Aus dem Dr. Rolf M. Schwiete Zentrum

für Limbusstammzellforschung und kongenitale Aniridie

Universität des Saarlandes, Homburg/Saar

Institutsleiterin: Prof. Dr. Nóra Szentmáry

Charakteristika der kongenitalen Aniridie
Baselinedaten und chirurgische Therapieansätze aus dem Homburger Aniridie-Register

Kumulative Dissertation zur Erlangung des Grades eines Doktors der Medizin

der Medizinischen Fakultät

der UNIVERSITÄT DES SAARLANDES

2023

vorgelegt von

Fabian Fries

geb. am: 18.07.1990 in Ottweiler

Tag der Promotion: 13.05.2024

Dekan: Prof. Dr. Michael D. Menger

1. Berichterstatter: Prof. Dr. Nóra Szentmáry

2. Berichterstatter: Prof. Dr. Bergita Ganse

Inhaltsverzeichnis

Zusammenfassung

1.1 Deutsche Zusammenfassung

Die „kongenitale Aniridie“ ist eine seltene angeborene Erkrankung (1:40.000 bis 1:100.000), die mit schweren Fehlbildungen fast aller Augensegmente einhergeht. In Deutschland wird die Zahl der Betroffenen auf etwa 1000 geschätzt. Charakteristisches und namensgebendes Merkmal der Aniridie ist das teilweise oder vollständige Fehlen der Iris. Regelhaft gehen damit weitere Fehlbildungen des Auges einher, von denen manche bereits unmittelbar nach Geburt (z. B. Makulahypoplasie und Papillenhypoplasie) diagnostiziert werden können, und wiederum andere Veränderungen (z. B. Keratopathie und Glaukom) erst mit zunehmendem Lebensalter zum Vorschein treten. Insbesondere die Aniridie-assoziierte Keratopathie und das Aniridie-assoziierte Sekundärglaukom stellen für die Betroffenen ein großes Risiko der Erblindung im Lauf des Lebens dar. Von besonderer Bedeutung ist auch die angeborene Limbusstammzellinsuffizienz, die mit Wundheilungsstörungen einhergeht und zur Folge hat, dass jede Operation bei Aniridie-Patienten als erschwert und risikobehaftet eingestuft werden muss.

In der ersten Arbeit wurden die Ergebnisse und das Transplantatüberleben nach perforierender Keratoplastik bei Aniridie-assoziierter Keratopathie ausgewertet. Es konnte gezeigt werden, dass die besten Ergebnisse durch eine Kombination von Modifikationen der Routinekeratoplastik erreicht werden. Da die Krankheitsverläufe aber so heterogen und schwierig zu vergleichen waren, entstand hier der Gedanke, die weiteren Auswertungen vor einem größeren Hintergrund, nämlich einem Aniridie-Register, durchzuführen.

Die zweite Studie beschreibt die Auswertung der ersten Daten aus dem Homburger Aniridie-Register. Zum Zeitpunkt der Fertigstellung dieser Arbeit handelt es sich hierbei um die weltweit größte Patientendatenbank kongenitaler Aniridie. Als Hauptrisikofaktoren für eine Erblindung konnten die Aniridie-assoziierte Keratopathie und die bei Aniridie-Patienten häufig auftretende Glaukomerkrankung bestätigt werden. Es konnte gezeigt werden, dass das komplette Fehlen der Iris (klinisch „komplette Aniridie“) einen signifikanten prädiktiven Marker für das Auftreten von Visus-bedrohenden Komplikationen/Verläufen darstellt.

Die dritte Arbeit befasst sich mit Risikofaktoren des Voranschreitens der Aniridie-assoziierten Keratopathie in Bezug auf die Glaukomtherapie. Auch hierfür wurden Daten aus dem Aniridie-Register herangezogen. Es konnte gezeigt werden, dass die Notwendigkeit einer lokalen Ma-

ximaltherapie mit einem fortgeschrittenen Stadium der Aniridie-assoziierten Keratopathie assoziiert ist. Zwischen den einzelnen Substanzklassen gab es keinen Unterschied. Dieser Zusammenhang blieb auch nach Korrektur bezüglich Lebensalter bestehen und wurde zum Anlass genommen, die Pathophysiologie/Kausalität in einem Zellkulturmodell zu untersuchen. Zum Zeitpunkt der Fertigstellung dieser Arbeit stehen die Ergebnisse des Zellkulturmodells noch aus.

Unser Aniridie-Register wird weitere Analysen der Krankheitsverläufe bei Patienten mit kongenitaler Aniridie im Langzeitverlauf ermöglichen. Da Operationen bei Aniridie als sehr risikobehaftet eingestuft werden müssen, erhoffen wir uns hierdurch, weitere prognostische Marker und bestenfalls Interventionsmöglichkeiten vor Operationsnotwendigkeit identifizieren zu können.

Für die *Klinischen Monatsblätter für Augenheilkunde* als Journal für unsere Baselinedaten haben wir uns deshalb entschieden, weil sie sowohl eine Publikation auf Deutsch als auch auf Englisch anbieten. Dies ermöglicht uns, einerseits mehr Zuweiser aus dem deutschsprachigen Raum für unser Aniridiezentrum zu akquirieren, aber auch gleichzeitig unsere Daten der internationalen, englischsprachigen Forschungsgemeinschaft via *Open Access* zur Verfügung zu stellen.

1.2 Abstract

Congenital aniridia is a rare congenital disease (1:40,000 to 1:100,000) that is associated with severe malformations of almost all segments of the eye. In Germany, the number of people affected is estimated at around 1000. The characteristic and name-giving feature of aniridia is the partial or complete absence of the iris. It is usually accompanied by other malformations of the eye, some of which can be diagnosed immediately after birth (e.g. macular hypoplasia and papillary hypoplasia), while other changes (e.g. keratopathy and glaucoma) only become apparent with increasing age. Aniridia-associated keratopathy and aniridia-associated secondary glaucoma in particular pose a major risk of blindness for those affected in the course of their lives. Of particular importance is also the congenital limbal stem cell insufficiency, which is associated with wound healing disorders and means that every operation in aniridia patients must be categorised as difficult and risky.

In the first study, the results and graft survival after penetrating keratoplasty for aniridia-associated keratopathy were analysed. It was shown that the best results are achieved by a combination of modifications of routine keratoplasty. However, as the course of the disease was so heterogeneous and difficult to compare, the idea arose to carry out further analyses against a larger background, namely an aniridia register.

The second paper describes the analysis of the first data from the Homburg Aniridia Register. At the time of writing, this is the world's largest patient database of congenital aniridia. The main risk factors for blindness were confirmed to be aniridia-associated keratopathy and glaucoma, which frequently occurs in aniridia patients. It could be shown that the complete absence of the iris (clinically "complete aniridia") is a significant predictive marker for the occurrence of sight-threatening complications/progressions.

The third study focused on risk factors for the progression of aniridia-associated keratopathy in relation to glaucoma therapy. Data from the aniridia register was also used for this study. It was shown that the need for local maximum therapy is associated with an advanced stage of aniridia-associated keratopathy. There was no difference between the individual substance classes. This correlation remained even after correction for age and was taken as an opportunity to investigate the pathophysiology/causality in a cell culture model. At the time of finalising this paper, the results of the cell culture model are still pending.

Our aniridia registry will enable further analyses of disease progression in patients with congenital aniridia over the long term. As surgery for aniridia must be categorised very risky, we hope to be able to identify further prognostic markers and, at best, intervention options before surgery is necessary.

We decided in favour of the *Klinische Monatsblätter für Augenheilkunde* as the journal for our baseline data because it offers publication in both German and English. This enables us to acquire more referring physicians from German-speaking countries for our aniridia centre, but also to make our data available to the international, English-speaking research community via *Open Access*.

Einleitung und Motivation

Die Aniridie wird als seltene Erkrankung betrachtet mit einer weltweiten Prävalenz zwischen 1 zu 40.000 und 1 zu 100.000 [1]. Trotz ihrer Benennung handelt es sich bei der Aniridie um eine panokuläre Störung, die aufgrund der eindeutigen Iris-Hypoplasie benannt ist, die in den meisten Fällen auftritt (***Abbildung 1***). Diese Charakteristika können von einem beinahe vollständigen Verlust der Iris bis zu einer Vergrößerung und Unregelmäßigkeiten der Pupille reichen, die als Kolobom oder mikroskopische, schlitzartige Randanomalien mit einer Spaltlampe erkennbar sind. Die Auswirkungen auf das Sehvermögen variieren, oft mit angeborenem Sehverlust und nachfolgender pathologischer Sehentwicklung und Nystagmus [2]. Die kongenitale Aniridie unterteilt sich in PAX6-genassoziierte Formen und PAX6-genlose Formen, wobei die ersteren häufiger auftreten [2]. Klinisch manifestiert sich die PAX6-bedingte kongenitale Aniridie in verschiedenen Ausprägungen, einschließlich dominanter Vererbung, sporadischem Auftreten (mit dann dominanter Vererbung) sowie in Verbindung mit Syndromen wie WAGR oder WAGRO. Langzeitkomplikationen wie Glaukom oder Aniridie-assoziierte Keratopathie (AAK) treten bei PAX6-Aniridie stärker ausgeprägt auf [1-3]. Die Bezeichnung "Aniridie-Syndrom" oder "PAX6-Syndrom" wurde vorgeschlagen, da sich gezeigt hat, dass PAX6-Aniridie oft mit systemischen Begleiterkrankungen einhergeht. Die Etablierung eines Aniridie-Zentrums ist entscheidend für verbesserte Behandlungsmöglichkeiten dieser seltenen Krankheit [4].

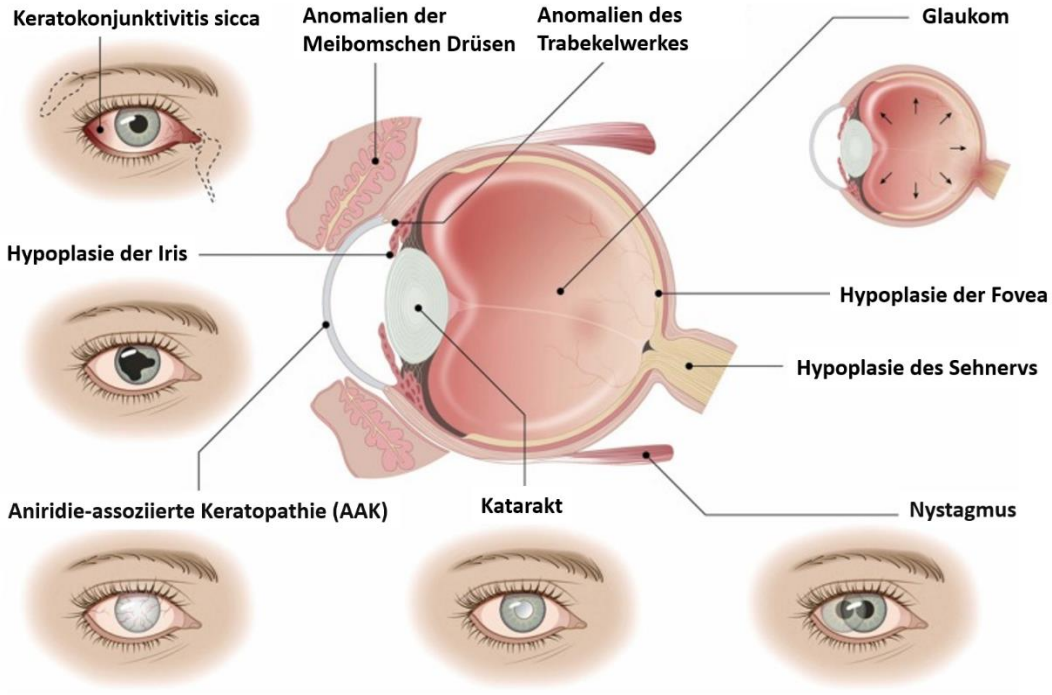


Abbildung 1. *Augenbeteiligung bei kongenitaler Aniridie. Modifiziert auf Basis von [5].*

Die Aniridie repräsentiert eine genetisch bedingte Fehlentwicklung (fast) sämtlicher Augensegmente. Während einige Anomalien im Verlauf des Lebens stabil bleiben, wie beispielsweise die Irisanomalie, nehmen andere einen chronisch-progressiven Verlauf [1-3].

Es existieren vier durch Aniridie verursachte Komplikationen, die zu einem Verlust der Sehschärfe und potenziell zu dauerhaftem Visusverlust führen können:

1. Juvenile Kataraktbildung [6, 7]
2. Sekundäres Glaukom (mit möglicherweise irreversibler Optikusatrophie) [1-4, 8]
3. Limbale Stammzellinsuffizienz mit Bildung von Hornhautnarben [1-4, 9]
4. Aniridie Fibrose Syndrom (AFS): Eine profibrotische, nicht-infektiöse entzündliche Reaktion des Auges auf intraokulare Operationen, die zu fibrokontraktiver Narbenbildung und Netzhautablösung führt [10-12].

Die Veränderungen unter Punkt 1 bis 3 können entweder kongenital vorhanden sein oder sich im Laufe des Lebens entwickeln.

Iris

Die anomale Entwicklung der Iris stellt das charakteristische phänotypische Merkmal bei Aniridie dar [1-4, 8, 9]. Diese kann, abhängig von der Mutation des PAX6-Gens, von einer vollständig fehlenden Iris bis hin zu einer leichten Verschiebung der Pupille (Korektopie) oder einem untypischen Kolobom variieren. Die Irismalformation ist eine der Auslöser für Photophobie.

Katarakt

Katarakte treten bei 50 bis 85 Prozent der Aniridie-Patienten auf. Oftmals zeigt sich bereits kongenital eine Katarakt sowohl am vorderen als auch am hinteren Linsenpol (Cataracta polaris anterior oder posterior), die häufig im Verlauf des Lebens stabil bleibt. Zusätzlich kann es zu einer fortschreitenden Trübung anderer Abschnitte der Linse kommen, sowie zu einer Subluxation der Augenlinse aufgrund von Zonulafaser-Insuffizienz. Beide Zustände können eine Indikation für die Entfernung der Linse und die Implantation einer intraokularen Linse darstellen, um die Sehschärfe zu erhalten [1-9].

Makula und Sehnerv

Aniridie-Patienten zeigen vermehrt eine Hypoplasie des Sehnervs (N. opticus) und der Makula. Die Makulahypoplasie tritt nicht zwangsläufig gleichzeitig mit der Optikushypoplasie auf; sie

kann auch als isoliertes Symptom im Kontext angeborener Sehbehinderungen auftreten. Zusätzlich sind häufig ein horizontaler Nystagmus und Strabismus zu beobachten [1, 3, 4].

Sekundärglaukom

Das Sekundärglaukom bei Aniridie tritt mit einer Inzidenz von 6 bis 75 Prozent, oft sogar vor dem Erwachsenenalter, auf. Aufgrund eines fehldifferenzierten Kammerwinkels entsteht eine Abflussbehinderung durch eine abnormale Positionierung des Schlemm-Kanals oder durch Irisrudimente, die den Kammerwinkel oder das Trabekelmaschenwerk verschließen und somit den Abfluss des Kammerwassers behindern [1-4, 8, 9].

Die Diagnose erfordert regelmäßige Kontrollen des Augeninnendrucks und des Sehnervs. Aufgrund der verdickten Hornhaut ist die Druckmessung häufig erschwert und erfordert eine Messung der Hornhautdicke sowie entsprechende Umrechnungen. Das Sekundärglaukom kann aufgrund der glaukomatösen Optikusatrophie zu einer irreversiblen Beeinträchtigung des Sehvermögens bis zur Erblindung führen und muss daher als potenziell schwerwiegendste Komplikation betrachtet werden (**Abbildung 2**).

Aniridie-assoziiertes Sicca-Syndrom

In Aniridie stellt besonders das Sicca-Syndrom als Vorläufer und Begleiter der Aniridie-assoziierten Keratopathie (AAK) eine erhebliche Herausforderung dar. Die Ursache für die gestörte Tränenfilmproduktion liegt in der PAX6-Mutation. PAX6 spielt eine entscheidende Rolle in der embryonalen Entwicklung des Auges, speziell in der Entwicklung des Oberflächenektoderms. Da die Meibom-Drüsen aus dem Ektoderm entstehen, liegt eine genetische Ursache für die Dysfunktion der Meibomschen Drüsen nahe [1-9].

Aniridie-assoziierte Keratopathie (AAK)

Bei bis zu 70 Prozent der von PAX6-Aniridie Betroffenen tritt im Laufe des Lebens eine Aniridie-assoziierte Keratopathie (AAK) auf, die durch die Kombination mehrerer Faktoren verursacht sein kann. Dazu zählen unter anderem eine ausgeprägte Sicca-Problematik, eine Limbusstammzellinsuffizienz mit gestörter Differenzierung der Hornhautepithelzellen sowie abnormale Zelladhäsion und Wundheilungsstörung an der Oberfläche der Hornhaut. Die Auswirkungen für den Patienten sind eine anhaltende Instabilität des Tränenfilms, wiederholte äußerst schmerzhafte Hornhauterosionen und eine Verschlechterung des Sehvermögens durch die

Bildung von vaskularisierten Narben (Pannus) [9]. Am Limbus befinden sich in den sogenannten Palisaden von Vogt die epithelialen Stammzellen, die für die Regeneration des Epithels, die korneale Homöostase und die Aufrechterhaltung der Grenze zwischen Hornhaut und Bindehaut verantwortlich sind [1-4, 9].

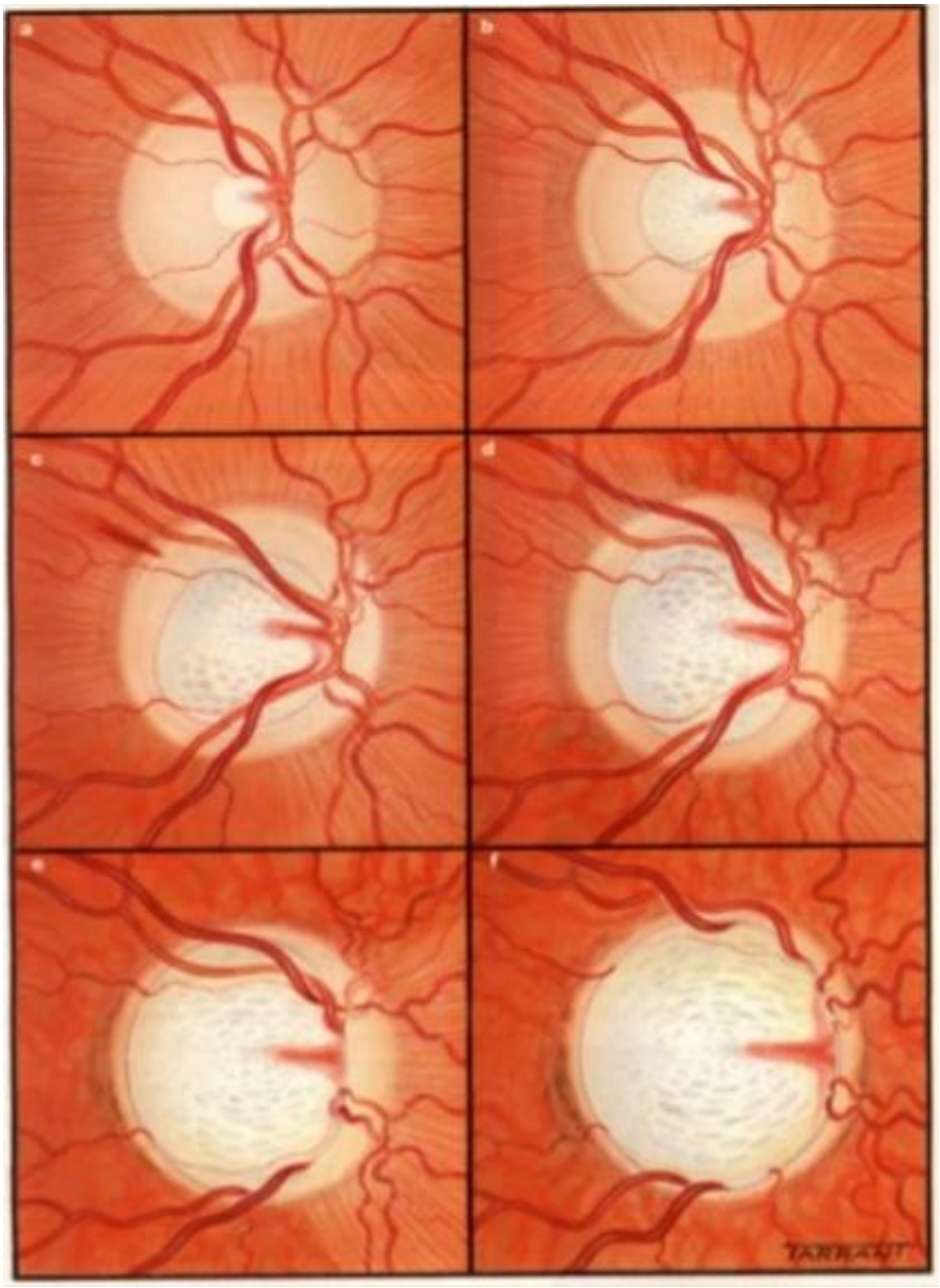


Abbildung 2. *Zunehmende Optikusatrophie durch Glaukom.*

Durch den kongenital anatomisch verändert-angelegten Schlemm-Kanal kommt es aufgrund von reduziertem Kammerwasserabfluss zu einem erhöhten Augeninnendruck. Die damit bedingte Reduktion der Perfusion führt zu einem kontinuierlichen Absterben von Sehnervenzellen. Funduskopisch lässt sich das an einer zunehmenden Exkavation und Abblassung (a → b → c → d → e → f) des Sehnervs beobachten. Bildquelle: https://www.allentownvision.com/glaucoma

Die Aniridie-assoziierte Keratopathie (AAK) hat ihre Ursache unter anderem in der Limbusstammzellinsuffizienz (LSZI), die durch den PAX6-Gendefekt verursacht wird [1-3]. Dies äußert sich durch eine sich zentripetal ausbreitende Vaskularisierung, Konjunktivalisierung und Dickenzunahme der Hornhaut. Die AAK ist ein fortschreitender Zustand, gekennzeichnet durch Entzündung, Vaskularisierung, gestörte Wundheilung, Schmerzen, Sehschärfebeeinträchtigung und Infiltration von konjunktivalen Becherzellen. Der natürliche Verlauf der AAK zeigt mehrere Progressionsstufen (***Abbildung 3***).

In der Jugend treten Anzeichen einer Keratopathie oft mit Dickenzunahme des peripheren Hornhautepithels ohne funktionelle Manifestation auf. Im zweiten Lebensjahrzehnt zeigen die Patienten chronische Irritationen und eine dünne, oberflächliche Vaskularisation in der peripheren Hornhaut, die kontinuierlich in die zentrale Hornhaut voranschreitet. Häufig sind Schmerzen, Lichtempfindlichkeit und wiederholte korneale epitheliale Erosionen. In fortgeschrittenen Stadien schreitet die Keratopathie voran, bis die gesamte Hornhaut durch Pannusbildung und vaskularisierte Narben betroffen ist, begleitet von einer starken Zunahme der zentralen Hornhautdicke [1-4, 9].

Die AAK ist als eine kombinierte Erkrankung aus Limbusstammzellversagen und dem defekten antiangiogenen Privileg der Hornhaut, wie in ***Abbildung 3*** dargestellt, einzustufen. Unterschiedliche Stadien von AAK mit Transparenzverlust der Hornhaut sind mit einer pathologischen Transgression von Gefäßen über den Limbus und zentripetal in die Hornhaut verbunden, nachweisbar durch die Angiographie des vorderen Augenabschnitts [4].

Aufgrund der Seltenheit der kongenitalen Aniridie gestaltet sich die Etablierung klinischer prognostischer Parameter und Behandlungsstandards als schwierig. Zur Entwicklung effektiverer Behandlungsmöglichkeiten ist die Einrichtung eines Aniridie-Registers unerlässlich. Unser Aniridiezentrum wird detaillierte Längsschnittanalysen von Augen- und Systemerkrankungen bei Patienten mit kongenitaler Aniridie während langfristiger Nachbeobachtungen durchführen.

Unsere Forschungen führen von der speziellen Fragestellung zur Optimierung der perforierenden Keratoplastik bei kongenitaler Aniridie, über die Untersuchung der Entstehung der AAK und möglicher prädiktiver Marker in einer zweiten Veröffentlichung, bis hin zur Erforschung von Risikofaktoren der AAK. Diese bilden die Grundlage für zukünftige geplante Zellkulturversuche.

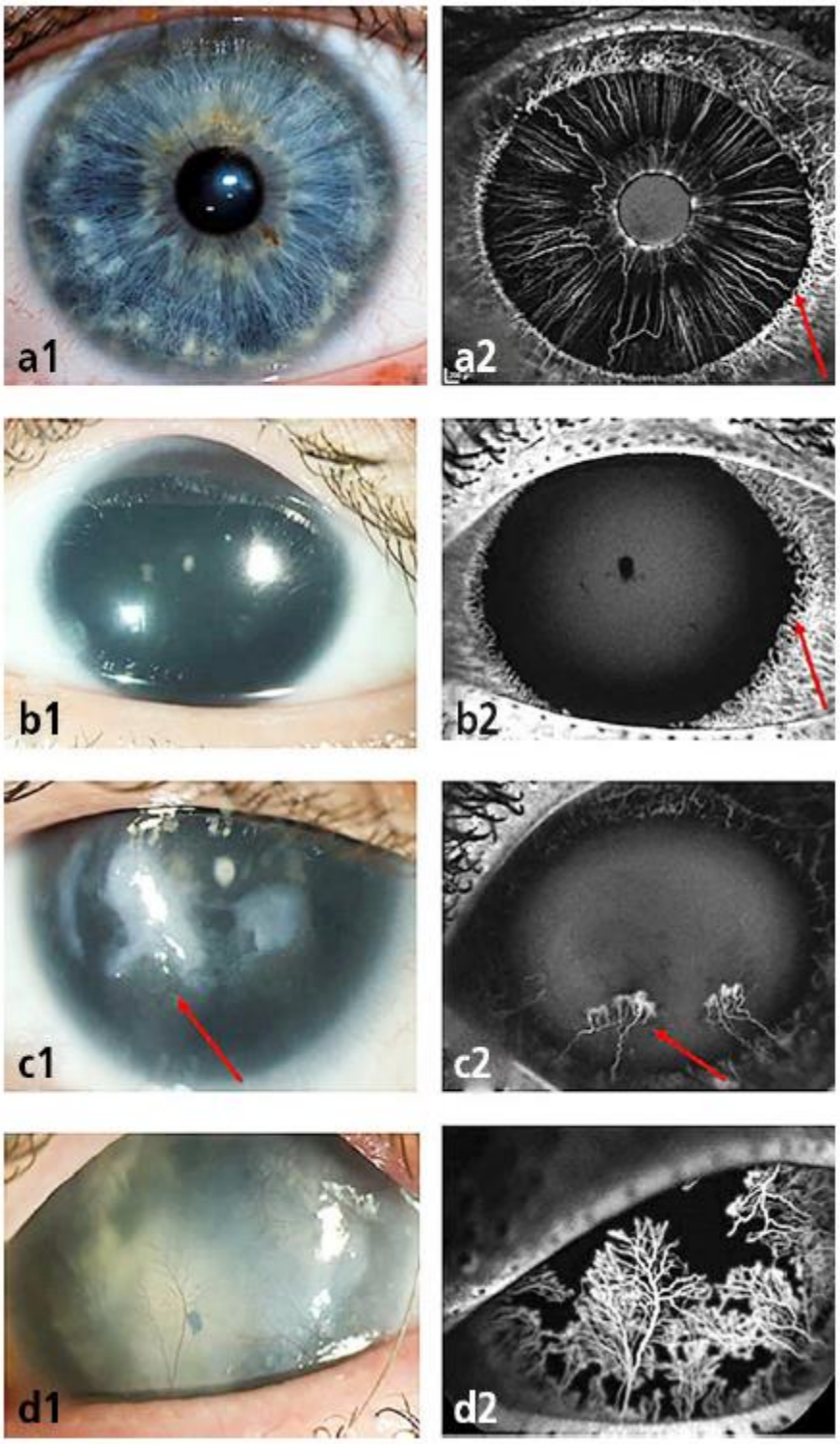


Abbildung 3. *Vorderabschnittsangiographien bei Aniridie-assoziierter Keratopathie.*

Während in a1 ein Normalbefund zu sehen ist, zeigt a2 die korrespondierende Angiographie. Von b über c zu d sieht man zunehmende Stadien der Aniridie-assoziierten Keratopathie. Der Limbus wird sukzessive von den Gefäßen überschritten. Sie breiten sich zentripetal aus bis sie schließlich auch das Zentrum bedecken. Eigene Aufnahme, bereits publiziert in [13].

Optimierungsversuch des Outcomes bei perforierender Keratoplastik in Fällen von kongenitaler Aniridie-assoziierter Keratopathie (AAK) (Veröffentlichung 1) [14]

In dieser Studie untersuchten wir die Verbesserungsmöglichkeiten sowohl mikrochirurgischer als auch konservativer Verfahren, um das Risiko von Komplikationen nach einer perforierenden Keratoplastik (PKP) bei Patienten mit Aniridie-assoziierter Keratopathie (AAK) zu minimieren. Unsere Analyse basierte auf einer retrospektiven Betrachtung von 25 PKP-Fällen bei 16 Patienten mit AAK. Die präoperativen Indikationen umfassten endotheliale Dekompensation und vaskularisierte Narben (68 %) sowie Transplantatversagen (32 %), verursacht durch Limbusstammzellinsuffizienz. Unser optimierter Ansatz beinhaltete die Verwendung kleiner Hornhauttransplantate (ca. 7,0 mm Durchmesser), 10-0-Nylon-Einzelknopfnähte, simultane Amnionmembrantransplantation als Abdeckung, Anwendung großer Verbandskontaktlinsen, temporäre laterale Tarsorrhaphie, postoperative autologe Serum-Augentropfen und systemische Immunsuppression. Die Hauptzielgrößen unserer Studie umfassten die Sehschärfe, die Überlebensrate der Transplantate und aufgetretene Komplikationen während einer durchschnittlichen Nachbeobachtungszeit von 107 Wochen. Die PKP bei kongenitaler Aniridie stellt eine Hochrisiko-Keratoplastik dar. Unser optimierter therapeutischer Ansatz zeigte vielversprechende Ergebnisse, um die postoperative Komplikationsrate bei diesen anspruchsvollen Fällen zu reduzieren.

Trotz umfassender Vor- und Nachbehandlung zeigten die Ergebnisse signifikante Verschlechterungen im Vergleich zu Standardkeratoplastiken. Dies führte zu einer Fokussierung auf frühere Stadien der Keratopathie, um potenzielle Ansätze zur Verlangsamung des Krankheitsfortschritts zu erforschen und die PKP als ultimative Behandlungsoption zu bewahren.

Querschnittsanalyse von 556 Augen aus dem Homburger Aniridiezentrum (Veröffentlichung 2) [15]

In unserer retrospektiven, monozentrischen Studie wurden Patienten einbezogen, die sich zwischen Juni 2003 und Januar 2022 einer umfangreichen augenärztlichen Untersuchung durch die Leitung der Homburger KiOLoN-Abteilung (Kinderophthalmologie, Orthoptik, Low Vision und Neuroophthalmologie) unterzogen. Berücksichtigt wurden Daten des ersten Untersuchungszeitpunkts. Die Studie umfasste 556 Augen von 286 Teilnehmern (Durchschnittsalter: 20,1 Jahre; 45,5% männlich). Bei 518 Augen (93,7%) wurde Nystagmus diagnostiziert, während 327 Augen (58,8%) Strabismus aufwiesen. Von den untersuchten Augen hatten 436 (78,4%) eine altersentsprechende Achsenlänge, 104 (18,7%) zeigten Mikrophthalmus und 13 (2,3%) Buphthalmus. Irisfehlbildungen traten in 34 Augen (6,1%) als atypisches Kolobom, in 61 Augen (10,9%) als Irisreste über mehr als 6 Uhrzeiten, in 96 Augen (17,2%) als Irisreste unter 6 Uhrzeiten und in 320 Augen (57,5%) als vollständige Aniridie auf. Die Einteilung der Patienten in die verschiedenen Stadien der Aniridie-assoziierten Keratopathie (AAK) ergab: Stadium 0 (96 Augen [17,2%], keine Keratopathie), Stadium 1 (178 Augen [32,0%]), Stadium 2 (107 Augen [19,2%]), Stadium 3 (67 Augen [12,0%]), Stadium 4 (62 Augen [11,1%]) und Stadium 5 (45 Augen [8,0%]). Sekundäres Glaukom wurde bei 307 (55,5%) Augen festgestellt, Makulahypoplasie bei 395 (71,4%) und angeborene Pathologien des Sehnervenkopfes bei 223 (40,3%) Augen. Ein signifikanter Zusammenhang bestand zwischen dem Typ der Irisfehlbildung und dem AAK-Stadium, Linseneigenschaften, Glaukompräsenz sowie angeborenen Makula- und Sehnervenkopf-Pathologien ($p < 0,001$ für alle), wobei vollständige Aniridie mit den meisten Komplikationen assoziiert war. Im Homburger Aniridiezentrum waren AAK, Irisfehlbildung, Katarakt und Makulahypoplasie die vorherrschenden ophthalmologischen Merkmale bei kongenitaler Aniridie. Der Irisfehlbildungstyp könnte auf die zukünftige Entwicklung von AAK, Katarakt und Glaukom hinweisen und korreliert mit angeborenen Pathologien des Sehnervenkopfes und der Makula.

Diese Ergebnisse motivierten uns, weitere Subgruppenanalysen durchzuführen, um Risikofaktoren für die Entwicklung der Aniridie-assoziierten Keratopathie zu identifizieren. Eine dieser Subgruppen wird im folgenden Abschnitt detaillierter beschrieben.

Die Auswirkung der Glaukombehandlung auf die Aniridie-assoziierte Keratopathie (AAK) – Ein Bericht aus dem Homburger Register für kongenitale Aniridie (Veröffentlichung 3) [16]

Diese Studie analysierte dieselbe Patientenkohorte wie in Veröffentlichung 2, wobei jedoch eine Unterteilung in neue Subgruppen erfolgte, um eine andere Fragestellung zu adressieren. Es wurden insgesamt 556 Augen von 286 Probanden (Durchschnittsalter: 20,1 Jahre; 45,5% männlich) einbezogen. Bei 307 dieser Augen (55,2%) von 163 Patienten (Durchschnittsalter: 27,5 Jahre; 43,1% männlich) wurde ein Glaukom diagnostiziert. Der durchschnittliche Augeninnendruck in der Glaukomgruppe betrug 19,0 mmHg (±8,0), verglichen mit 14,1 mmHg (±3,6) in der Nicht-Glaukom-Gruppe ($p < 0,001$). Unter den Glaukompatienten befanden sich 68 Personen mit einer Monotherapie, 51 mit einer Zweifachtherapie, 41 mit einer Dreifachtherapie, 7 mit einer Vierfachtherapie und 140 ohne lokale Therapie (z. B. nach drucksenkender Operation, bei schmerzfreiem Endstadium des Glaukoms oder bei Noncompliance).

Die Patienten wurden gemäß den AAK-Stadien klassifiziert: Stadium 0 (96 Augen, 17,2%, keine Keratopathie), Stadium 1 (178 Augen, 32,0%), Stadium 2 (107 Augen, 19,2%), Stadium 3 (67 Augen, 12,0%), Stadium 4 (62 Augen, 11,1%) und Stadium 5 (45 Augen, 8,0%). Das durchschnittliche AAK-Stadium variierte je nach Therapie: 1,4 in der Gruppe ohne Augentropfen, 1,9 in der Monotherapie-Gruppe, 1,8 in der Zweifachtherapie-Gruppe, 1,9 in der Dreifachtherapie-Gruppe, 3,4 in der Vierfachtherapie-Gruppe und 3,3 nach drucksenkender Operation. Es zeigte sich eine signifikante positive Korrelation zwischen dem AAK-Stadium und der Anzahl der drucksenkenden Medikamente ($p < 0,05$) sowie einer vorangegangenen drucksenkenden Operation ($p < 0,05$). Interessanterweise war die Verwendung von Prostaglandin-Analoga, trotz bekannter proinflammatorischer Nebenwirkungen, nicht signifikant mit einem höheren AAK-Stadium korreliert.

In unserem Aniridiezentrum wiesen Patienten, die eine lokale antiglaukomatöse Vierfachtherapie erhielten oder eine antiglaukomatöse Operation hatten, deutlich höhere AAK-Stadien auf. Die Art der verwendeten Wirkstoffe zeigte keinen Einfluss auf das AAK-Stadium. Zudem wurde eine Zellkulturuntersuchung zur Erforschung der Pathophysiologie und Kausalität von der Ethikkommission der Ärztekammer des Saarlandes genehmigt und eingeleitet. Die Ergebnisse dieser Studie stehen noch aus. Unser Register wird zukünftig eine detaillierte Analyse der ophthalmischen und systemischen Erkrankungen bei Patienten mit kongenitaler Aniridie im Langzeitverlauf ermöglichen.

Veröffentlichung 1

Int Ophthalmol (2021) 41:4091–4098
https://doi.org/10.1007/s10792-021-01982-z

ORIGINAL PAPER

An attempt to optimize the outcome of penetrating keratoplasty in congenital aniridia-associated keratopathy (AAK)

C. J. Farah · F. N. Fries · L. Latta · B. Käsmann-Kellner · B. Seitz

Received: 9 February 2021 / Accepted: 16 July 2021 / Published online: 29 July 2021

Abstract
Purpose To propose an optimized microsurgical and medical approach to reduce the risk of complications after penetrating keratoplasty (PKP) in patients with aniridia-associated keratopathy (AAK).
Methods Retrospective observational case series of 25 PKP performed in 16 patients with AAK. Preoperative indications were endothelial decompensation and vascularized scars (68%) or graft failure (32%) due to limbal stem cell deficiency. The optimized approach included a combination of a small corneal graft size (around 7.0 mm), interrupted 10–0nylon sutures, simultaneous AMT as a patch, large bandage contact lens, temporary lateral tarsorrhaphy, postoperative autologous serum eye drops, and systemic immunosuppression. Main outcome measures included: visual acuity, transplant survival, and complications encountered during follow-up of 107 weeks on average.
Results A complete modified keratoplasty scheme was used in 10 of 25 PKP (group 1), while at least one of the modifications was missing in the other 15 PKP (group 2). After 8 weeks of follow-up, the epithelium was closed in 23 eyes. Visual acuity improved in 19 eyes at 6 months of follow-up, and remained stable in six eyes. None of the eyes showed a decrease in visual acuity. At the last post-operative follow-up, this visual improvement persisted in 14 eyes and graft survival rate after 156 weeks (3 years) was 69% in group 1 versus 44% in group 2 ($p = 0.39$, log-rank test). Secondary corneal neovascularization (8%), scarring (4%), ulcer (4%), or graft rejection (8%) happened mostly in the second group which was missing at least one of the suggested modifications.
Conclusions PKP in congenital aniridia must be considered as a high-risk keratoplasty. An optimized therapeutic approach seems to be promising in order to reduce the postoperative complication rate in these most difficult eyes.

Keywords Aniridia-associated keratopathy · Penetrating keratoplasty · Limbal stem cell deficiency · Amnion membrane transplantation · Autologous serum

C. J. Farah (✉) · F. N. Fries · B. Käsmann-Kellner · B. Seitz
Department of Ophthalmology, Saarland University Medical Center, 100 Kirrbergerstr., Building 22, 66421 Homburg, Saar, Germany
e-mail: farah.chr.j@gmail.com

L. Latta
Dr. Rolf M. Schwiete Center for Limbal Stem Cell Research and Congenital Aniridia, Saarland University Medical Center, Homburg, Saar, Germany

Introduction

First described as "congenital irideremia" in the nineteenth century, congenital aniridia is a rare (1:60.000–1:90.000) pan-ocular disease that can be differentiated in two major categories depending on the presence of a PAX-6-Gene mutation [1–4]. Among those with a mutation of the PAX-6-Gene, frequent mutations involve point mutations and deletions. A deletion of the short arm of chromosome 11(p13) may be autosomal dominant or sporadic, and to a lesser extent autosomal recessive, e.g., in the Gillespie syndrome [5]. A PAX-6-Gene mutation is more frequently associated with ocular complications compared to aniridia triggered by other mutations than the PAX-6-Gene [2, 3]. Congenital aniridia is linked to different malformations such as iris, macular or optic nerve hypoplasia, but also to deteriorating progressive major ocular dysfunctions such as limbal stem cell deficiency, premature onset of cataract, and secondary glaucoma that can lead to blindness throughout life [2, 3, 6–8]. The incidence and severity of aniridia-associated keratopathy (AAK) increases with age affecting about 20–30% of those patients and leads to corneal opacities, scarring, and vascularization due to a unique form of limbal stem cell deficiency [2, 9–11].

With time, it may result in corneal ulcers, dense vascularized scars, or endothelial decompensation (especially after complicated cataract and/or glaucoma surgery), where a penetrating keratoplasty becomes indicated [8]. Persisting epithelial defects, suture loosening, and an increased risk of graft rejection are typical postoperative complications in those high-risk keratoplasties [8]. We hypothesized that the combination of a small corneal graft size, interrupted sutures, simultaneous amniotic membrane transplantation (AMT) as a patch [12, 13], large bandage contact lens, temporary lateral tarsorrhaphy, postoperative autologous serum eye drops [14], and systemic immunosuppression [15, 16] may improve the outcome after PKP in congenital aniridia.

Patients and methods

This study is a retrospective observational case series of 25 penetrating keratoplasties (PKP) performed in 20 eyes of 16 patients with AAK at the Department of Ophthalmology of the Saarland University Medical Center in Germany between 2012 and 2019. Four patients received a bilateral surgery. All procedures were performed in accordance with the ethical standards of the 1964 Helsinki Declaration and its later amendments. Transplantation had been proposed as a last resort therapy in corneas with severe endothelial decompensation and stromal scars (68%) and graft failure (32%), due to limbal stem cell deficiency with no success of conservative treatments (stage IV or V according to Yazdanpanah et al. [17]).

The mean age during keratoplasty was 52 ± 8 (from 26 to 64) years, and most eyes (92%) had a history of previous surgeries (Table 1). We defined a small graft as ranging from 6.0 to 7.5 mm diameter. Two types of 10–0 nylon sutures were used as follows: double-running cross-stitch sutures according to Hoffmann versus 24–32 interrupted sutures (Fig. 1). Amniotic membranes were collected from healthy women with their consent and properly processed in the eye bank before transplantation [18]. They were transplanted at the end of the keratoplasty as a single 16-mm layer membrane with the stromal side facing the corneal graft and fixed with a running 10–0 nylon episcleral suture as a patch [12]. The membrane was covered with a large 17-mm bandage contact lens, and both sutures and contact lens were removed after a period of 4–6 weeks. Temporary lateral tarsorrhaphy was performed using 5–0 silk and left typically for 4–6 weeks after PKP.

After exclusion of systemic infectious diseases, 100%-concentrated autologous serum eye drops were prepared [19, 20]. During the first postoperative days, they were applied hourly alternating with preservative-free hyaluronic acid containing artificial tear eye drops. A topical antibiotic coverage for at least 4 weeks with ofloxacin or moxifloxacin eyedrops five times daily was necessary to prevent infections until complete epithelial healing was achieved, and the bandage contact lens was removed, typically after 4 weeks. An additional long-term therapy with topical acyclovir (five times daily) and systemic acyclovir (starting with 400 mg five times daily for 6 weeks, then two times daily for 1 year) was given to patients with history of herpetic keratitis.

Postoperative topical corticosteroids (starting at five times daily, being reduced by 1 drop every 6–8 weeks) and systemic corticosteroid (prednisolone, prednisolonacetat, and methylprednisolone) were slowly tapered over 4 weeks, starting at 100 mg

Table 1 Characteristics of 16 patients with congenital aniridia (25 keratoplasties)

Patients	*n* = 16 (100%)
Males	6 (37,5%)
Females	10 (62,5%)
Genetics	*n* = 16 (100%)
PAX-6	7 (43,75%)
WAGR(O)	1 (6,25%)
Genetically not analyzed	8 (50%)
Type of Keratoplasty	*n* = 25 (100%)
First keratoplasty (with or without simultaneous pannus removal)	13 (52%)
Repeat keratoplasty (with or without simultaneous pannus removal)	5 (20%)
Classical triple procedure (combined with cataract surgery and intraocular lens implantation)	4 (16%)
Pole-to-pole surgery (keratoplasty combined with lens and vitreoretinal surgery)	2 (8%)
HLA-typed keratoplasty	1 (4%)
History of previous surgeries before keratoplasty (Multiple choices possible)	*n* = 25 (100%)
Glaucoma surgery	11 (44%)
Trabeculotomy	3 (12%)
Cyclophotocoagulation	9 (36%)
Ahmed valve	5 (20%)
Corneal surgery	12 (48%)
Pannus removal (“pannectomy”)	3 (12%)
Phototherapeutic keratectomy (PTK)	1 (4%)
Penetrating keratoplasty (in domo)	5 (20%)
Penetrating keratoplasty (ex domo)	3 (12%)
Cataract surgery	8 (32%)
Retinal surgery	4 (24%)
None	2 (8%)

daily and being reduced by 20 mg every second day until 20 mg daily, then being reduced slowly. A systematic preoperative evaluation for long-term use of immunosuppression with mycophenolate mofetil or cyclosporin A and systemic follow-up were established in collaboration with the family physician, and doses were adapted to the general condition of the patient.

Transplantations were combined with simultaneous cataract or vitreoretinal surgeries where indicated. Characteristics of recipients are shown in Table 1. The optimized approach included a combination of a small corneal graft size (around 7.0 mm), interrupted 10–0 nylon sutures, simultaneous AMT as a patch, large bandage contact lens, temporary lateral tarsorrhaphy, postoperative autologous serum eye drops, and systemic immunosuppression. Main outcome measures included: visual acuity, transplant survival, and complications encountered during the follow-up of 107 weeks on average.

For statistical analysis, eyes were separated into two groups based on the therapeutic scheme. A complete, modified keratoplasty scheme was used in 10 of 25 PKP (group 1). The second group included all other PKP, where at least one modification of the modified keratoplasty scheme was missing (group 2). Techniques used are illustrated in Table 2.

Statistical analysis was performed using SPSS v. 20.0.0 (IBM Corp., Armonk, NY, USA). Visual acuity was recorded in decimal and converted into logMAR before analysis. Graft survival was analyzed using the Kaplan–Meier method and log-rank test, which is appropriate to compare small sample sizes and unequal censoring groups.

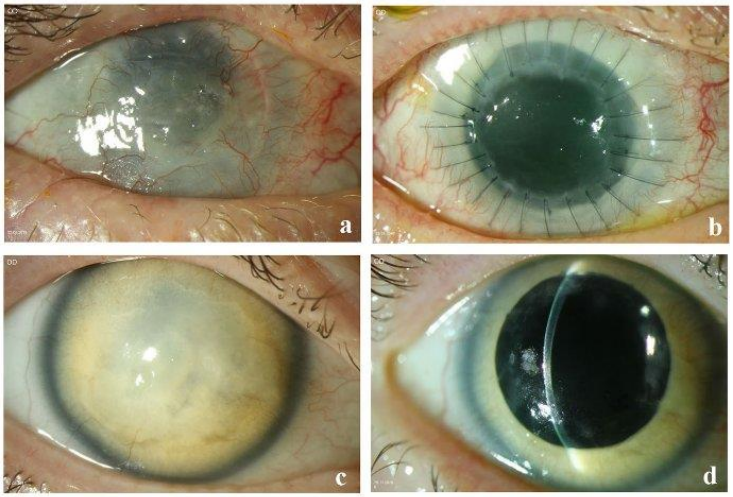


Fig. 1 **a** Patient 1: Preoperative picture with decompensated and severely vascularized graft, 6 years after penetrating keratoplasty. Visual acuity: Hand motion (logMar 2.7). **b** Patient 1: 8 months postoperative picture with 26 interrupted sutures after repeat penetrating keratoplasty with Barron trephine (7.00/7.25 mm). Visual acuity has reached logMar 2.3. **c** Patient 2: 64-year old woman with corneal decompensation, vascularized cornea, secondary amyloidosis, and premature cataract in congenital aniridia. Visual acuity: Hand motion (logMAR 2.7). **d** Patient 2: Clear corneal graft, 18 months after classical excimer laser-assisted triple procedure (7.0/7.1 mm) and suture removal. Visual acuity improved to logMAR 1.0

Table 2 Techniques used in 25 keratoplasties (Multiple choices possible)

Diameter of recipient openings (diameter in mm)	*n* = 25 (100%)
6.5 mm	1 (4%)
7.0 mm	14 (56%)
7.5 mm	9 (36%)
12.0 mm (Limbo-keratoplasty)	1 (4%)
Sutures	*n* = 25 (100%)
Interrupted sutures	18 (72%)
Double-running cross-stitch sutures according to Hoffmann (1976)	7 (28%)
Use of simultaneous amnion membrane transplantation (Patch)	*n* = 25 (100%)
Yes	24 (96%)
No	1 (4%)
Use of simultaneous lateral tarsorrhaphy	*n* = 25 (100%)
Yes	14 (56%)
No	11 (44%)
Use of postoperative autologous serum eye drops primarily	*n* = 25 (100%)
Yes	19 (76%)
No	6 (24%)
Use of long-term immunosuppressive therapy	*n* = 25 (100%)
Mycophenolate mofetil	20 (80%)
Cyclosporin-A	3 (12%)
None	2 (8%)

Results

Visual acuity

At 6 months of follow-up, the mean visual acuity (VA) improved from logMAR 2.18 to logMAR 1.65. None of the eyes showed a decreased VA. On the other hand, the VA remained unchanged in six eyes and improved in 19 eyes. The last examination was made on average 107 weeks postoperatively with a mean VA of logMAR 1.69 in group 1 and logMAR 1.77 in group 2.

All patients reported a subjective improvement in their visual acuity, either by increased clarity of the image, or by decreased visual discomfort caused by corneal optical phenomena such as glare. However, no specific questionnaires have been used to standardize subjective assessment of patients. This visual improvement persisted in 14 eyes until the end of follow-up (107 weeks on average).

Graft survival

At 8 weeks of follow-up, the epithelium was closed in 23 eyes and only 2 eyes needed a second AMT. Graft survival was referred to as a clear and transparent graft, without endothelial decompensation scars or corneal opacities. Graft survival is demonstrated in a Kaplan–Meier chart (Fig. 2). The mean postoperative follow-up was 119 weeks in group 1 and 216 weeks in group 2. Graft survival rate after 156 weeks (3 years) was 69% in group 1 and 44% in group 2 ($p = 0.39$, log-rank test). The median graft survival time was 97 weeks in group 1 and 81 weeks in group 2.

Adverse events

While only two eyes needed a repeat keratoplasty in group 1 and 3 eyes in group 2, severe corneal complications such as graft rejection (8%), anterior segment fibrosis syndrome (4%) or graft ulcer (16%), mostly occurred in group 2. Other, less severe corneal complications such as persistent epithelial defects (20%) and premature suture removal/replacement (28%) were present in both groups (Table 3).

A second AMT as patch was used to treat persistent epithelial defects in five eyes. Loose corneal sutures were removed as quickly as possible to prevent infiltrates and infections. Graft rejections were primarily treated with topical intracameral and systemic steroids, followed by a repeat keratoplasty, if needed. The two eyes that required a repeat keratoplasty after graft rejection had contraindications to systemic immunosuppressive therapy.

Extracorneal complications, regardless of the chosen therapeutic scheme, were changes in ocular pressure (hypo- and hypertension) (20%), retinal detachment (4%), retinal vein occlusion (4%), and intraocular lens luxation (4%). Those complications were handled according to the respective German guidelines.

Discussion

The development of microsurgical techniques and the knowledge of limbal stem cell function has led to a considerable improvement in the treatment and visual prognosis of eyes with congenital aniridia over the years [3–9, 21–23]. Each small improvement is helpful for those progressively visually impaired patients. The global therapeutic approach should always consider a high risk of concomitant glaucomatous damage with irreversible optic atrophy due to a mispositioning of the ciliary body processes toward the iris stump and a very short ciliary body [2, 7, 24].

In well-selected cases, the treatment of AAK with PKP has shown to be beneficial, even though the postoperative complications turned out to be more frequent in those high-risk eyes [6, 8, 25]. Our modified treatment scheme appears to reduce the severe postoperative complications and improve visual prognosis as well as graft survival in the mid-term follow-up.

At first glance, the Kaplan–Meier curve may suggest a different trend between the groups, but the log-rank test could not detect a statistically significant difference between the two groups in terms of graft survival ($p = 0.39$). We attribute this to the low number of cases, which is due to the rarity of the disease. Based on our clinical experience, we refrain from randomizing patients as we do not want to deprive them of what we believe to be the optimal treatment nowadays.

We explain this difference by the following preventive measures: the use of interrupted sutures allows a quick removal of loose sutures at the slit lamp without risking graft slippage, and thus reduces the risk of infections and secondary immune reactions

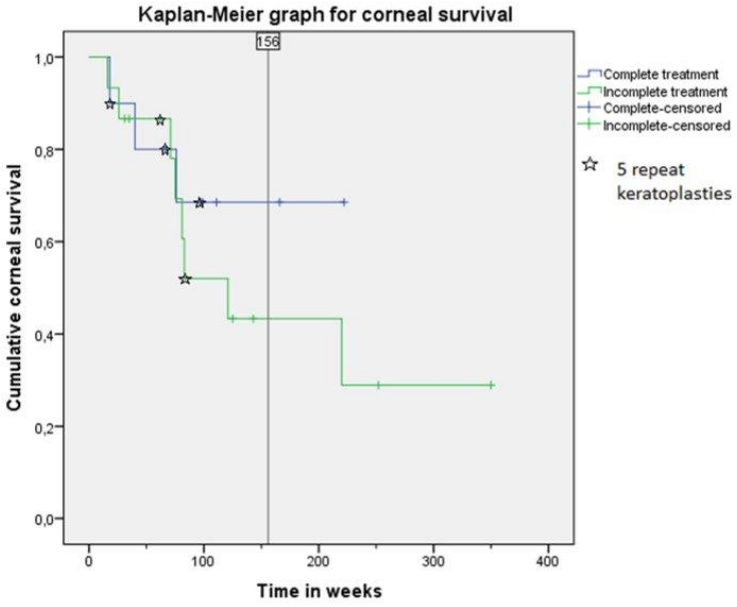


Fig. 2 Survival of corneal grafts in group 1 (complete treatment) versus group 2 (incomplete treatment). Graft survival rate after 156 weeks (3 years) was 69% in group 1 and 44% in group 2, (p = 0.39, logrank test)

Table 3 Corneal complications during follow-up. n = 25 keratoplasties (100%). Group 1: Complete therapeutic modified scheme (n = 10). Group 2: Incomplete therapeutic scheme (n = 15)

Persistent epithelial defect	n = 5 (20%)
Group 1	3 (12%)
Group 2	2 (8%)
Premature suture removal/replacement	n = 7 (28%)
Group 1	3 (12%)
Group 2	4 (16%)
Corneal endothelial decompensation	n = 4 (16%)
Group 1	2 (8%)
Group 2	2 (8%)
Immunological graft rejection	n = 2 (8%)
Group 1	0 (0%)
Group 2	2 (8%)
Graft ulcer/neovascularization/scarring/anterior segment fibrosis syndrome	n = 5 (20%)
Group 1	1 (4%)
Group 2	4 (16%)

[27]. A temporary lateral tarsorrhaphy combined with simultaneous AMT as a patch and 17-mm bandage contact lens considerably reduces postoperative epithelial defects due to a mechanical protective effect [12, 28, 29].

Furthermore, the amnion membrane supports epithelialization, it has anti-fibrotic effects (due to a reduced expression of TGF β1, β2, β3 isoforms and TGF-beta receptor II), and anti-inflammatory effects (through inhibition of proinflammatory cytokines). Moreover, anti-angiogenic effects (due to production of thrombospondin-1, endostatin and tissue inhibitors of metalloproteases), and immunomodulatory effects have been reported [12, 13, 18, 26]. Similar positive effects have been described for the autologous serum eye drops [13, 18, 25]. Due to the higher risk of graft

rejection, a systemic immunosuppressive therapy proved to be useful in most patients [15, 16].

The limitations of the study are the small sample size due to the rare condition and the lack of long-term observation periods to demonstrate a significant difference between groups. A prospective randomized study is not possible for ethical reasons. As our proposed therapeutic scheme does not directly address the cause of AAK, it could be combined with a prior transplantation of allogenic limbal stem cells, even though limbal stem cell transplantation alone in patient with AAK has been significantly associated with progression of limbal stem cell deficiency severity and visual impairment [30]. However, the survival curve looks promising, with a major separation after only 2 years of follow-up. We may expect an average graft survival of 50% at 5 years for the group which was treated with the complete scheme, and only 30% for the other group (Fig. 2). This highlights the difficulty of surgical management in AAK compared to other corneal pathologies with limbal stem cell deficiency such as herpes keratitis (survival rate of 90% after 2 years and 49% after 10 years), while chemical burn-associated limbal stem cell deficiency could best benefit from our modified scheme to enhance graft survival (survival rate of 35% after 5 years and 14% after 10 years) [31, 32]. Moreover, the four patients that received a bilateral surgery showed a better graft survival in the eyes who benefited from the optimized treatment scheme. This may also speak in favor of the efficacy of our modified scheme.

Although the Boston keratoprosthesis shows promising results in small series of patients with AAK, other complications such as retroprosthetic membrane formation or device extrusion exists [6, 33]. Regarding the limbo-keratoplasty, Lang et al. reported a median graft survival of 3.2 years over a long-term observation, indicating that this might also be a favour of approach in "partial" limbal stem cell deficiency [34].

In conclusion, an optimized high-risk PKP approach seems to be promising to reduce the postoperative complication rate in these most difficult eyes with congenital aniridia. Furthermore, an extension to treat other high-risk corneal pathologies with limbal stem cell deficiency seems promising (e.g., chemical burn).

Author contribution CJF collected data, conceived, and designed the analysis and wrote the paper. FNF performed analysis and collected data. LL contributed data or analysis tools. BDD contributed data and designed the analysis. BS designed the analysis and wrote the paper.

Funding Open Access funding enabled and organized by Projekt DEAL. No funding was received for this research.

Data availability The data were attached to the manuscript as a supplementary file.

Declartions

Conflict of interest All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest (such as honoraria; educational grants; participation in speakers' bureaus; membership, employment, consultancies, stock ownership, or other equity interest; and expert testimony or patent-licensing arrangements), or non-financial interest (such as personal or professional relationships, affiliations, knowledge or beliefs) in the subject matter or materials discussed in this manuscript.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional research committee (Center for Limbal Stem Cell Research and Congenital Aniridia) and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. This manuscript did not involve any kind of animal research. All the authors consent to the publication of this manuscript in International Ophthalmology. For this type of study, formal consent is not required.

Informed consent Informed consent was obtained from all individual participants included in the study.

References

1. Foster MC (1898) Congenital irideremia. Arch Ophthalmol 27(4):593–615
2. Käsmann-Kellner B, Seitz B (2014) Congenital aniridia or PAX6-Syndrome? Ophthalmologe 111:1144

3. Käsmann-Kellner B, Viestenz A, Seitz B (2015) Aniridia guides and aniridia syndrome (PAX6 syndrome): Dos and Don´ts in clinical care & Implementation of supra-regional "Aniridia Guides" can lessen progressive vision loss and improve comprehensive and individualized medical care. In: Parekh M, Poli B, Ferrari S, Teofil C, Ponzi D (eds) Aniridia—recent development in scientific and clinical research. Springer, Switzerland, pp 123–154
4. Lagali N, Wowra B, Fries FN et al (2020) Early phenotypic features of aniridia-associated keratopathy and association with PAX6 coding mutations. Ocul Surf 18(1):130–140
5. Jordan T, Hanson I, Zalatayev D et al (1992) The human PAX6 gene is mutated in two patients with aniridia. Nat Genet 1(5):328–332
6. Akpek EK, Harissi-Dagher M, Petrarca R et al (2007) Outcomes of Boston keratoprosthesis in aniridia: a retrospective multicenter study. Am J Ophthalmol 144:227–231
7. Chen TC, Walton DS (1999) Goniosurgery for prevention of aniridic glaucoma. Arch Ophthalmol 117:1144–1148
8. Seitz B, Käsmann-Kellner B, Viestenz A (2014) Stage-related therapy of congenital aniridia. Ophthalmologe 111:1164–1171
9. Ramaesh K, Ramaesh T, Dutton GN et al (2005) Evolving concepts on the pathogenic mechanisms of aniridia related keratopathy. Int J Biochem Cell Biol 37:547–557
10. Brandt JD, Casuso LA, Budenz DL (2004) Markedly increased central corneal thickness: an unrecognized finding in congenital aniridia. Am J Ophthalmol 137:348–350
11. Lagali N, Wowra B, Fries FN et al (2020) PAX6 mutational status determines aniridia-associated keratopathy phenotype. Ophthalmology 127(2):273–275
12. Seitz B, Das S, Sauer R et al (2011) Simultaneous amniotic membrane patch in high-risk keratoplasty. Cornea 30:269–272
13. Borderie VM, Levy O, Georgeon C et al (2018) Simultaneous penetrating keratoplasty and amniotic membrane transplantation in eyes with a history of limbal stem cell deficiency. J Fr Ophtalmol 4:583–591
14. Lopez-Garcia JS, Rivas L, Garcia-Lozano I et al (2008) Autologous serum eyedrops in treatment of aniridic keratopathy. Ophthalmology 115:262–267
15. Bali S, Filek R, Si F, Hodge W (2016) Systemic immunosuppression in high-risk penetrating keratoplasty: a systematic review. J Clin Med Res 8(4):269–276
16. Chow S, Cook SD, Tole DM (2015) Long-term outcomes of high-risk keratoplasty in patients receiving systemic immunosuppression. Cornea 34(11):1395–1399
17. Yazdanpanah G, Bohm KJ, Hassan OM et al (2020) Management of congenital aniridia-associated keratopathy: long-term outcomes from a tertiary referral center. Am J Ophthalmol 210:8–18
18. Thomasen H, Schroeter J, Reinhard T et al (2018) Good practice procedures for acquisition and preparation of cryopreserved human amniotic membranes from donor placentas. Ophthalmologe 115(10):855–867
19. Wu MF, Stachon T, Seitz B et al (2017) Effect of human autologous serum and fetal bovine serum on human corneal epithelial cell viability, migration and proliferation in vitro. Int J Ophthalmol 10(6):908–913
20. Spaniol K, Koerschgen L, Sander O et al (2014) Comparison of application systems for autologous serum eye drops. Curr Eye Res 39(6):571–579
21. Vicente A, Byström B, Pedrosa Domellöf F (2018) Altered signaling pathways in aniridia-related keratopathy. Invest Ophthalmol Vis Sci 59(13):5531–5541
22. Latta L, Nordström K, Stachon T et al (2019) Expression of retinoic acid signaling components ADH7 and ALDH1A1 is reduced in aniridia limbal epithelial cells and a siRNA primary cell based aniridia model. Exp Eye Res 179:8–17
23. Rubelowski JM, Latta L, Katiyar P et al (2020) HCE-T cell line lacks cornea-specific differentiation markers compared to primary limbal epithelial cells and differentiated cornea epithelium. Graefes Arch Clin Exp Ophthalmol 258(3):565–575
24. Viestenz A, Seitz B, Deland E et al (2018) Clinical anatomy of the anterior chamber angle in congenital aniridia and consequences for trabeculotomy/cyclophotocoagulation. Clin Anat 31(1):64–67
25. Lee H, KhanO'keefe RM (2008) Aniridia: current pathology and management. Acta Opthalmol 86:708–715
26. Jirsova K, Gary L, Jones A (2017) Amniotic membrane in ophthalmology: properties, preparation, storage and indications for grafting—a review. Cell Tissue Bank 18:193–204
27. Eliason JA, McCulley JP (1990) A comparison between interrupted and continuous suturing techniques in keratoplasty. Cornea 9(1):10–16
28. Seitz B, Resch MD, Schlötzer-Schrehardt U et al (2006) Histopathology and ultrastructure of human corneas after amniotic membrane transplantation. Arch Ophthalmol 142(10):1487–1490
29. Resch MD, Schlötzer-Schrehardt U, Hofmann-Rummelt C et al (2006) Integration patterns of cryopreserved amniotic membranes into the human cornea. Ophthalmology 113(11):1927–1935
30. Shimpei K, Yoshinori O, Satoshi K et al (2020) Quantitative analysis of the association between follow-up duration and severity of limbal stem cell deficiency or visual acuity in aniridia. Invest Ophthalmol Vis Sci 61(6):57
31. Alfaro Rangel R, Lepper S, Szentmary N et al (2021) Herpes simplex virus keratitis in a university tertiary referral centre—clinical features and surgical approaches. Klin Monbl Augenheilkd Epub: Feb 19
32. Arraquer RI, Pareja-Aricò L, Gómez-Benlloch A et al (2019) Risk factors for graft failure after penetrating keratoplasty. Medicine (Baltimore) 98(17):e15274
33. Bakhtiari P, Chan C, Welder JD et al (2012) Surgical and visual outcomes of the type I Boston Keratoprosthesis for the management of aniridic fibrosis syndrome in congetinal aniridia. Am J Ophthalmol 153(5):967–971
34. Lang S, Böhringer D, Geerling G et al (2017) Long-term results of allogenic penetrating limbo-keratoplasty: 20 years of experience. Eye 31:372–378

Veröffentlichung 2

Klinische Studie

Thieme

A Cross-sectional Analysis of 556 Eyes Entering the Homburg Aniridia Centre

Eine Querschnittsanalyse von 556 Augen aus dem Homburger Aniridiezentrum

OPEN ACCESS

Authors

Fabian Norbert Fries[1,2], Annamária Náray[1,3], Cristian Munteanu[2], Tanja Stachon[1], Neil Lagali[4], Berthold Seitz[2], Nóra Szentmáry[1*], Barbara Käsmann-Kellner[2*]

Affiliations

1 Dr. Rolf M. Schwiete Center for Limbal Stem Cell and Aniridia Research, Saarland University, Homburg/Saar, Germany
2 Department of Ophthalmology, Saarland University Medical Center, Homburg/Saar, Germany
3 Department of Ophthalmology, Semmelweis University, Budapest, Hungary
4 Department of Biomedical and Clinical Sciences, Faculty of Medicine, Linköping University, Linköping, Sweden

Key words

congenital aniridia, Homburg Aniridia Center, glaucoma, macular hypoplasia, aniridia-associated keratopathy, Pax-6 gene

Schlüsselwörter

kongenitale Aniridie, Pax-6-Gen, Glaukom, Makulahypoplasie, Homburger Aniridiezentrum, Aniridie-assoziierte Keratopathie

received 27. 11. 2022
accepted 15. 2. 2023
published online

Bibliography

Klin Monatsbl Augenheilkd 2023
DOI 10.1055/a-2065-8405
ISSN 0023-2165

Georg Thieme Verlag KG, Rüdigerstraße 14, 70469 Stuttgart, Germany

Correspondence

Fabian Norbert Fries, MD
Department of Ophthalmology, Saarland University Hospital and Saarland University Faculty of Medicine
Kirrberger Str. 100, 66421 Homburg/Saar, Germany
Phone: + 496 84 11 62 23 87, Fax: + 49 6 84 1 16 2 24 00
fries.fn@gmail.com

ABSTRACT

Purpose Congenital aniridia is a severe malformation of almost all eye segments. In addition, endocrinological, metabolic, and central nervous systems diseases may be present. In order to develop better treatment options for this rare disease, an aniridia center must be established. The purpose of this work is to summarize ophthalmic findings of aniridia subjects examined at the Department of Ophthalmology, Saarland University Medical Center in Homburg.

Methods Our retrospective single-center study included patients who underwent a comprehensive ophthalmic examination through the head of the KiOLoN ("Kinderophthalmologie", Orthoptics, Low Vision and Neuroophthalmology) Unit of the department between June 2003 and January 2022. Data at the first examination time point have been included.

Results Of 286 subjects, 556 eyes of (20.1 ± 20.1 years; 45.5 % males) were included. There was nystagmus in 518 (93.7 %) eyes, and strabismus in 327 (58.8 %) eyes. There were 436 (78.4 %) eyes with age-appropriate axial length, 104 (18.7 %) eyes with microphthalmos, and 13 (2.3 %) eyes with buphthalmos. There was iris malformation with atypical coloboma in 34 eyes (6.1 %), more than 6 clock hours of iris remnants in 61 eyes (10.9 %), less than 6 clock hours of iris remnants in 96 eyes (17.2 %), and complete aniridia in 320 (57.5 %) eyes. The patients were graded according to the following aniridia-associated keratopathy (AAK) stages: Stage 0 (96 eyes [17.2 %], no keratopathy), Stage 1 (178 eyes [32.0 %]), Stage 2 (107 eyes [19.2 %]), Stage 3 (67 eyes [12.0 %]), Stage 4 (62 eyes [11.1 %]), Stage 5 (45 eyes [8.0 %]). There was secondary glaucoma in 307 (55.5 %), macular hypoplasia in 395 (71.4 %), and congenital optic nerve

* Prof. Szentmáry and Prof. Käsmann-Kellner are co-senior authors of the manuscript.

head pathology in 223 (40.3 %) eyes. The iris malformation type was significantly positively correlated with AAK stage, lens properties, presence of glaucoma, congenital macular, and optic nerve head properties ($p < 0.001$ for all), while complete aniridia showed the most complications.

Conclusions At the Homburg Aniridia Center, the most common ophthalmic signs in congenital aniridia were AAK, iris malformation, cataract, and macular hypoplasia. The iris malformation type may indicate future expression of AAK, cataract, and glaucoma development and it is correlated with a congenital optic nerve head and macular pathology. Our registry will support further detailed longitudinal analysis of ophthalmic and systemic diseases of aniridia subjects during long-term follow-up.

ZUSAMMENFASSUNG

Hintergrund Die kongenitale Aniridie ist eine schwere Fehlbildung fast aller Augensegmente. Darüber hinaus können auch endokrinologische, metabolische und zentralnervöse Erkrankungen vorliegen. Um bessere Behandlungsmöglichkeiten für diese seltene Krankheit zu entwickeln, ist die Einrichtung eines Aniridie-Zentrums notwendig. Unser Ziel ist es, die ophthalmologischen Befunde von Aniridiepatienten zusammenzufassen, die in der Klinik für Augenheilkunde am Universitätsklinikum des Saarlandes (UKS) in Homburg untersucht wurden.

Methoden Unsere retrospektive, monozentrische Studie umfasste Patienten, die sich zwischen Juni 2003 und Januar 2022 einer umfassenden augenärztlichen Untersuchung durch die Leiterin der Abteilung KiOLoN (Kinderophthalmologie, Orthoptik, Low Vision und Neuroophthalmologie) unterzogen. Die Daten des ersten Untersuchungszeitpunkts wurden einbezogen.

Ergebnisse Es wurden 556 Augen von 286 Probanden (20,1 ± 20,1 Jahre; 45,5% Männer) eingeschlossen. Bei 518 (93,7%) Augen lag ein Nystagmus vor, Strabismus bei 327 (58,8%) Augen. 436 (78,4%) Augen wiesen eine altersentsprechende Achsenlänge auf, 104 (18,7%) Augen hatten einen Mikrophthalmus und 13 (2,3%) Augen einen Buphthalmus. Bei 34 Augen (6,1%) lag eine Irisfehlbildung mit atypischem Kolobom vor, bei 61 Augen (10,9%) ein Irisrest von mehr als 6 Uhrzeiten, bei 96 Augen (17,2%) ein Irisrest von weniger als 6 Uhrzeiten und bei 320 Augen (57,5%) eine vollständige Aniridie. Die Patienten wurden nach den folgenden Stadien der aniridieassoziierten Keratopathie (AAK) eingeteilt: Stadium 0 (96 Augen [17,2%], keine Keratopathie), Stadium 1 (178 Augen [32,0%]), Stadium 2 (107 Augen 819,2%]), Stadium 3 (67 Augen [12,0%9), Stadium 4 (62 Augen [11,1%]), Stadium 5 (45 Augen [8,0%]). Ein sekundäres Glaukom lag bei 307 (55,5%), eine Makulahypoplasie bei 395 (71,4%) und eine angeborene Pathologie des Sehnervenkopfes bei 223 (40,3%) Augen vor. Die Art der Irisfehlbildung war signifikant positiv korreliert mit dem AAK-Stadium, den Linseneigenschaften, dem Vorhandensein eines Glaukoms, den angeborenen Makula- und Sehnervenkopf-Eigenschaften ($p < 0,001$ für alle), wobei eine komplette Aniridie die meisten Komplikationen aufwies.

Schlussfolgerungen Im Homburger Aniridiezentrum waren die häufigsten ophthalmologischen Zeichen bei kongenitaler Aniridie AAK, Irisfehlbildung, Katarakt und Makulahypoplasie. Der Typ der Irisfehlbildung kann auf die zukünftige Ausprägung von AAK, Katarakt und Glaukom hinweisen und ist mit der angeborenen Pathologie des Sehnervenkopfes und der Makula korreliert. Unser Register wird eine weitere detaillierte Analyse der ophthalmischen und systemischen Erkrankungen von Aniridie-Patienten im Langzeitverlauf ermöglichen.

Introduction

Aniridia is considered a rare disease, with a global prevalence of 1 in 40 000 to 1 in 100 000 [1 – 5]. Despite its name, aniridia is a panocular disorder that takes its name from the obvious hypoplasia of the iris, which is present in most cases. This feature can range from a conspicuous, almost complete loss of the iris to enlargement and irregularity of the pupil, representing a coloboma, to microscopic slit-like anomalies of the pupillary margin that can only be seen with slit lamp illumination. The effects on vision are also variable. In most cases, there is already congenital severe visual loss and, consequently, pathological visual development and nystagmus [1 – 4, 6].

In congenital aniridia, one can distinguish between the PAX6 gene-associated forms and other forms without alterations in the PAX6 gene, with the PAX6 forms being significantly more common [7, 8]. The typical clinical PAX6-related congenital aniridia occurs in several forms: dominant inheritance, occurring sporadically (then inherited dominantly), as part of the WAGR (Wilms tumor, aniridia, genitourethral anomalies, retardation) or WAGRO (WAGR plus "obesity") syndrome, and associated with other syndromes. Long-term complications with visual impairment, such as glaucoma or severe aniridia-associated keratopathy (AAK), are more frequent in PAX6-related aniridia [1, 3].

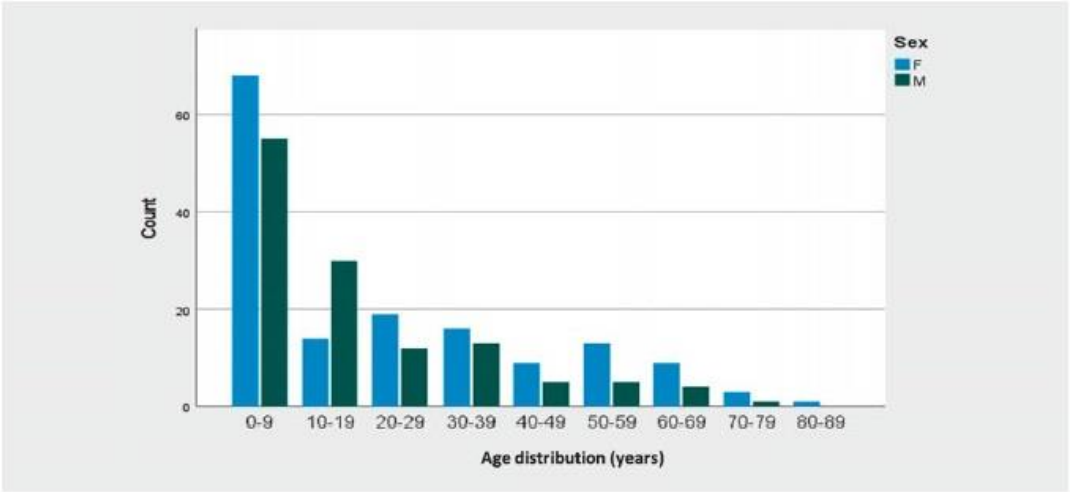
Since it has become more and more evident in recent years that so-called "isolated" PAX6 aniridia can also frequently have systemic concomitant diseases (hormonal, gastrointestinal, metabolic, cerebral), the term "aniridia syndrome" or "PAX6 syndrome" has been recommended [1, 3].

In order to develop better treatment options for the rare disease congenital aniridia, establishment of an aniridia center is necessary. The purpose of this work is to summarize ophthalmic properties of aniridia subjects examined at the Department of Ophthalmology, Saarland University Medical Center, in Homburg/Saar, Germany.

Patients and Methods

Ethical considerations

Our retrospective single-center study included patients from the Department of Ophthalmology, Saarland University Medical Cen-



▶ **Fig. 1** Age distribution of subjects at the Homburg Aniridia Center at the first examination time point.

ter in Homburg/Saar, Germany. This study was approved by the Ethics Committee of Saarland/Germany (No 144/15) and followed regulations of the Declaration of Helsinki. Informed consent was obtained from all participants. In case of minors or guardianship, informed consent was obtained from the legal representative or legal guardian.

Inclusion criteria, data collection, and examination methods

Inclusion criteria was the presence of partial or complete congenital aniridia, visible at slit lamp examination. All subjects underwent a structured ophthalmic examination through the Head of the KiOLoN ("Kinderophthalmologie", Orthoptics, Low Vision and Neuroophthalmology) Unit of the Department of Ophthalmology of Saarland University, Prof. Dr. Barbara Käsmann-Kellner. Uncorrected and best-corrected visual acuity (UCVA and BCVA) measurement using Snellen charts, intraocular pressure (IOP) measurement using Goldmann applanation tonometry or iCare (Icare Finland Oy, Vantaa, Finland), and a detailed slit lamp and fundus examination were performed.

Iris malformation was classified as atypical coloboma, more than 6 clock hours of iris remnants, less than 6 clock hours of iris remnants, and complete aniridia (no iris remnant tissue is visible at slit lamp examination, without gonioscopy). Limbal stem cell insufficiency (LSCI) was classified as follows: (1) no limbal changes, (2) avascular pannus with less than 3 mm width, (3) vascularized pannus with less than 3 mm width, (4) vascularized pannus over 3 mm width. AAK was classified as Stage 0 (no limbal changes), Stage 1 (conjunctival tissue just crosses the limbal border but remains 1 mm or less from the limbus), Stage 2 (the pannus extends across the peripheral cornea and is typically present in 360 degrees of the cornea), Stage 3 (the pannus invades the central cornea, typically covering the entire cornea with vessels), Stage 4 (the cornea is completely vascularized), or Stage 5 (end-stage with an opaque, thick, vascularized cornea) [7,8].

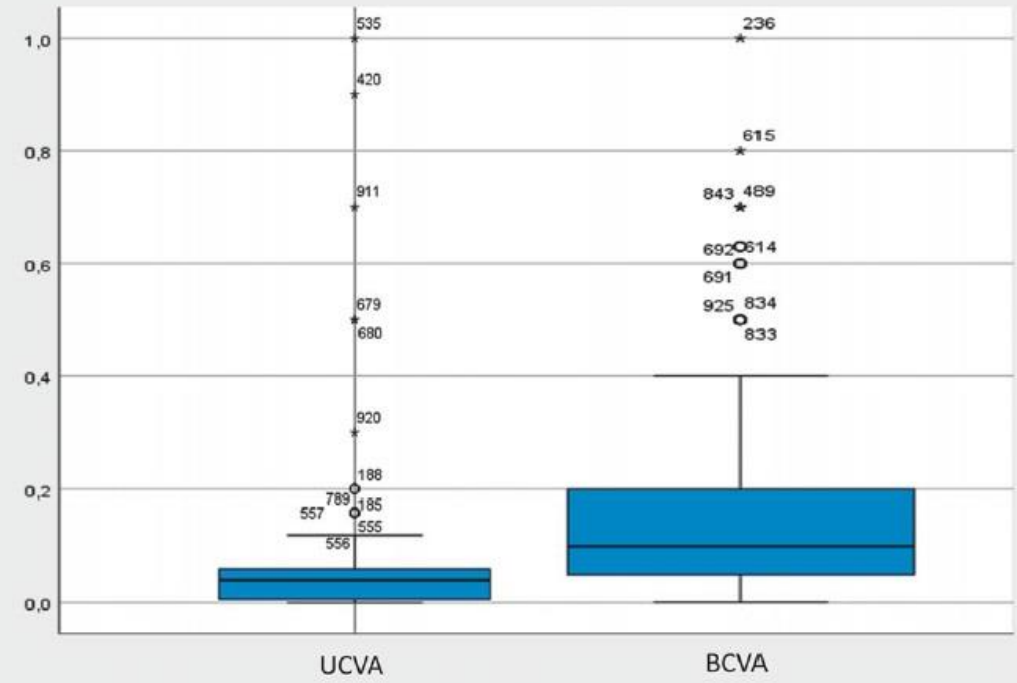
All patient data were entered pseudonymized in a Microsoft Access database. In collaboration with the Department of Ophthalmology, Saarland University Medical Center in Homburg/Saar (Chair: Prof. Dr. B. Seitz) and the Dr. Rolf M. Schwiete Center for Limbal Stem Cell and Aniridia Research, Homburg/Saar (Chair: Prof. Dr. N. Szentmáry), our aim was to build up a database in order to get better insight into the pathomechanisms and stage-appropriated treatment options of congenital aniridia. The present study summarizes patient data at the first examination time point for subjects examined between June 2003 and January 2022.

Results

Of 286 subjects, 556 eyes (age 20.1 ± 20.1 years; 45.5 % males) were included. Age distribution of the subjects is displayed in ▶ **Fig. 1**. UCVA was 0.074 ± 0.013 (0.001–1.0) and BCVA was 0.15 ± 0.08 (0.001–1.0; ▶ **Fig. 2**) at the first examination time point.

There was nystagmus in 518 (93.7 %) eyes and strabismus in 327 (58.8 %) eyes. There were 436 (78.4 %) eyes with age-appropriate axial length, 104 (18.7 %) eyes with microphthalmos, 13 (2.3 %) eyes with buphthalmos, and in 3 (0.6) eyes, no axial length measurement was performed at the first examination time point. There was iris malformation with atypical coloboma in 34 eyes (6.1 %), more than 6 clock hours of iris remnants in 61 eyes (10.9 %), less than 6 clock hours of iris remnants in 96 eyes (17.2 %), and complete aniridia in 320 (57.5 %) eyes (▶ **Fig. 3a–d**). Nevertheless, in 45 (8.3 %) eyes, we could not collect data on the exact iris malformation type, mainly due to corneal opacities.

LSCI was classified as follows: (1) no limbal changes in 97 eyes (17.4 %), (2) avascular pannus with less than 3 mm width in 174



▸ **Fig. 2** Uncorrected visual acuity (UCVA) and best-corrected visual acuity (BCVA; Snellen) of the analyzed subjects at the first examination time point.

eyes (31.2%), (3) vascularized pannus with less than 3 mm width in 79 eyes (14.2%), and (4) vascularized pannus over 3 mm width in 190 eyes (34.1%). In 16 eyes (3.1%), no data on LSCI was available at the first time point of examination.

There was AAK Stage 0 (no keratopathy) in 96 eyes (17.2%), Stage 1 in 178 eyes (32.0%) eyes, Stage 2 in 107 eyes (19.2%) eyes, Stage 3 in 67 eyes (12.0%) eyes, Stage 4 in 62 eyes (11.1%), and Stage 5 in 45 eyes (8.0%). One eye (0.3%) could not be included in any of the groups along the available clinical data (▸ **Fig. 3e–i**).

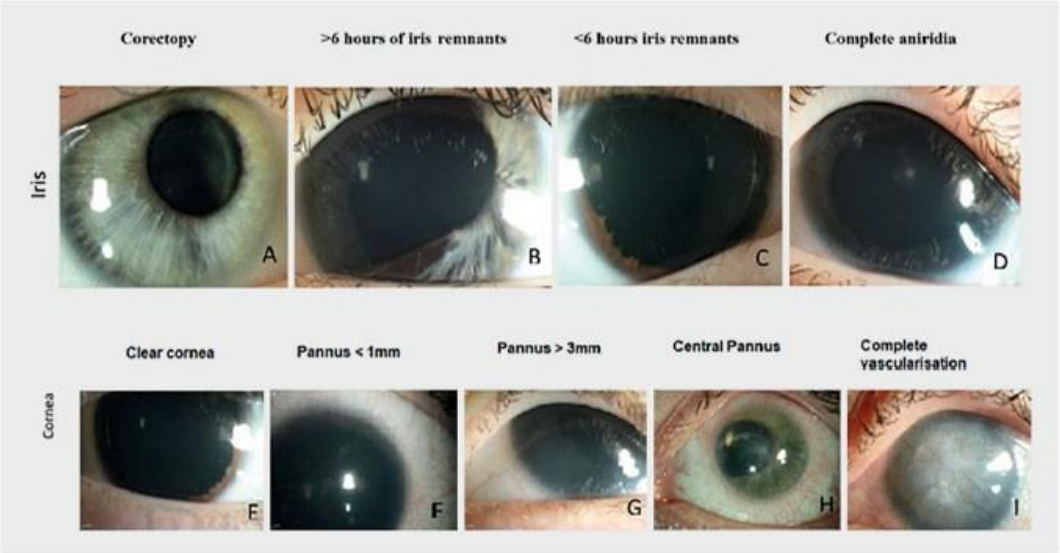
The lens was clear in 127 eyes (22.8%), there was cataract in 224 eyes (40.2%), subluxated lens in 9 eyes (1.6%), pseudophakia in 129 eyes (23.2%), aphakia in 32 eyes (5.7%), and in 35 eyes (6.5%), the lens status could not be assessed. There was secondary glaucoma in 307 eyes (55.5%), macular hypoplasia in 395 eyes (71.4%), and congenital optic nerve head pathology in 223 eyes (40.3%) eyes (▸ **Fig. 4**).

The iris malformation type was significantly positively correlated with AAK stage, lens properties, presence of glaucoma, congenital macular, and optic nerve head properties ($p < 0.001$ for all), with complete aniridia showing the most complications.

Discussion

Collecting data of 556 eyes of 286 subjects from one of the largest worldwide aniridia databases, the Homburg Aniridia Center could be established in Homburg/Saar. About one-fourth of the included subjects were children. With a mean UCVA below 0.1 and a mean BCVA below 0.2, most of the analyzed subjects necessitated special education and visual aids during life [1–3].

The misdevelopment of the iris is the characteristic phenotypic appearance in aniridia. It can range from a complete absence of the iris to a slight shift of the pupil (corectopy) or an atypical coloboma [7,8]. Iris malformation is one of the causes of photophobia. We could observe that about two-thirds of the analyzed subjects (75.3%) had less than 6 clock hours of iris remnants (17.4%), or complete aniridia (in 320 eyes [57.9%]), which enables a relatively obvious immediate diagnosis for ophthalmologists. Nevertheless, ophthalmologists also have to take into consideration that about 17% of the patients with congenital aniridia may present with an atypical coloboma (6.1%), or more than 6 clock hours of iris remnants (11%), but these signs may also indicate congenital aniridia. Additionally, in some cases, due to corneal



▶ **Fig. 3** Iris malformation (**a – d**) and aniridia-associated keratopathy (**e, f, g, h**) in congenital aniridia subjects.

opacities, the lack of iris might not be observable, which may result in a wrong diagnosis.

Most interestingly, statistical analysis confirmed the clinical suspicion that the iris malformation type was significantly positively correlated with AAK stage, lens properties, presence of glaucoma, congenital macular, and optic nerve head properties ($p < 0.001$ for all), with complete aniridia showing the most complications.

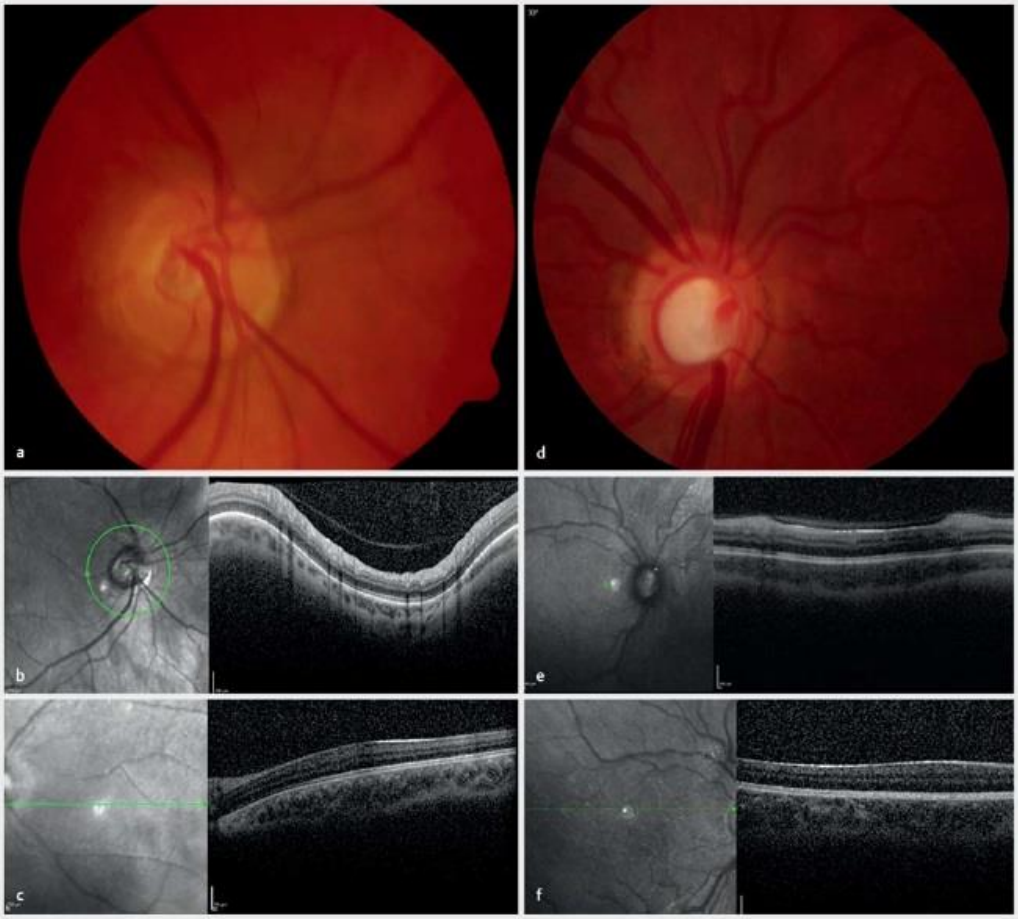
Up to 70% of PAX6 aniridia sufferers develop AAK with age, which may be due to a combination of several factors [1–3, 6–10]. These include a pronounced dry eye problem, LSCI with impaired corneal epithelial cell differentiation, abnormal cell adhesion, and wound healing [1–3, 5–10]. The consequences for the patients are permanent tear film instability as well as recurrent extremely painful corneal erosions, and a progression of visual loss due to vascularized corneal pannus and/or scars [5, 7–9]. Among our subjects, with relatively young age, most of the subjects belonged to the Stage 1 AAK group (38.8% of the eyes), followed by Stage 2 with 19.3%, Stage 0 with 17.4%, Stage 3 in 12.1%, Stage 4 in 11.2%, and Stage 5 in 8.1% of the eyes.

AAK is characterized by centripetal spreading vascularization, conjunctivalization, and thickening of the cornea, which is, in part, due to LSCI [5, 10–14]. There was LSCI with avascular pannus with less than 3 mm width in 174 (31.2%) eyes, vascularized pannus with less than 3 mm width in 79 (14.2%) eyes, and vascularized pannus over 3 mm width in 190 (34.1%) eyes.

The natural history of AAK shows several stages of progression. Signs of keratopathy often appear in early youth with thickening of the peripheral corneal epithelium but no functional manifestation. In the second decade, patients show chronic irritation and thin superficial vascularization in the peripheral cornea, which gradually progresses to the central cornea. Pain, photophobia, and recurrent corneal epithelial erosions are common. In later stages, the keratopathy progresses until the entire cornea is involved, with a severe increase in central corneal thickness due to pannus formation and vascularized scars [5, 7, 9].

Cataract occurs in 50–85% of aniridia patients. In the Homburg Aniridia Center, there was a clear lens in 127 eyes (23.0%), cataract in 224 eyes (40.6%), subluxated lens in 9 eyes (1.6%), pseudophakia in 129 eyes (23.3%), aphakia in 32 eyes (5.8%), and in 21 eyes (3.8%), the lens status could not be assessed. In many cases, a cataract of the anterior and posterior lens pole (cataracta polaris anterior or posterior) is found congenitally, which often remains stable during life. In addition, progressive opacification of the other lens segments may occur, as well as subluxation or luxation of the crystalline lens due to lack/insufficiency of zonular fibers. Both may be an indication for lens removal and if possible, implantation of an intraocular lens to preserve visual acuity [1–3, 6, 9].

Secondary glaucoma occurs with an incidence of 6–75% in aniridia subjects, often before adulthood. There was secondary glaucoma in 307 eyes (55.5%) of our subjects. Aniridia-associated glaucoma is caused by an abnormal localization of the Schlemm canal or by iris rudiments, which close the chamber angle or the trabecular meshwork and thus obstruct the outflow of aqueous humor. Diagnosis requires regular monitoring of IOP as well as the optic nerve. Often, a pressure measurement is difficult due to the thickened cornea and requires a corneal thickness measurement. Secondary glaucoma can lead to irreversible visual loss, even ending up in blindness due to glaucomatous optic atrophy



▶ **Fig. 4** Congenital optic nerve head pathology (**a**, **b**) and macular hypoplasia (**c**), glaucomatous excavation (**d**, **e**), and macular hypoplasia (**f**) in congenital aniridia.

and thus must be potentially considered the most serious irreversible complication [1–3,6].

The Homburg Aniridia Center examined 395 (71.4%) eyes with macular hypoplasia and 223 (40.3%) eyes with congenital optic nerve head pathology. In aniridia patients, hypoplasia of the optic nerve and macula was more common. Macular hypoplasia does not necessarily occur together with optic hypoplasia, it can also occur as an isolated symptom in the context of congenital visual impairment. Furthermore, a mostly horizontal nystagmus as well as strabismus can be observed. We observed nystagmus in 518 (93.7%) eyes and strabismus in 327 (58.8%) eyes.

With regard to therapy, there are currently no generally accepted treatment modalities. In case of corneal involvement (AAK), different therapeutic options arise depending on the severity, which range from autologous serum eye drops, amniotic membrane transplantation, and phototherapeutic keratectomy to lamellar and penetrating keratoplasty. For penetrating keratoplasties, the following procedure has been proven helpful in our Department of Ophthalmology in these high-risk patients: systemic immunosuppression, small-sized penetrating keratoplasty with single knot sutures, simultaneous transplantation of an amniotic membrane as a patch, temporary lateral tarsorrhaphy, and autologous serum eye drops [9].

As treatment for limbal stem cell deficiency in congenital aniridia, the use of limbal allografts (4 eyes), keratolimbal allografts (31 eyes), cultivated limbal epithelial cells (10 eyes), and

cultivated oral mucosal epithelial cells (17 eyes) have been reported [15]. These procedures may be combined with systemic immunosuppression, simultaneous transplantation of an amniotic membrane as a patch, temporary lateral tarsorrhaphy, and postoperative use of autologous serum eye drops. Following surgery, visual acuity improves during the first 6 months, which thereafter, gradually declines. Patients were followed for 12–18 months after epithelial (stem) cell transplantation, however, longer term outcomes and further procedures have not been reported, yet [1–4, 9, 14, 15].

Because of the feared aniridia-fibrosis syndrome and the increased risk of glaucoma due to obstruction of the chamber angle and thus of the aqueous humor outflow, the surgical insertion of an artificial iris should be avoided. Anti-glaucomatous therapy should be started before visual field loss occurs. If an IOP increase is resistant to therapy, trabeculotomy is the method of first choice. Lens opacification can be surgically treated by insertion of an artificial lens, if necessary, with a capsular tension ring (small incision, "in-the-bag"), if visual acuity is clearly impaired [1–4, 9].

Prognosis limiting is the observation that surgical therapy options in aniridia patients have a multiple higher complication rate than in patients not affected by this disease. One well-known complication after repeated ocular surgery is progressive anterior segment fibrosis syndrome. In this case, a non-acute inflammatory fibrotic membrane develops in the anterior chamber starting from the iris remnants, which grows into the posterior chamber over the ciliary body, detaching the ciliary body and thus causing bulbar hypotony, with consecutive retinal detachment [1–4, 9].

Due to the rarity of aniridia, all ophthalmic clinics in Germany, university based or others, care for relatively few patients. Thus, no sound treatment guidelines exist for PAX6 aniridia and its complications. We established a clinical aniridia registry (observational study) at our hospital in order to systematically record the course of this rare disease, to optimize its diagnostics and therapy, and thus to further improve the treatment of the disease in the future.

Currently, 460 patients with congenital aniridia are regularly followed at our clinic (2/3 are children and adolescents under 16 years of age, and 1/3 are adults over 16 years), more than 98 % of whom live outside Saarland. Nevertheless, it is estimated that there are currently about 940 patients in Germany with the diagnosis "congenital aniridia" (about 85 % may be PAX6-associated aniridia). Most of these patients or their parents or legally designated caregivers have registered themselves in the national self-help organization "AWS Aniridie-WAGR e. V." (www.aniridie-wagr.de).

Prof. Käsmann-Kellner has been a volunteer medical advisor to the association since its inception, and the association supported the establishment of our Aniridia Center. A large proportion of the patients we care for are children, and accordingly – as with most of the rare congenital diseases – measurable effects of our planned interventions may not become apparent for many years. The aim is therefore to store the data collected in the registry for the long term, initially for 10 years, allowing a longitudinal long-term follow-up.

CONCLUSION BOX

Already known:

- In congenital aniridia, there is an increased risk of developing blindness during life.
- As congenital aniridia is a rare disease, it is difficult to establish clinical prognostic parameters and treatment standards.
- In order to develop better treatment options in congenital aniridia, establishment of an aniridia center is necessary.

Newly described:

- The most prevalent ophthalmic signs in congenital aniridia are AAK, iris malformation, cataract, and macular hypoplasia.
- The iris malformation type may indicate future expression of AAK, cataract, and glaucoma development and it is correlated with congenital optic nerve head and macular pathology.
- Our Aniridia Center will support further detailed longitudinal analysis of ophthalmic and systemic diseases of these difficult patients with congenital aniridia during long-term follow-up.

Acknowledgements

The work of Dr. Szentmáry at the Dr. Rolf M. Schwiete Centre was generously supported by the Dr. Rolf M. Schwiete Foundation, Mannheim. This article is based upon work from COST Action CA18116, supported by COST (European Cooperation in Science and Technology).

Conflict of Interest

The authors declare that they have no conflict of interest.

References

[1] Käsmann-Kellner B, Fries FN, Latta L et al. Aniridie bei PAX6-Syndrom: Eine potenziell zur Erblindung führende Erkrankung. Etablierung eines Deutschen Aniridie-Registers. Concept Ophthalmologie 2019; 12–18. Im Internet (Stand: 03.04.2023): https://www.researchgate.net/publication/353515480_Aniridie_bei_PAX6-Syndrom_Eine_potenziell_zur_Erblindung_fuhrende_Erkrankung_Etablierung_eines_Deutschen_Aniridie-Registers

[2] Käsmann-Kellner B, Seitz B. [Aniridia syndrome: clinical findings, problematic courses and suggestions for optimization of care ("aniridia guide")]. Ophthalmologe 2014; 111: 1145–1156. doi:10.1007/s00347-014-3060-x

[3] Käsmann-Kellner B, Seitz B. [Congenital aniridia or PAX6 syndrome]. Ophthalmologe 2014; 111: 1144. doi:10.1007/s00347-014-3058-4

[4] Seitz B, Käsmann-Kellner B, Viestenz A. [Stage-related therapy of congenital aniridia]. Ophthalmologe 2014; 111: 1164–1171. doi:10.1007/s00347-014-3061-9

[5] Latta L, Figueiredo FC, Ashery-Padan R et al. Pathophysiology of aniridia-associated keratopathy: Developmental aspects and unanswered questions. Ocul Surf 2021; 22: 245–266. doi:10.1016/j.jtos.2021.09.001

[6] Käsmann-Kellner B, Latta L, Fries FN et al. Diagnostic impact of anterior segment angiography of limbal stem cell insufficiency in PAX6-related aniridia. Clin Anat 2018; 31: 392–397. doi:10.1002/ca.22987

[7] Lagali N, Wowra B, Fries FN et al. PAX6 Mutational Status Determines Aniridia-Associated Keratopathy Phenotype. Ophthalmology 2020; 127: 273–275. doi:10.1016/j.ophtha.2019.09.034

[8] Lagali N, Wowra B, Fries FN et al. Early phenotypic features of aniridia-associated keratopathy and association with PAX6 coding mutations. Ocul Surf 2020; 18: 130–140. doi:10.1016/j.jtos.2019.11.002

[9] Farah CJ, Fries FN, Latta L et al. An attempt to optimize the outcome of penetrating keratoplasty in congenital aniridia-associated keratopathy (AAK). Int Ophthalmol 2021; 41: 4091–4098. doi:10.1007/s10792-021-01982-z

[10] Katiyar P, Stachon T, Fries FN et al. Decreased FABP5 and DSG1 protein expression following PAX6 knockdown of differentiated human limbal epithelial cells. Exp Eye Res 2022; 215: 108904. doi:10.1016/j.exer.2021.108904

[11] Latta L, Knebel I, Bleil C et al. Similarities in DSG1 and KRT3 Downregulation through Retinoic Acid Treatment and PAX6 Knockdown Related Expression Profiles: Does PAX6 Affect RA Signaling in Limbal Epithelial Cells? Biomolecules 2021; 11: 1651. doi:10.3390/biom11111651

[12] Latta L, Ludwig N, Krammes L et al. Abnormal neovascular and proliferative conjunctival phenotype in limbal stem cell deficiency is associated with altered microRNA and gene expression modulated by PAX6 mutational status in congenital aniridia. Ocul Surf 2021; 19: 115–127. doi:10.1016/j.jtos.2020.04.014

[13] Latta L, Nordström K, Stachon T et al. Expression of retinoic acid signaling components ADH7 and ALDH1A1 is reduced in aniridia limbal epithelial cells and a siRNA primary cell based aniridia model. Exp Eye Res 2019; 179: 8–17. doi:10.1016/j.exer.2018.10.002

[14] Schlötzer-Schrehardt U, Latta L, Gießl A et al. Dysfunction of the limbal epithelial stem cell niche in aniridia-associated keratopathy. Ocul Surf 2021; 21: 160–173. doi:10.1016/j.jtos.2021.06.002

[15] Landsend ECS, Lagali N, Utheim TP. Congenital aniridia – A comprehensive review of clinical features and therapeutic approaches. Surv Ophthalmol 2021; 66: 1031–1050. doi:10.1016/j.survophthal.2021.02.011

Veröffentlichung 3

Klinische Studie

Thieme

The Effect of Glaucoma Treatment on Aniridia-Associated Keratopathy (AAK) – A Report from the Homburg Register for Congenital Aniridia

Die Auswirkung der Glaukombehandlung auf die Aniridie-assoziierte Keratopathie (AAK) – Ein Bericht aus dem Homburger Register für kongenitale Aniridie

OPEN ACCESS

Authors
Fabian Norbert Fries[1,2], Annamária Náray[1,3], Cristian Munteanu[2], Tanja Stachon[1], Neil Lagali[4], Berthold Seitz[2], Barbara Käsmann-Kellner[2], Nóra Szentmáry[1]

Affiliations
1 Dr. Rolf M. Schwiete Center for Limbal Stem Cell and Aniridia Research, Saarland University, Homburg/Saar, Germany
2 Department of Ophthalmology, Saarland University Medical Center, Homburg/Saar, Germany
3 Department of Ophthalmology, Semmelweis University, Budapest, Hungary
4 Department of Biomedical and Clinical Sciences, Faculty of Medicine, Linköping University, Linköping, Sweden

Key words
congenital aniridia, Homburg Aniridia Center, severity of glaucoma, number of antiglaucomatous eye drops, progression of aniridia-associated keratopathy

Schlüsselwörter
kongenitale Aniridie, Homburger Aniridie-Zentrum, Schweregrad des Glaukoms, Anzahl der antiglaukomatösen Augentropfen, Progression der Aniridie-assoziierten Keratopathie

received 29.7.2023
accepted 18.8.2023

Bibliography
Klin Monatsbl Augenheilkd 2023
DOI 10.1055/a-2194-1580
ISSN 0023-2165

Georg Thieme Verlag KG, Rüdigerstraße 14, 70469 Stuttgart, Germany

Correspondence
Fabian Norbert Fries, MD
Department of Ophthalmology, Saarland University Medical Center
Kirrberger Str. 100, 66421 Homburg/Saar, Germany
Phone: + 49 6841 1623 87, Fax: + 49 6841 1624 00
fries.fn@gmail.com

ABSTRACT

Background Congenital aniridia is a severe malformation of almost all eye segments. Aniridia-associated keratopathy (AAK) and secondary glaucoma, which occur in more than 50% of affected individuals, are typically progressive and pose a high risk of blindness for patients with congenital aniridia. Our aim was to investigate the effect of glaucoma treatment on AAK in patients of the Homburg Aniridia Center.

Methods Our retrospective monocentric study included patients who underwent a comprehensive ophthalmological examination at the Homburg Aniridia Center between June 2003 and January 2022.

Results There were 556 eyes of 286 subjects (20.1 ± 20.1 years; 45.5% males) included. In 307 (55.2%) eyes of 163 subjects (27.5 ± 16.3 years; 43.1% males), glaucoma was present at the time of examination. The mean intraocular pressure in the glaucoma group was 19.0 mmHg (± 8.0), while in the non-glaucoma group, it was 14.1 mmHg (± 3.6) ($p < 0.001$). In the glaucoma group, 68 patients used antiglaucomatous topical monotherapy, 51 patients used 2 agents, 41 patients used 3 agents, 7 patients used quadruple therapy, and 140 did not use topical therapy (e.g., after pressure-lowering surgery, pain-free end-stage glaucoma, or incompliance). Patients were classified according to the following stages of AAK: Stage 0 (96 eyes [17.2%], no keratopathy), Stage 1 (178 eyes [32.0%]), Stage 2 (107 eyes [19.2%]), Stage 3 (67 eyes [12.0%]), Stage 4 (62 eyes [11.1%]), Stage 5 (45 eyes [8.0%]). The mean stage of AAK was 1.4 (1.2–1.5) in the group without eye drops, 1.9 (1.5–2.2) in the group with

monotherapy, 1.8 (1.5–2.1) in the group with 2 drugs, 1.9 (1.5–2.2) in the group with 3 drugs, 3.4 (2.3–4.6) in the group with 4 drugs, and 3.3 (3.1–3.6) after antiglaucomatous surgery. The stage of AAK was significantly positively correlated with the number of pressure-lowering eye drops ($p < 0.05$) and prior pressure-lowering surgery ($p < 0.05$). Prostaglandin analogues were not correlated with a higher AAK stage compared to the other drug groups.

Conclusions At the Homburg Aniridia Center, patients using topical antiglaucomatous quadruple therapy or who had previously undergone antiglaucomatous surgery had by far the highest AAK stage. The different drug groups had no influence on the AAK stage.

ZUSAMMENFASSUNG

Hintergrund Die kongenitale Aniridie ist eine schwere Fehlbildung fast aller Augensegmente. Insbesondere die Aniridie-assoziierte Keratopathie (AAK) sowie das bei mehr als 50 % der Betroffenen auftretende Sekundärglaukom verlaufen typischerweise progressiv und stellen ein hohes Risiko der Erblindung für Patienten mit kongenitaler Aniridie dar. Unser Ziel war es, bei Patienten des Homburger Aniridie-Zentrums die Auswirkung der Glaukombehandlung auf die AAK zu untersuchen.

Methoden Unsere retrospektive, monozentrische Studie umfasste Patienten, die sich zwischen Juni 2003 und Januar 2022 einer umfassenden augenärztlichen Untersuchung durch das Homburger Aniridie-Zentrum unterzogen.

Ergebnisse Es wurden 556 Augen von 286 Probanden (20,1 ± 20,1 Jahre; 45,5% Männer) eingeschlossen. Bei 307 (55,2%) Augen von 163 Patienten (27,5 ± 16,3 Jahre; 43,1 % Männer) lag zum Zeitpunkt der Untersuchung ein Glaukom vor. Der Augeninnendruck lag in der Glaukomgruppe im Mittel bei 19,0 mmHg (± 8,0) während er bei den Patienten ohne Glaukom bei 14,1 mmHg (± 3,6) lag ($p < 0,001$). In der Glaukomgruppe nutzten 68 Patienten eine lokale antiglaukomatöse Monotherapie, 51 Patienten nutzten 2 Wirkstoffe, 41 Patienten nutzten 3 Wirkstoffe, 7 Patienten nutzten eine Vierfachtherapie, 140 nutzten keine Lokaltherapie (z.B. nach drucksenkender Operation, schmerzfreies Glaukom-Endstadium oder Incompliance). Die Patienten wurden nach den folgenden Stadien der Aniridie-assoziierten Keratopathie (AAK) eingeteilt: Stadium 0 (96 Augen [17,2%], keine Keratopathie), Stadium 1 (178 Augen [32,0%]), Stadium 2 (107 Augen [19,2%]), Stadium 3 (67 Augen [12,0%]), Stadium 4 (62 Augen [11,1%]), Stadium 5 (45 Augen [8,0%]). Das Stadium der Aniridie-assoziierten Keratopathie lag in der Gruppe ohne Augentropfen im Mittel bei 1,4 (1,2–1,5), in der Gruppe mit Monotherapie bei 1,9 (1,5–2,2), in der Gruppe mit 2 Wirkstoffen bei 1,8 (1,5–2,1), in der Gruppe mit 3 Wirkstoffen bei 1,9 (1,5–2,2), in der Gruppe mit 4 Wirkstoffen bei 3,4 (2,3–4,6) und nach antiglaukomatöser Operation bei 3,3 (3,1–3,6). Das Stadium der Aniridie-assoziierten Keratopathie war signifikant positiv korreliert mit der Anzahl der drucksenkenden Augentropfen ($p < 0,05$) und einer zuvor durchgeführten drucksenkenden Operation ($p < 0,05$). Prostaglandinanaloga waren im Vergleich zu den anderen Wirkstoffgruppen nicht mit einem höherem AAK-Stadium korreliert.

Schlussfolgerungen Im Homburger Aniridie-Zentrum wiesen die Patienten, die eine lokale antiglaukomatöse Vierfachtherapie nutzten oder zuvor antiglaukomatös operiert wurden mit Abstand das höchste AAK-Stadium auf. Die verschiedenen Wirkstoffgruppen hatten keinen Einfluss auf das AAK-Stadium.

Introduction

The introduction as well as the patients and methods section of this study have already been described and published in detail [1]. The authors therefore limit themselves to presenting an abbreviated topic-specific manuscript.

Congenital aniridia is a hereditary bilateral ocular disorder characterized by autosomal dominant inheritance. During a lifetime, over 50% of patients with aniridia will develop aniridia-associated glaucoma. While open-angle glaucoma is more prevalent in individuals with aniridia, there have also been reports of anatomical malformations linked to the underdeveloped iris obstructing the trabecular meshwork [2–4].

Diagnosing and monitoring aniridia-associated glaucoma present challenges due to the presence of keratopathy, nystagmus, and foveal and optic nerve head hypoplasia. Topical glaucoma therapy for aniridia does not significantly differ from general glaucoma treatment; however, the use of preservative-free formulations is recommended. Often, monotherapy alone proves insufficient, necessitating a combination of treatments. In case the effect of conservative treatment is not sufficient, an antiglaucomatous surgical intervention may become necessary [5–8].

Nevertheless, in case of a painless eye, without light perception, there is no more necessity of antiglaucomatous surgery. Since congenital aniridia is characterized by a pathologically altered conjunctiva, a more or less evident inflammatory state and healing disorders due to limbal stem cell insufficiency, the risks of glaucoma surgery are significantly higher, and the interval of surgically induced pressure reduction is usually shorter compared to non-aniridia (glaucoma) patients [6, 8, 9]. In our clinical routine, trabeculotomy is considered the primary option if the anatomy of the chamber angle permits [6, 8]. Otherwise, trabeculectomy is an option, but drainage implants are also effective in lowering intraocular pressure (IOP) [10]. In severely damaged eyes, cyclophotocoagulation can be performed as the last option, whereby aniridia fibrosis syndrome and phthisis bulbi are feared complications [4, 8].

There is limited literature on glaucoma therapy specifically for congenital aniridia and no randomized controlled studies have explored the efficacy of different treatment options and their evi-

dence-based use in this context. The progression of keratopathy further complicates glaucoma follow-up assessments, as it leads to diminishing visual clarity and must, therefore, be taken into consideration when devising a glaucoma treatment plan [8, 9].

The purpose of this cross-sectional study was to compare the different substance classes with regard to their effect on the AAK in order to create more evidence for therapy recommendations.

Patients and Methods

Ethical considerations

Our retrospective single-center study included patients at the Department of Ophthalmology, Saarland University Medical Center in Homburg/Saar, Germany. This study was approved by the Ethics Committee of Saarland/Germany (No 144/15) and followed regulations of the Declaration of Helsinki. Informed consent was obtained from all participants. In case of minors or guardianship, informed consent was obtained from the legal representative or legal guardian.

Diagnosis of glaucoma

As there are some disease specifics to consider when examining aniridia patients, we would like to go into more detail about the convention we use regarding the presence of glaucoma. The classical definition of glaucoma includes the triad of increased IOP, nerve fiber damage, and visual field loss. We also used these criteria in order to define glaucoma disease within our congenital aniridia patients. Nevertheless, we were confronted with several difficulties during examination of congenital aniridia subjects.

Due to nystagmus and large daily fluctuations in ocular surface integrity, the visual field examination is very stressful for aniridia patients without providing satisfactory reproducibility to determine progression. Our examination and clinical evaluation therefore focus mainly on the morphology and morphological changes of the optic nerve head during progression, taking into account IOP. Both increasing optic disc excavation in the presence of increased IOP (over 21 mmHg) and increasing optic disc excavation in the presence of non-increased IOP (neurological cause excluded) are considered by us to be positive for the presence of glaucoma. Taking optic disc photos regularly facilitates the assessment of excavation progression. The measurement of the nerve fiber layer thickness by means of optical coherence tomography is included as a supportive measure. Here, too, it must be borne in mind in congenital aniridia that due to congenital optic nerve anomalies (often hypoplastic), a single measurement does not allow any conclusion to be drawn about increasing damage to the nerve cells, but only the evaluation of several examinations in the course allows this assessment to be made. In this context, morphological assessment and diagnosis should be reserved for an experienced examiner. Frequent changes of the examiner should also be avoided in order not to complicate the assessment of the course. This was taken into account and implemented in our department.

Inclusion criteria, data collection, and examination methods

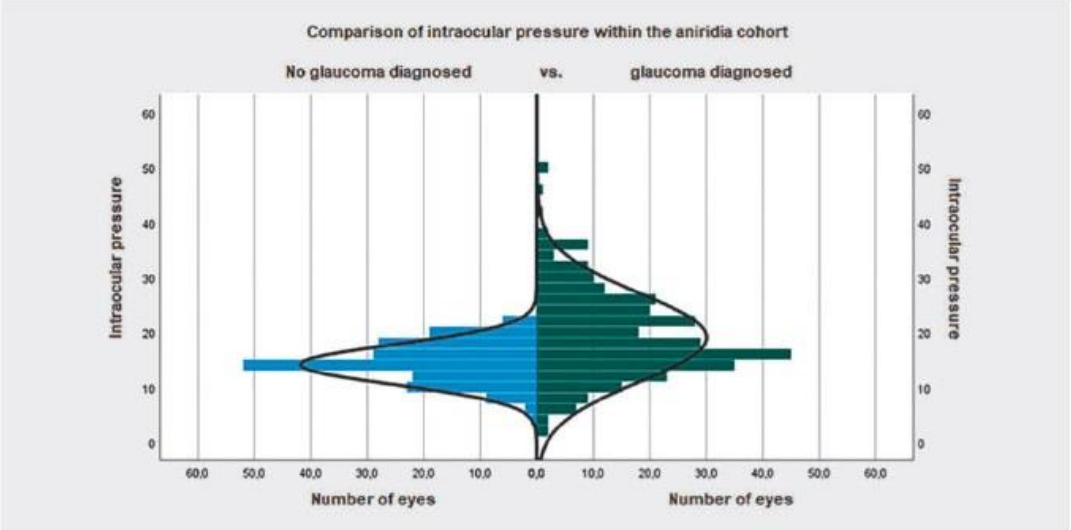
Inclusion criterion was the presence of partial or complete congenital aniridia, visible at slit lamp examination. All subjects underwent a structured ophthalmic examination through the head of the KiOLoN ("Kinderophthalmologie", Orthoptics, Low Vision and Neuroophthalmology) Unit of the Department of Ophthalmology of Saarland University (Prof. Dr. Barbara Käsmann-Kellner). Uncorrected and best-corrected visual acuity (UCVA and BCVA) measurements using Snellen charts, IOP measurement using Goldmann applanation tonometry or iCare (Icare Finland Oy, Vantaa, Finland), and detailed slit lamp and fundus examinations were performed. If there was sufficient cooperation, a measurement was taken using Goldmann applanation tonometry, whereas younger children were more likely to be measured using iCare. For the refinement of the IOP measurement, the Dresden correction table according to Kohlhaas was used to compensate for systematic measurement errors caused by a change in corneal thickness [11].

AAK was classified as follows: Stage 0 (no limbal changes), Stage 1 (conjunctival tissue just crosses the limbal border but remains 1 mm or less from the limbus), Stage 2 (the pannus extends across the peripheral cornea and is typically present in 360 degrees of the cornea), Stage 3 (the pannus invades the central cornea, typically covering the entire cornea with vessels), Stage 4 (the cornea is completely vascularized), Stage 5 (end-stage with an opaque, thick, vascularized cornea) [1,12]. In the evaluation of the eye drops, mono preparations were evaluated as one eye drop, and combination preparations were assigned according to their active ingredients and were accordingly included in the evaluation as two eye drops. This made it possible to evaluate the different substance classes in relation to each other.

All patient data were entered pseudonymized in a Microsoft Access database. Data analysis was performed using IBM SPSS Statistics for Windows Version 29.0 (IBM Corp., Armonk, NY, USA). Descriptive statistics were used to characterize data and assess the distribution of study variables. Categorical variables were summarized in frequencies or percentages. A chi-square test was performed to check for the presence of an association between dependent variables and independent variables, a t-test was used to compare normally distributed variables, and a Pearson correlation was used after adjusting for confounding variables to describe the correlation between the identified risk factors and the progression of AAK. The statistical significance was considered at a p value < 0.05 and a 95% confidence interval (CI).

In collaboration with the Department of Ophthalmology, Saarland University Medical Center in Homburg/Saar (Chair: Prof. Dr. B. Seitz) and the Dr. Rolf M. Schwiete Center for Limbal Stem Cell and Aniridia Research, Homburg/Saar (Chair: Prof. Dr. N. Szentmáry), our aim was to build up a database in order to get a better insight into the pathomechanisms and stage-appropriate treatment options of congenital aniridia.

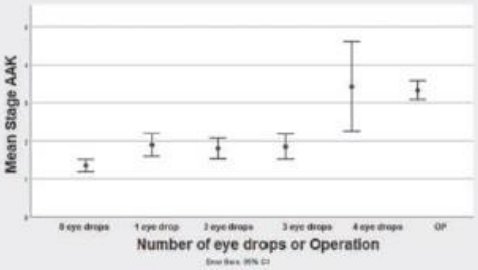
The present study summarizes patient data for subjects examined between June 2003 and January 2022.



▶ **Fig. 1** Combined histogram showing intraocular pressure distribution of subjects at the Homburg Aniridia Center. The patients with glaucoma are shown in green (right), the others are shown in blue (left).

Results

There were 556 eyes of 286 subjects (20.1 ± 20.1 years; 45.5% males) included. In 307 (55.2%) eyes of 163 subjects (27.5 ± 16.3 years; 43.1% males), glaucoma was present at the time of examination. The mean IOP in the glaucoma group was 19.0 mmHg (± 8.0), while in the non-glaucoma group, it was 14.1 mmHg (± 3.6; $p < 0.001$, ▶ **Fig. 1**). In the glaucoma group, 68 (20.5%) patients used topical monotherapy, 51 (16.6%) patients used 2 agents, 41 (13.4%) patients used 3 agents, 7 (2.3%) patients used quadruple therapy, and 140 (45.6%) did not use topical therapy (e.g., after pressure-lowering surgery or pain-free end-stage of glaucoma). Patients were classified according to the following stages of AAK: Stage 0 (96 eyes [17.2%], no keratopathy), Stage 1 (178 eyes [32.0%]), Stage 2 (107 eyes [19.2%]), Stage 3 (67 eyes [12.0%]), Stage 4 (62 eyes [11.1%]), Stage 5 (45 eyes [8.0%]) [1]. The mean stage of AAK was 1.4 (1.2–1.5) in the group without eye drops, 1.9 (1.5–2.2) in the group with monotherapy, 1.8 (1.5–2.1) in the group with 2 drugs, 1.9 (1.5–2.2) in the group with 3 drugs, 3.4 (2.3–4.6) in the group with 4 drugs, and 3.3 (3.1–3.6) after antiglaucomatous surgery. The stage of AAK was significantly positively correlated with the number of pressure-lowering eye drops ($p < 0.05$) and prior antiglaucomatous surgery ($p < 0.05$; (▶ **Fig. 2**). Even after correction for age, there was still a significant correlation between the number of eye drops and AAK stage ($r = 0.166$ and $p < 0.001$, Pearson correlation). After previous corneal surgery (PKP, PTK, AMT, pannus abrasion), no correlation between glaucoma therapy and AAK stage was found (▶ **Fig. 3**). Prostaglandin analogues were not correlated with a higher AAK stage compared to the other drug groups, despite their pro-inflammatory side effect ($p > 0.05$).

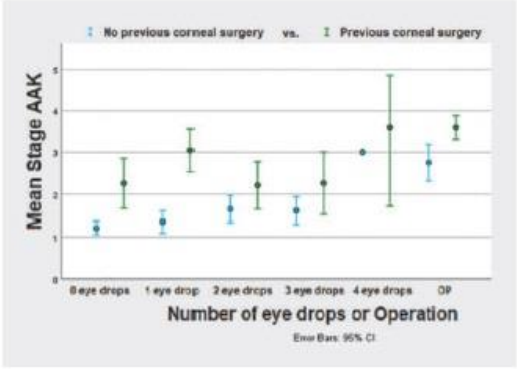


▶ **Fig. 2** Comparison of stage of aniridia-associated keratopathy (AAK) considering the glaucoma therapy.

Discussion

Keratopathy and glaucoma are considered the main causes of secondary blindness in the context of congenital aniridia. Both pathologies are progressive and require different therapeutic approaches [5–9, 13]. So far, there are only a few studies dealing with secondary glaucoma in the context of congenital aniridia [14, 15]. The incidence of glaucoma in aniridia is reported to be 50–75% [3, 16]. Our experience confirms these descriptions also in the present larger cohort, with an incidence of 55.2% regarding the presence of glaucoma overall.

Limbal stem cell insufficiency and altered ocular surface as well as a continuous inflammatory processes are discussed as the cause of the progression of keratopathy [17–25]. Therefore,



► **Fig. 3** Comparison of stage of aniridia-associated keratopathy (AAK) considering the glaucoma therapy correcting for previous corneal surgery (PKP, PTK, AMT, pannus abrasion). The patients who did not have any previous corneal surgery are represented in blue, the patients who did have previous corneal surgery before assessment are represented in green.

there is a consensus among experts to use preservative-free preparations for topical therapy, if possible, in order to avoid aggravation of the inflammatory state by proinflammatory additives such as benzalkonium chloride [6–8, 12].

Despite these precautions, more intensive pressure-lowering topical therapy seems to be associated with a higher stage of AAK. However, no comparative studies are currently available that would allow a closer discussion against the background of already published literature.

Prostaglandin analogues have established themselves as the most effective substance class in general glaucoma therapy but are well known for their undesirable proinflammatory effects. Interestingly, no difference between the individual substance classes (beta-blockers, alpha-agonists, carbonic anhydrase inhibitors, prostaglandin analogues) and AAK could be demonstrated in the present study. It should be taken into account that due to the division of our cohort into smaller subgroups, small differences may no longer be evident for statistical reasons. For this reason, the available data do not allow us to make a reliable statement about a causative impact of a pressure-lowering substance class on the stage of AAK.

However, a closer look at our larger group of patients suggests that there is a correlation between advanced glaucoma or the need for more intensive pressure-lowering therapy and the progression of keratopathy. A significant difference was found for the number of pressure-lowering eye drops, whereby patients who were not (or no longer) dependent on topical therapy or who did not apply eye drops due to noncompliance showed the least pronounced AAK. No significant difference was found between monotherapy, double therapy, and triple therapy. However, it should be pointed out that larger numbers of cases and further studies are necessary to be able to make a reliable statement. The most advanced keratopathy was seen in patients with quadruple therapy and after antiglaucomatous surgery. This clinical observation alone does not yet allow us to draw any conclusions about the pathomechanism and can therefore only be seen as a suggestion for future basic research studies. In addition, a specific approach is necessary in order to verify glaucoma progression in time. Due to the very small number of patients, a further differentiation between the surgical procedures (e.g., trabeculotomy, trabeculectomy, cyclophotocoagulation, drainage implant) is currently not considered useful and is therefore not discussed further.

Our goal is to include more patients in our registry and to validate the previously observed trends with larger numbers of cases and in the long-term follow-up in terms of a longitudinal study outline.

CONCLUSION BOX

Already known:

- In congenital aniridia, there is an increased risk of developing blindness during life.
- AAK and glaucoma are the most common causes of blindness in congenital aniridia.
- In order to develop better treatment options in congenital aniridia, establishment of an aniridia register is necessary.

Newly described:

- A more intensive pressure-lowering topical therapy is associated with a higher stage of AAK.
- AAK stage of patients that had previously undergone glaucoma surgery was comparable to patients who used antiglaucomatous topical quadruple therapy.
- Our aniridia register will support further validation of previously observed trends with growing numbers of cases.

Conclusions:

At the Homburg Aniridia Center, patients using topical antiglaucomatous quadruple therapy or who had previously undergone pressure-lowering surgery had the highest AAK stage. The different drug substance groups had no influence on the AAK stage. Our registry will allow further detailed analysis of ophthalmic and systemic disease in patients with congenital aniridia over the long term.

Conflict of Interest

The authors declare that they have no conflict of interest.

Acknowledgements

The work of Dr. Szentmáry at the Dr. Rolf M. Schwiete Center was generously supported by the Dr. Rolf M. Schwiete Foundation, Mannheim. This article is based upon work from COST Action CA18116, supported by COST (European Cooperation in Science and Technology).

References

[1] Fries FN, Näray A, Munteanu C et al. A Cross-sectional Analysis of 556 Eyes Entering the Homburg Aniridia Centre. Klin Monbl Augenheilkd 2023; 240: 1–8. doi:10.1055/a-2065-8405

[2] Käsmann-Kellner B, Latta L, Fries FN et al. Diagnostic impact of anterior segment angiography of limbal stem cell insufficiency in PAX6-related aniridia. Clin Anat 2018; 31: 392–397. doi:10.1002/ca.22987

[3] Muñoz-Negrete FJ, Teus MA, García-Feijoó J et al. Aniridic glaucoma: An update. Arch Soc Esp Oftalmol (Engl Ed) 2021; 96 (Suppl. 1): S52–S59. doi:10.1016/j.oftale.2020.11.011

[4] Viestenz A, Seitz B, Deland E et al. Clinical anatomy of the anterior chamber angle in congenital aniridia and consequences for trabeculotomy/cyclophotocoagulation. Clin Anat 2018; 31: 64–67. doi:10.1002/ca.22935

[5] Käsmann-Kellner B, Fries FN, Latta L et al. Aniridie bei PAX6-Syndrom: Eine potenziell zur Erblindung führende Erkrankung. Etablierung eines Deutschen Aniridie-Registers. Concept Ophthalmologie 2019 (05)

[6] Käsmann-Kellner B, Seitz B. [Aniridia syndrome: clinical findings, problematic courses and suggestions for optimization of care ("aniridia guide")]. Ophthalmologe 2014; 111: 1145–1156. doi:10.1007/s00347-014-3060-x

[7] Käsmann-Kellner B, Seitz B. [Congenital aniridia or PAX6 syndrome]. Ophthalmologe 2014; 111: 1144. doi:10.1007/s00347-014-3058-4

[8] Seitz B, Käsmann-Kellner B, Viestenz A. [Stage-related therapy of congenital aniridia]. Ophthalmologe 2014; 111: 1164–1171. doi:10.1007/s00347-014-3061-9

[9] Farah CJ, Fries FN, Latta L et al. An attempt to optimize the outcome of penetrating keratoplasty in congenital aniridia-associated keratopathy (AAK). Int Ophthalmol 2021; 41: 4091–4098. doi:10.1007/s10792-021-01982-z

[10] Soyugelen Demirok G, Ekşioğlu Ü, Yakin M et al. Short- and Long-term Results of Glaucoma Valve Implantation for Aniridia-related Glaucoma: A Case Series and Literature Review. Turk J Ophthalmol 2019; 49: 183–187. doi:10.4274/tjo.galenos.2019.07348

[11] Kohlhaas M, Boehm AG, Spoerl E et al. Effect of central corneal thickness, corneal curvature, and axial length on applanation tonometry. Arch Ophthalmol 2006; 124: 471–476. doi:10.1001/archopht.124.4.471

[12] Landsend ECS, Lagali N, Utheim TP. Congenital aniridia – A comprehensive review of clinical features and therapeutic approaches. Surv Ophthalmol 2021; 66: 1031–1050. doi:10.1016/j.survophthal.2021.02.011

[13] van Velthoven AJH, Utheim TP, Notara M et al. Future directions in managing aniridia-associated keratopathy. Surv Ophthalmol 2023; 68: 940–956. doi:10.1016/j.survophthal.2023.04.003

[14] Grainger RM, Lauderdale JD, Collins JL et al. Report on the 2021 Aniridia North America symposium on PAX6, aniridia, and beyond. Ocul Surf 2023; 29: 423–431. doi:10.1016/j.jtos.2023.05.010

[15] Gupta V, Somarajan BI, Gupta S et al. A new association of PAX6 variation with Juvenile onset open angle glaucoma. J Hum Genet 2023; 68: 355–358. doi:10.1038/s10038-022-01115-z

[16] Bremond-Gignac D. [Glaucoma in aniridia]. J Fr Ophtalmol 2007; 30: 196–199. doi:10.1016/s0181-5512(07)89576-3

[17] Fries FN, Moslemani K, Utheim TP et al. Early ocular surface and tear film status in congenital aniridia indicates a supportive treatment window. Br J Ophthalmol 2022. doi:10.1136/bjo-2021-320774

[18] Katiyar P, Stachon T, Fries FN et al. Decreased FABP5 and DSG1 protein expression following PAX6 knockdown of differentiated human limbal epithelial cells. Exp Eye Res 2022; 215: 108904. doi:10.1016/j.exer.2021.108904

[19] Lagali N, Wowra B, Fries FN et al. PAX6 Mutational Status Determines Aniridia-Associated Keratopathy Phenotype. Ophthalmology 2020; 127: 273–275. doi:10.1016/j.ophtha.2019.09.034

[20] Lagali N, Wowra B, Fries FN et al. Early phenotypic features of aniridia-associated keratopathy and association with PAX6 coding mutations. Ocul Surf 2020; 18: 130–140. doi:10.1016/j.jtos.2019.11.002

[21] Latta L, Figueiredo FC, Ashery-Padan R et al. Pathophysiology of aniridia-associated keratopathy: Developmental aspects and unanswered questions. Ocul Surf 2021; 22: 245–266. doi:10.1016/j.jtos.2021.09.001

[22] Latta L, Knebel I, Bleil C et al. Similarities in DSG1 and KRT3 Downregulation through Retinoic Acid Treatment and PAX6 Knockdown Related Expression Profiles: Does PAX6 Affect RA Signaling in Limbal Epithelial Cells? Biomolecules 2021; 11: 1651. doi:10.3390/biom11111651

[23] Latta L, Ludwig N, Krammes L et al. Abnormal neovascular and proliferative conjunctival phenotype in limbal stem cell deficiency is associated with altered microRNA and gene expression modulated by PAX6 mutational status in congenital aniridia. Ocul Surf 2021; 19: 115–127. doi:10.1016/j.jtos.2020.04.014

[24] Latta L, Nordström K, Stachon T et al. Expression of retinoic acid signaling components ADH7 and ALDH1A1 is reduced in aniridia limbal epithelial cells and a siRNA primary cell based aniridia model. Exp Eye Res 2019; 179: 8–17. doi:10.1016/j.exer.2018.10.002

[25] Schlötzer-Schrehardt U, Latta L, Giessl A et al. Dysfunction of the limbal epithelial stem cell niche in aniridia-associated keratopathy. Ocul Surf 2021; 21: 160–173. doi:10.1016/j.jtos.2021.06.002

Literaturverzeichnis

1. Latta L, Figueiredo FC, Ashery-Padan R, et al. Pathophysiology of aniridia-associated keratopathy: Developmental aspects and unanswered questions. Ocul Surf. 2021; 22:245-266.
2. Lagali N, Wowra B, Fries FN, et al. Early phenotypic features of aniridia-associated keratopathy and association with PAX6 coding mutations. Ocul Surf. 2020;18(1):130-140.
3. Landsend ECS, Lagali N, Utheim TP. Congenital aniridia - A comprehensive review of clinical features and therapeutic approaches. Surv Ophthalmol. 2021;66(6):1031-1050.
4. Käsmann-Kellner B, Seitz B. Aniridiesyndrom: Klinische Befunde, problematische Verläufe und Vorschlag zur Betreuungsoptimierung ("Aniridielotse") [Aniridia syndrome: clinical findings, problematic courses and suggestions for optimization of care ("aniridia guide")]. Ophthalmologe. 2014;111(12):1145-1156.
5. van Velthoven AJH, Utheim TP, Notara M et al. Future directions in managing aniridia-associated keratopathy. Surv Ophthalmol. 2023 Sep-Oct;68(5):940-956.
6. D'Oria F, Barraquer R, Alio JL. Crystalline lens alterations in congenital aniridia. Arch Soc Esp Oftalmol (Engl Ed). 2021;96 Suppl 1:38-51.
7. Eden U, Lagali N, Dellby A, et al. Cataract development in Norwegian patients with congenital aniridia. Acta Ophthalmol. 2014;92(2):e165–7.
8. Náray A, Csidey M, Kéki-Kovács K, Németh O, Knézy K, Bausz M, Szigeti A, Csorba A, Kormányos K, Szabó D, Stachon T, Corton M, Tory K, Nagy ZZ, Maka E, Szentmáry N. [Congenital aniridia - Hungarian data of a spectrum disease]. Orv Hetil. 2023 Jan 29;164(4):148-155.
9. Náray A, Fries FN, Csidey M, Kéki-Kovács K, Németh O, Knézy K, Bausz M, Szigeti A, Csorba A, Kormányos K, Szabó D, Corton M, Tory K, Nagy ZZ, Maka E, Szentmáry N. [Staging of aniridia-associated keratopathy]. Orv Hetil. 2023 Jul 9;164(27):1063-1069.
10. Tsai JH, Freeman JM, Chan CC, et al. A progressive anterior fibrosis syndrome in patients with postsurgical congenital aniridia. Am J Ophthalmol. 2005;140(6):1075-1079.

11. Kothari M, Rao K, Moolani S. Recurrent progressive anterior segment fibrosis syndrome following a descemet-stripping endothelial keratoplasty in an infant with congenital aniridia. Indian J Ophthalmol. 2014;62(2):246-248.

12. Bakhtiari P, Chan C, Welder JD, de la Cruz J, Holland EJ, Djalilian AR. Surgical and visual outcomes of the type I Boston Keratoprosthesis for the management of aniridic fibrosis syndrome in congenital aniridia. Am J Ophthalmol. 2012;153(5):967-971.

13. Käsmann-Kellner B, Latta L, Fries FN, Viestenz A, Seitz B. Diagnostic impact of anterior segment angiography of limbal stem cell insufficiency in PAX6-related aniridia. Clin Anat. 2018 Apr;31(3):392-397.

14. Farah CJ, Fries FN, Latta L, Käsmann-Kellner B, Seitz B. An attempt to optimize the out-come of penetrating keratoplasty in congenital aniridia-associated keratopathy (AAK). Int Ophthalmol. 2021 Dec;41(12):4091-4098.

15. Fries FN, Náray A, Munteanu C, Stachon T, Lagali N, Seitz B, Szentmáry N, Käsmann-Kellner B. A cross-sectional analysis of 556 eyes entering The Homburg Aniridia Center. Klin Monbl Augenheilkd 2023 Aug 30. English. doi: 10.1055/a-2065-8405. Epub ahead of print.

16. Fries FN, Náray A, Munteanu C, Stachon T, Lagali N, Seitz B, Käsmann-Kellner B, Szentmáry N. The effect of glaucoma treatment on aniridia-associated keratopathy (AAK) - A report from the Homburg Register for Congenital Aniridia. Klin Monbl Augenheilkd 2023 2023 Oct 18. German. doi: 10.1055/a-2194-1580. Epub ahead of print.

Publikationsverzeichnis

Veröffentlichungen im Zusammenhang mit der Dissertation

1 Farah CJ, Fries FN, Latta L, Käsmann-Kellner B, Seitz B. An attempt to optimize the outcome of penetrating keratoplasty in congenital aniridia-associated keratopathy (AAK). Int Ophthalmol. 2021 Dec;41(12):4091-4098.

2 Fries FN, Náray A, Munteanu C, Stachon T, Lagali N, Seitz B, Szentmáry N, Käsmann-Kellner B. A cross-sectional analysis of 556 eyes entering The Homburg Aniridia Center. Klin Monbl Augenheilkd 2023 2023 Aug 30. English. doi: 10.1055/a-2065-8405. Epub ahead of print.

3 Fries FN, Náray A, Munteanu C, Stachon T, Lagali N, Seitz B, Käsmann-Kellner B, Szentmáry N. The effect of glaucoma treatment on aniridia-associated keratopathy (AAK) - A report from the Homburg Register for Congenital Aniridia. Klin Monbl Augenheilkd 2023 Oct 18. German. doi: 10.1055/a-2194-1580. Epub ahead of print.

Andere Veröffentlichungen

1 Fries FN, Berger T, Fraenkel D, Seitz B, Suffo S. Subtotale traumatische Linsenluxation in die Vorderkammer [Subtotal traumatic lens luxation into the anterior chamber]. Ophthalmologie. 2023 Nov 9. German. doi: 10.1007/s00347-023-01952-z.

2 Ax T, Ganse B, Fries FN, Szentmáry N, de Paiva CS, March de Ribot F, Jensen SO, Seitz B, Millar TJ. Dry eye disease in astronauts: a narrative review. Front Physiol. 2023 Oct 19;14:1281327. doi: 10.3389/fphys.2023.1281327.

3 Chan C, Fries FN, Käsmann-Kellner B, Seitz B, Abdin AD. Grubenpapillen-Makulopathie: Inzidenz und klinischer Verlauf [Optic pit maculopathy: incidence and clinical course]. Ophthalmologie. 2023 Dec;120(12):1267-1272.

4 Náray A, Fries FN, Csidey M, Kéki-Kovács K, Németh O, Knézy K, Bausz M, Szigeti A, Csorba A, Kormányos K, Szabó D, Corton M, Tory K, Nagy ZZ, Maka E, Szentmáry N. [Staging of aniridia-associated keratopathy]. Orv Hetil. 2023 Jul 9;164(27):1063-1069.

5 Mühl-Benninghaus R, Fries FA, Reith W, Hendrix P, Seitz B, Fries FN. Acute bilateral vision loss after endovascular treatment of an intracranial aneurysm. Wien Med Wochenschr. 2023 Apr 20. doi: 10.1007/s10354-023-01015-7. Epub ahead of print.

6 Fries FN, Moslemani K, Utheim TP, Seitz B, Käsmann-Kellner B, Lagali NS. Early ocular surface and tear film status in congenital aniridia indicates a supportive treatment window. Br J Ophthalmol. 2023 Dec 18;108(1):30-36. doi: 10.1136/bjo-2021-320774.

7 Vandebroek AC, Saternus R, Becker SL, Fries FN, Pérez Guerra N, Rickmann A. Ulzerierende Raumforderung am Oberlid als Manifestation einer primären kutanen CD30+-T-Zell-lymphoproliferativen Erkrankung [Ulcerating mass on the upper eyelid as manifestation of a primary CD30+T-cell lymphoproliferative disease]. Ophthalmologie. 2023 Aug;120(8):847-850.

8 Vandebroek AC, Rickmann A, Boden K, Szurman G, Bozzato V, Seitz B, Fries FN. Erratum zu: Therapierefraktäre Epiphora nach Dakryozystorhinostomie – Stellenwert von Bildgebung und Histopathologie. Ophthalmologie. 2023 Jan;120(Suppl 1):68.

9 Katiyar P, Stachon T, Fries FN, Parow F, Ulrich M, Langenbucher A, Cayless A, Seitz B, Käsmann-Kellner B, Latta L, Szentmáry N. Decreased FABP5 and DSG1 protein expression following PAX6 knockdown of differentiated human limbal epithelial cells. Exp Eye Res. 2022 Feb;215:108904.

10 Latta L, Knebel I, Bleil C, Stachon T, Katiyar P, Zussy C, Fries FN, Käsmann-Kellner B, Seitz B, Szentmáry N. Similarities in DSG1 and KRT3 Downregulation through Retinoic Acid Treatment and PAX6 Knockdown Related Expression Profiles: Does PAX6 Affect RA Signaling in Limbal Epithelial Cells? Biomolecules. 2021 Nov 8;11(11):1651.

11 Maamri A, Fries FN, Spira-Eppig C, Eppig T, Seitz B. Mitarbeiterbefragung nach Einführung der elektronischen Patientenakte FIDUS an der Universitätsaugenklinik des Saarlandes [Employee survey after introduction of the FIDUS electronic patient file at the Saarland University Eye Hospital]. Ophthalmologe. 2022 May;119(5):471-480.

12 Weinstein I, Fries FN, Hartmann M, Abdin AD, Seitz B. Ausgeprägte Bandkeratopathie bei Refsum-Syndrom [Pronounced band keratopathy in Refsum‘s syndrome]. Ophthalmologie. 2022 Jul;119(7):741-743.

13 Schlötzer-Schrehardt U, Latta L, Gießl A, Zenkel M, Fries FN, Käsmann-Kellner B, Kruse FE, Seitz B. Dysfunction of the limbal epithelial stem cell niche in aniridia-associated keratopathy. Ocul Surf. 2021 Jul;21:160-173.

14 Abdin AD, Suffo S, Fries FN, Kaymak H, Seitz B. Einheitliche Klassifikation des pachychorioidalen Krankheitsspektrums [Uniform classification of the pachychoroid spectrum disorders]. Ophthalmologe. 2021 Aug;118(8):865-878.

15 Shi L, Fries FN, Xanthopoulou K, Stachon T, Daas L, Zemova E, Langenbucher A, Seitz B, Szentmáry N. Correction: Endothelial Cell Density and Central Corneal Thickness following Penetrating Keratoplasty of Acanthamoeba Keratitis Patients - A Retrospective Cross-Sectional Observational Study. Klin Monbl Augenheilkd. 2021 Apr 14.

16 Shi L, Fries FN, Xanthopoulou K, Stachon T, Daas L, Zemova E, Langenbucher A, Seitz B, Szentmáry N. Endothelial Cell Density and Central Corneal Thickness following Penetrating Keratoplasty of Acanthamoeba Keratitis Patients – A Retrospective Cross-Sectional Observational Study. Klin Monbl Augenheilkd. 2022 Aug;239(8):1007-1012.

17 Bedersdorfer M, Rickmann A, Bisorca-Gassendorf L, Szurman P, Boden KT, Seitz B, Fries FN. Intravenöse Fibrinolyse bei einem nichtarteriitischen Zentralarterienverschluss – eine Behandlungsoption? [Intravenous fibrinolysis for nonarteritic central retinal artery occlusion-a treatment option?]. Ophthalmologe. 2022 Jan;119(Suppl 1):98-101.

18 Fries FN, Hendrix P, Abdin AD, Seitz B, Sourlis C, Fries FA, Mühl-Benninghaus R. Akuter binokularer Visusverlust bei Basilarisaneurysma-bedingter Subarachnoidalblutung [Acute binocular visual loss due to basilar artery aneurysm-induced subarachnoid hemorrhage]. Ophthalmologe. 2021 Oct;118(10):1054-1056.

19 Wykrota AA, Löw U, Fries FN, Seitz B, Abdin AD. Unilaterale makuläre Pigmentepithelunregelmäßigkeiten bei einem 38-jährigen Patienten – „Akute retinale Pigmentepitheliitis“ [Unilateral Irregularities in the Macular Pigment Epithelium in a 38-Year-Old Patient – „Acute Retinal Pigment Epitheliitis“]. Klin Monbl Augenheilkd. 2022 Mar;239(3):304-307.

20 Szentmáry N, Fries FN, Daas L, Shi L, Laurik KL, Langenbucher A, Seitz B. Chamäleonartige Hornhautveränderungen: Akanthamöbenkeratitis [Chameleon-Like Corneal Disorders: Acanthamoeba Keratitis]. Klein Monbl Augenheilkd. 2020 Jun;237(6):754-760.

21 Latta L, Ludwig N, Krammes L, Stachon T, Fries FN, Mukwaya A, Szentmáry N, Seitz B, Wowra B, Kahraman M, Keller A, Meese E, Lagali N, Käsmann-Kellner B. Abnormal neovascular and proliferative conjunctival phenotype in limbal stem cell deficiency is associated with altered microRNA and gene expression modulated by PAX6 mutational status in congenital aniridia. Ocul Surf. 2021 Jan;19:115-127.

22 Weinstein I, Fries FN, Szentmáry N, Seitz B, Daas L. Ausgeprägter Wessely-Immunring bei Keratitis – ein Chamäleon [Distinctive Wessely immune ring in keratitis-a chameleon]. Ophthalmologe. 2021 Jan;118(1):53-55.

23 Shi L, Hager T, Fries FN, Daas L, Holbach L, Hofmann-Rummelt C, Zemova E, Seitz B, Szentmáry N. Reactive uveitis, retinal vasculitis and scleritis as ocular end-stage of Acanthamoeba keratitis: a histological study. Int J Ophthalmol. 2019 Dec 18;12(12):1966-1971.

24 Lagali N, Wowra B, Fries FN, Latta L, Moslemani K, Utheim TP, Wylegala E, Seitz B, Käsmann-Kellner B. Early phenotypic features of aniridia-associated keratopathy and association with PAX6 coding mutations. Ocul Surf. 2020 Jan;18(1):130-140.

25 Lagali N, Wowra B, Fries FN, Latta L, Moslemani K, Utheim TP, Wylegala E, Seitz B, Käsmann-Kellner B. PAX6 Mutational Status Determines Aniridia-Associated Keratopathy Phenotype. Ophthalmology. 2020 Feb;127(2):273-275.

26 Boden KT, Rickmann A, Fries FN, Xanthopoulou K, Alnaggar D, Januschowski K, Seitz B, Käsmann-Kellner B, Schrecker J. Evaluierung eines VR-Simulators zum Erlernen der direkten Ophthalmoskopie in der studentischen Lehre [Evaluation of a virtual reality simulator for learning direct ophthalmoscopy in student teaching]. Ophthalmologe. 2020 Jan;117(1):44-49.

27 Latta L, Nordström K, Stachon T, Langenbucher A, Fries FN, Szentmáry N, Seitz B, Käsmann-Kellner B. Expression of retinoic acid signaling components ADH7 and ALDH1A1 is reduced in aniridia limbal epithelial cells and a siRNA primary cell based aniridia model. Exp Eye Res. 2019 Feb;179:8-17.

28 Fries FN, Pattmöller M, Seitz B, Berger F, Kampen H, Szentmáry N, Becker SL. Ophthalmomyiasis externa due to Oestrus ovis in a traveller returning from Greece. Travel Med Infect Dis. 2018 May-Jun;23:101-102.

29 Gyenes A, Orosz E, Sándor GL, Fries FN, Seitz B, Zoltán NZ, Szentmáry N. Frühdiagnose und erfolgreiche medikamentöse Therapie bei Akanthamöbenkeratitis [Early Diagnosis and Successful Medical Treatment of Acanthamoeba Keratitis]. Klin Monbl Augenheilkd. 2018 Dec;235(12):1407-1410.

30 Manouvakhova OV, Macchi V, Fries FN, Loukas M, De Caro R, Oskouian RJ, Spinner RJ, Tubbs RS. Landmarks for Identifying the Suprascapular Foramen Anteriorly: Application to Anterior Neurotization and Decompressive Procedures. Oper Neurosurg (Hagerstown). 2018 Feb 1;14(2):166-170.

31 Faria BL, Brieske CM, Cosgarea I, Omlor AJ, Fries FN, de Faria COM, Lino HA, Oliveira ACC, Lisboa OC, Klode J, Schadendorf D, Bernardes-Souza B, Brinker TJ. A smoking prevention photoageing intervention for secondary schools in Brazil delivered by medical students: protocol for a randomised trial. BMJ Open. 2017 Dec 10;7(12):e018589.

32 Wang JM, Fries FN, Hendrix P, Brinker T, Loukas M, Tubbs RS. Orbital Blowout Fracture with Complete Dislocation of the Globe into the Maxillary Sinus. Cureus. 2017 Sep 29;9(9):e1728.

33 Käsmann-Kellner B, Latta L, Fries FN, Viestenz A, Seitz B. Diagnostic impact of anterior segment angiography of limbal stem cell insufficiency in PAX6-related aniridia. Clin Anat. 2018 Apr;31(3):392-397.

34 Montibeller GR, Hendrix P, Fries FN, Becker KW, Oertel J. Comparison of microscopic and endoscopic view 40ft he internal acoustic meatus: A cadaveric study. Clin Anat. 2018 Apr;31(3):398-403.

35 Fries FN, Suffo S, Daas L, Seitz B, Fiorentzis M, Viestenz A. Tenonplasty for closing defects during sclerocorneal surgery-A brief review of its anatomy and clinical applications. Clin Anat. 2018 Jan;31(1):72-76.

36 Brinker TJ, Owczarek AD, Seeger W, Groneberg DA, Brieske CM, Jansen P, Klode J, Stoffels I, Schadendorf D, Izar B, Fries FN, Hofmann FJ. A Medical Student-Delivered Smoking Prevention Program, Education Against Tobacco, for Secondary Schools in Germany: Randomized Controlled Trial. J Med Internet Res. 2017 Jun 6;19(6):e199.

37 Daas L, Viestenz A, Schnabel PA, Fries FN, Hager T, SzentmÁry N, Seitz B. Confocal microscopy as an early relapse marker for acanthamoeba keratitis. Clin Anat. 2018 Jan;31(1):60-63. doi: 10.1002/ca.22925.

38 Iwanaga J, Granger A, Vahedi P, Loukas M, Oskouian RJ, Fries FN, Lotfinia I, Mortazavi MM, Oakes WJ, Tubbs RS. Mapping the Internal Anatomy of the Lateral Brainstem: Anatomical Study with Application to Far Lateral Approaches to Intrinsic Brainstem Tumors. Cureus. 2017 Feb 3;9(2):e1010.

39 Fries FN, Hendrix P, Brinker TJ, Loukas M, Tubbs RS. Otto Mennicke (1876-) and the first description of skull base anomalies causing cerebellar tonsillar ectopia: one of the first mentions of the Chiari I malformation. Childs Nerv Syst. 2017 May;33(5):825-827.

40 Tubbs RS, Maldonado AA, Stoves Y, Fries FN, Li R, Loukas M, Oskouian RJ, Spinner RJ. A novel method of lengthening the accessory nerve for direct coaptation during nerve repair and nerve transfer procedures. J Neurosurg. 2018 Jan;128(1):272-276.

41 Xavier LE, Bernardes-Souza B, Lisboa OC, Seeger W, Groneberg DA, Tran TA, Fries FN, Corrêa PC, Brinker TJ. A Medical Student-Delivered Smoking Prevention Program, Education Against Tobacco, for Secondary Schools in Brazil: Study Protocol for a Randomized Trial. JMIR Res Protoc. 2017 Jan 30;6(1):e16.

42 Brinker TJ, Holzapfel J, Baudson TG, Sies K, Jakob L, Baumert HM, Heckl M, Cirac A, Suhre JL, Mathes V, Fries FN, Spielmann H, Rigotti N, Seeger W, Herth F, Groneberg DA, Raupach T, Gall H, Bauer C, Marek P, Batra A, Harrison CH, Taha L, Owczarek A, Hofmann FJ, Thomas R, Mons U, Kreuter M. Photoaging smartphone app promoting poster campaign to reduce smoking prevalence in secondary schools: the Smokerface Randomized Trial: design and baseline characteristics. BMJ Open. 2016 Nov 7;6(11):e014288.

43 Fries FN, Youssef P, Irwin PA, Tubbs RI, Loukas M, Tubbs RS. Comparing the left and right Whitnall‘s tubercles and their relation 41ot he frontozygomatic suture: Application to symmetry following lateral orbital surgery. Orbit. 2016 Dec;35(6):305-308.

44 Tubbs RS, Fries FN, Kulwin C, Mortazavi MM, Loukas M, Cohen-Gadol AA. Modified skin incision for avoiding the lesser occipital nerve and occipital artery during retrosigmoid craniotomy: potential applications for enhancing operative working distance and angles while minimizing the risk of postoperative neuralgias and intraoperative hemorrhage. J Clin Neurosci. 2016 Oct;32:83-7.

45 Tubbs RS, Ajayi OO, Fries FN, Spinner RJ, Oskouian RJ. Variations of the accessory nerve: anatomical study including previously undocumented findings-expanding our misunderstanding of this nerve. Br J Neurosurg. 2017 Feb;31(1):113-115.

46 Ivashchuk G, Fries FN, Loukas M, Paulson D, Monteith SJ, Chapman JR, Oskouian RJ, Tubbs RS. Arterial variations around the atlas: a comprehensive review for avoiding neurosurgical complications. Childs Nerv Syst. 2016 Jun;32(6):1093-100.

47 Tubbs RS, Malefant J, Loukas M, Jerry Oakes W, Oskouian RJ, Fries FN. Enigmatic human tails: A review of their history, embryology, classification, and clinical manifestations. Clin Anat. 2016 May;29(4):430-8.

48 Fries FN, Bischoff M, Käsmann-Kellner B, Hager T, Seitz B, Viestenz A. Akuter monokularer Visusverlust bei kindlicher akuter lymphatischer Leukämie [Acute monocular visual loss in a child with acute lymphoblastic leukemia]. Ophthalmologe. 2016 Aug;113(8):712-4.

Danksagung

Zuerst bin ich meiner Doktormutter, Frau Prof. Dr. Nóra Szentmáry zu besonderem Dank für die Vergabe des Themas, für die Betreuung der Durchführung und wissenschaftliche Aufarbeitung dieser Arbeit und dafür, dass sie mir stets mit Rat und Tat hilfreich zur Seite stand, verpflichtet.

Danken möchte ich ebenfalls besonders Herrn Prof. Dr. med. Berthold Seitz und Frau Prof. Dr. med. Barbara Käsmann-Kellner für die Möglichkeit, dass ich mich der Aniridie-Forschung widmen durfte. Ihre langjährige klinische Tätigkeit war die Grundlage für diese Arbeit. Vor allem bedanke ich mich bei Frau Prof. Käsmann-Kellner, die mir bei Fragestellungen stets freundlich und hilfsbereit zur Seite stand.

Insbesondere möchte ich mich bei Herrn Cristian Munteanu für die Unterstützung bei der statistischen Auswertung und Beratung zur Auswahl der geeigneten Testverfahren bedanken.

Mein Dank gilt allen Kollegen des Dr. Rolf M. Schwiete Zentrums der Universität des Saarlandes und der Klinik für Augenheilkunde am Universitätsklinikum des Saarlandes, die diese Arbeit unterstützt haben.

Ein besonderer Dank geht an meine Familie und meine Freunde für die Unterstützung während der Dissertation sowie dafür, dass sie mich in meinem Handeln immer bestärkt haben.