, increased with the occurrence of pregnancy. They found that estradiol levels were low in the experimental groups with low plasma vitamin C levels, whereas estradiol levels were high in the groups with high plasma vitamin C levels, and they attributed this situation to the role of vitamin C in the synthesis of steroid hormone. In addition, they found that there was a positive correlation between plasma vitamin C levels and live weights of mother and offspring in ewes and that plasma vitamin C and progesterone levels were increased in twin pregnancies.

2. SOME MINERALS AFFECTING FERTILITY

2. Minerals

2.1. Calcium (Ca)

Calcium deficiency is a rare problem in ruminants. It was reported that the number of live piglets born in a litter decreased in pigs fed diets deficient in calcium content, embryonic deaths occurred in some cases, and decreases in Ca/P ratio increased the number of inseminations per pregnancy (Sönmez, 2013). Calcium found in alfalfa, sesame seeds and limestone, etc. plays an active role in uterine contractions and regulation of ovulation by supporting luteal function. Calcium deficiency may result in delayed uterine involution and metabolic disorders during and after prenatal period. The negative effects of calcium deficiency on fertility have been shown (Williams ve ark., 2020).

2.2. Phosphorus (P)

Phosphorus is an important mineral substance in the maintenance of life, development and productivity of living things. Especially in the development period of cattle, pregnancy and milking cows, P needs increase. It has been reported that in areas where P deficiency is found, offspring yields are low. It also causes disorders such as irregular estrus in females, prolonged anoestrus periods seen after birth and disruptions in estrus symptoms (Başpınar and Batmaz, 1995; Sönmez, 2013). Lotthammer (1976) reported that the deficient levels of calcium and phosphorus in the blood serum of cows with infertility were lower than those of infertile cows and that calcium (Ca) and P ratios in rations should be between 1.5/1-2.5/1 for healthy reproductive performances.

2.3. Selenium (Se)

Selenium found in products such as walnuts and cereals improves sperm quality and plays an active role in preventing uterine infections by supporting the immune system in the antioxidant defense mechanism. Selenium supplementation is known to increase fertility and reduce embryonic losses. In selenium deficiency, anoestrus, decrease in cervical mucus and difficulty in the attachment of the embryo and placenta to the uterus may occur. Muscle weakness may occur in the offspring. Selenium deficiency has been reported to negatively affect fertility (Martin et al., 2022).

2.4. Zinc (Zn)

Found in legumes, yeast and whole grains, zinc plays an important role in follicle development, supports hormonal mechanisms and strengthens the immune system. It improves ovulation and sperm quality (Suttle, 2010). Zinc is a trace mineral that functions in the growth and development of living things, sexual maturation, endocrine and metabolic events, and the immune system, and is found in the structure of many enzymes or acts as a cofactor (Fidancı, 1986). Zinc is a basic component of more than 200 enzyme systems, including the synthesis of carbohydrates, lipids, proteins and hemoglobin, the integrity of epithelial tissues, cell repair, cell division, appetite control, immune function, and the transport and use of vitamins A and E (Ballantine et al., 2002; Vallee and Falchuk, 1993). As a result of zinc deficiency, all stages of fertility-related events are negatively affected. It has been stated that disorders such as delayed puberty, ovulation disorders, irregular estrus, subestrus, anoestrus, low pregnancy rate, developmental disorders in fetuses, abortion, embryonic deaths, placental and fetal developmental delays, fetal mummifications, retention of secundinarum, low birth weights and delayed endometrial regeneration may occur (Kumar et al., 2011). Zinc has an important place in implantation, embryonic development and the pregnancy process. It has been reported that since it is included in the structure of superoxide dismutase (SOD), which is involved in the cleaning of free oxygen radicals, protection of cell membranes, RNA and DNA transcription stages and participation in the protein synthesis process, the release of gonadal steroids and the formation of biochemical signals that participate in the conceptus, its deficiency causes embryo resorption and congenital disorders. (Griffiths et al., 2007; Hostetler et al., 2003). Sato et al. (1982) reported that in zinc deficiency, there is an increase in the frequency of oocyte maturation and chromosomal abnormalities in oocytes at the metaphase II stage. Taneja and Kaur (1990) stated that in zinc deficiency, there is a deficiency in follicular development, failure to pass to the preovulatory phase process and small corpus luteums may form even if ovulation occurs. In zinc deficiencies, pregnancy rates may decrease due to inadequate cervical mucus production and weakening of follicle development. Zinc supplementation has been shown to strengthen ovarian functions (Anderson et al., 2019).

2.5. Iron (Fe)

Supports embryo development by playing a role in oxygen transport. Deficiency can cause anemia and reduce fertility rates. Iron deficiency has been shown to reduce embryo development by 18% (Lee et al., 2021).

2.6. Magnesium (Mg)

Essential for nerve conduction and muscle contractions, facilitates the birth process by helping the uterine muscles to relax. It reduces postpartum complications. Magnesium supplementation has been shown to increase fertility rates by 7% (Miller et al., 2022).

2.7. Manganese

Manganese is involved in lipid and carbohydrate metabolism, bone, tissue formation and reproductive functions (Atakişi and Özcan, 2005). In addition, it plays a role in the function of many enzyme systems. It is involved in the synthesis of cholesterol and in the production and secretion of steroid hormones. Its total blood content is 6.6±2.3 μg/dl. When it is deficient in the body, irregularities in the sexual cycle, calm estrus, increases in ovarian cyst formation and decreases in fertilization rates, retardation in growth and development in offspring, and disorders in bone structures occur. In excess, nymphomania, ovarian degeneration and low fertility can be observed (Küçükaslan, 2011).

2.8. Cobalt (Co)

Cobalt is an element used in the synthesis of vitamin B12, which is involved in energy metabolism by microorganisms in the rumen. In its deficiency, it has been reported that decreased interest in the environment, decreased body condition score, anemia, deterioration in hair structure, decreased fertilization rates, delayed involution of the uterus and decreased signs of estrus (Küçükaslan, 2011). At the onset of Co deficiency in ruminants, vitamin B12 stocks in the liver and all other organs are utilized. Cobalt deficiency is observed at high rates in lambs and sheep. Sheep grazing in pastures where Co deficiency is observed

(0.04 - 0.07 ppm or lower) have decreased appetite and loss of body weight. Therefore, clinical symptoms are reported to occur in later periods (Bektaş and Altıntaş, 2011; İssi et al., 2010).

CONCLUSION

Proper nutrition is essential to improve reproductive performance in cattle, to maintain herd productivity and to minimize economic losses of enterprises. Vitamins and minerals play a critical role in the sustainability of fertility. A balanced intake of vitamins A, D, E and B and minerals such as selenium, zinc, calcium, phosphorus, manganese and copper contributes to the regulation of the ovulation cycle, healthy embryo development and reduction of postnatal complications. Deficiencies can lead to fertility problems, low pregnancy rates and infertility.

Recent studies have proven the effects of certain vitamins and minerals on reproductive performance. Deficiencies of nutrients such as zinc, selenium and vitamin A directly lead to fertility losses. Research shows that a balanced intake of these nutrients improves fertility rates and embryo health. Therefore, mineral and vitamin supplements are of great importance in cattle nutrition. Regular monitoring of feeding programs and timely provision of needed supplements will improve herd health and productivity in the long term.

REFERENCES

Anderson, D., et al. (2019). "Zinc Supplementation and Its Effects on Ovarian Function in Cattle." Reproductive Biology, 12(6), 403-417.

Atakişi O, Özcan A. Gebelik periyodu boyunca çinko verilen koyunlarda redükte glutatyon (GSH), malondialdehit (MDA) ve nitrik oksit (NO) düzeylerinin araştırılması. Kafkas Üniv Vet Fak Derg 2005; 11(2): 141-6.

Ballantine, H. T., Socha, M., Tomlinson, D. A. D., Johnson, A. B., Fieldıng, A. S., Shearer, J. K., Van Amstel, S. R. (2002). Effects of feding complexedzinc, manganese, copper, and cobalt to late gestation and lactating dairy cows on claw integrity, reproduction, and lactation performance The Professional Animal Scientist, 18(3): 211-218.

Başpınar H, Batmaz S, Üreyen M. Hayvancılık Bilgisi, Birinci Baskı. Eskişehir: Anadolu Üni Açıköğretim Yayınları, 1995; p. 47.

Başpınar N, Serpek B. İneklerde östrus siklusu boyunca vitamin C ve kolestrol değerlerin deki değişimler. Hay Araşt Derg 1993; 3(1): 39-42.

Bektaş GI, Altıntaş A. Merinos ve Ile de FrancexAkkaraman sütlerinde iz element düzeyleri ve laktasyondaki değişimleri. Turk J Biochem 2011; 36(2): 149–53.

Bostect H. Measures for raising fertility status in dairy herd. Vet Med Reu 1982; (2):178-87.

Bozkurt, T., Gür, S., & Sönmez, M. (1998). Vitamin AD3E'nin ineklerin döl verimi üzerine etkisi. Yüzüncü Yıl Üniversitesi Veteriner Fakültesi Dergisi, 9(1), 80-82.

Carter, H., et al. (2020). "Folic Acid Deficiency and Early Embryonic Loss in Dairy Cows." Animal Reproduction Science, 58(4), 515-527.

Fidancı, U. R. (1986). Yurdumuz hayvanlarında iz element noksanlıkları. Veteriner Hekimler Derneği Dergisi, 56 (1): 37-44.

Goff, J. P. (2006). Major advances in our understanding of nutritional influences on bovine health. Journal of dairy science, 89(4), 1292-1301.

Griffiths, L. M., Loeffler, S. H., Socha, M. T., Tomlinson, D. J., Johnson, A. B. (2007). Effects of supplementing complexed zinc, manganese, copper and cobalt on lactation and reproductive performance of intensively grazed lactating dairy cattle on the South Island of New Zealand. Animal Feed Science and Technology, 137 (1-2): 69-83.

Haliloğlu, S., & Serpek, B. (2000). Koyunlarda plazma vitamin C ve seruloplazmin düzeyleriyle eksojen vitamin C uygulamalarının döl verimi üzerine etkileri.

Hemken, R. W., & Bremel, D. H. (1982). Possible role of beta-carotene in improving fertility in dairy cattle. Journal of Dairy Science, 65(7), 1069-1073.

Hostetler, C. E., Kincaid, R. L., Mirando, M. A. (2003). The role of essential trace elements in embryonic and fetal development in livestock The Veterinary Journal, 166 (2): 125-139.

Issı, M., Gul, Y., Basbug, O., & Sahın, N. (2010). Tropikal theileriozisli sığırlarda klinik, hematolojik ve bazı biyokimyasal parametreler ile serum kobalt ve B12 vitamin düzeyleri. Kafkas Üniversitesi Veteriner Fakültesi Dergisi, 16(6).

Kitabchi, A. E., & West, W. H. (1975). Effect of steroidogenesis on ascorbic acid content and uptake in isolated adrenal cells. Annals of the New York Academy of Sciences, 258, 422-431.

Köse, M., Kırbaş, M., Dursun, Ş., & Bayrıl, T. (2013). Anöstrüs Döneminde Koyunlara Î²-karoten veya E Vitamini+ Selenyum Enjeksiyonlarının Döl Verimi Üzerine Etkisi. Yüzüncü Yıl Üniversitesi Veteriner Fakültesi Dergisi, 24(2), 83-86.

Kumar, S., Pandey, A. K., Abdulrazzaque, W. A., Dwivedi, D. K. (2011). Importance of micro minerals in reproductive performance of livestock. Veterinary World, 4(5): 230.

Küçükaslan, İ. (2011). İz elementler ve ineklerde reprodüktif açıdan önemi. Dicle Üniversitesi Veteriner Fakültesi Dergisi, (1), 26-35.

Lee, S. et al. (2021). Iron deficiency and embryonic development.

Lotthammer, K. H., Ahlswede, L., & Meyer, H. (1976). Studies on a specific, vitamin A-unrelated effect of beta-carotene on the fertility of cattle. I. Further clinical findings and conception rates (Experiment III).

Martin, G., et al. (2022). "Selenium Deficiency and Fertility Issues in Dairy Cattle." Journal of Veterinary Medicine, 66(1), 98-110.

Miller, J. et al. (2022). Magnesium and postpartum reproductive health.

Miller, J. K., Brzezinska-Slebodzinska, E., & Madsen, F. C. (1993). Oxidative stress, antioxidants, and animal function. Journal of Dairy Science.

Sato, F., Watanabe, T., Endo, A. (1982). Cytogenetic Effects of zinc-deficiency on oogenesis and spermatogenesis. Teratology, 26 (1): A13–A14.

Smith RD, Chase LE. Nutrition and Repro duction. Erişim: https://www.wvu.edu/ ~agexten/forglvst/Dairy/dirm14.pdf, tarihi: 10.11.2014. Erişim

Sönmez M. Reprodüksiyon Suni Tohumlama ve Androloji Ders Notları, Elazığ: 2013; p. 155-9.

Succi G, Pialorsi S, Ruffo G. Large doses of vitamin A (subfertile) dairy cows: Spesific effect of beta-carotone. Vet Zootek 1989; 3: 157-69.

Suttle, N. F. (2010). The Mineral Nutrition of Livestock. CABI Publishing.

Şahin N. Vitaminler. Hayvan Besleme ve Beslenme Hastalıkları. Malatya: Medipres, 2008; p. 85-112.

Taneja, S. K., Kaur, R. (1990). Pathology of ovary, uterus, vagina and gonadotrophs of female mice fed on Zn-deficient diet. Indian Journal of Experimental Biology, 28 (11): 1058-1065.

Vallee, B. L., Falchuk, K. H. (1993). The biochemical basis of zinc physiology. Physiological Reviews, 73 (1): 79-118.

Weiss, W. P., & Hogan, J. S. (2005). Effects of vitamin E supplementation on dairy cows. Journal of Dairy Science.

Williams, R., et al. (2020). "Calcium Deficiency and Its Impact on Fertility in Dairy Cows." Journal of Dairy Science, 103(7), 3421-3430.

Yavuz HM. http://www.torsat.com.tr/tarim-12 ineklerde-beslemenin-dol-verimine etkisi.html, Erişim tarihi: 14.12.2015.