UNIVERSITE DE NANTES

– – – – –

FACULTE DE MEDECINE

Année 2016-2017 N° 121

T H E S E

pour le

DIPLOME D'ETAT DE DOCTEUR EN MEDECINE

par

Aurélie Najm

née le 09 février 1988 à
Saint-Sébastien sur-Loire.

– – – –

Présentée et soutenue publiquement le 11 Septembre 2017.

– – – –

Qualité et utilité des biopsies synoviales échoguidées réalisées en pratique courante.

– – – –

Président du jury : Monsieur le Professeur Yves MAUGARS

Directeur de thèse : Monsieur le Professeur Benoît LE GOFF

COMPOSITION DU JURY

Président du jury :

Monsieur le Professeur Yves MAUGARS

Directeur de thèse :

Monsieur le Professeur Benoît LE GOFF

Membres du jury :

Monsieur le Professeur François Gouin

Madame le Docteur Anne Moreau

Monsieur le Docteur Jean-Marie Berthelot

REMERCIEMENTS

A Monsieur le Professeur Benoît Le Goff

Tout d'abord un grand merci pour nos premiers échanges qui m'ont décidée à choisir la Rhumatologie comme spécialité.
Merci pour ton accompagnement, ton soutien et pour l'ensemble de tes conseils avisés dans ces multiples projets développés tout au long de mon internat, et aussi dans les projets à venir.
Merci de m'avoir donné l'opportunité de ce stage à Dublin, qui aura été une expérience humaine et professionnelle hors du commun et m'aura ouvert des portes à l'échelon européen.
Merci enfin pour m'avoir formée à la Rhumatologie et à l'échographie.

A Monsieur le Professeur Yves Maugars,

Merci pour m'avoir formée à la Rhumatologie et pour tout ce que vous m'avez enseigné au cours de ces 4 années d'internat, que ce soit en visite, en consultation ou en staff. Merci aussi pour m'avoir soutenue dans les différents projets débutés au cours de mon internat. Merci enfin de la confiance que vous m'accordez en me proposant un poste de chef de clinique dans votre service.

A Monsieur le Docteur Jean-Marie Berthelot

Merci d'avoir contribué activement à ma formation au cours de ces 4 années d'internat et pour vos excellentes présentations toujours très didactiques et passionnantes.

A Madame le Docteur Glemarec, A Madame le Docteur Guillot,

Merci d'avoir contribué activement à ma formation au cours de ces 4 années d'internat, merci pour votre disponibilité et vos conseils, surtout lorsque que je vous pose des questions entre 2 consultations. Merci Joelle pour m'avoir formée à la scopie et Merci Pascale pour m'aider et me former aux problématiques d'ostéoporose.

A Monsieur Le Professeur François Gouin

Un grand merci pour avoir accepté de participer à mon jury de thèse.

A Madame le Docteur Anne Moreau

Un grand merci pour avoir accepté de participer à mon jury de thèse.

Aux Docteurs Dernis, Haettich, Esparbes, Denis pour leur accompagnement et leur enseignement au cours de mon semestre au Mans, très formateur alors que j'étais encore jeune semestre. Merci au Docteur Dernis pour son soutien.

Aux Docteurs Cormier, André, Caulier, Tanguy et Cozic, pour ce super semestre à la Roche sur Yon, au cours duquel j'ai pu me perfectionner à la pratique de la rhumatologie clinique, de la scopie, des gestes échoguidés, etc… Merci aussi pour cette ambiance si agréable et détendue et votre bonne humeur !

Au Professeur Douglas Veale et à Carl Orr pour avoir contribué à ma formation au cours de ces 6 mois à Dublin, pour ces projets réalisés ensemble mais également pour les projets à venir, et pour leur soutien.

A mes collègues internes, chefs de clinique avec qui j'ai partagé ces excellentes années d'internat, à l'hôpital, en journées de DES, en congrès et parfois aussi à l'extérieur.

Aux formidables IDE, aides-soignantes, ASH du service de Rhumatologie pour leur travail quotidien auprès des malades et pour nos échanges toujours agréables, que ce soit dans les couloirs du service, pendant les week-end d'astreintes ou pendant les pauses café.

A Sabrina, Roselyne, Flavie, Béatrice, Claudine pour leur investissement à nous faciliter la vie au quotidien, et pour votre bonne humeur et tous ces moments agréables que l'on passe entre 2 consultations.

A mes collègues du laboratoire INSERM UMR1238 (mention spéciale pour Régis et Kevin) pour les excellents moments passés au labo et à l'extérieur.

A mes collègues de promo qui sont devenus mes amis, pour tout ce que nous avons partagé pendant nos longues études de médecine.

A la « pineapple team » et plus particulièrement Pierre-Antoine, pour ton soutien -as a Y&S Rheumatologist- !

A ceux que j'aime, je n'ai rien à écrire que vous ne sachiez pas déjà.

TABLE DES MATIERES

TABLE DES TABLEAUX **7**

TABLE DES FIGURES **8**

LISTE DES ABBREVIATIONS **9**

I. INTRODUCTION **10**

I.1. Généralités sur le tissu synovial **10**

I.1.1. Morphologie du tissu synovial normal 10

I.2. Intérêt diagnostic des biopsies synoviales. **13**

I.2.1. Places actuelle des biopsies synoviales en pratique clinique. 13

I.2.2. Place actuelle des biopsies synoviales en recherche translationnelle. 16

I.3. Techniques de biopsie synoviale. **21**

I.4. Introduction de l'article. **22**

II. MATERIAL AND METHODS **24**

II.1. Patients and histological diagnosis. **24**

II.2. US guided synovial biopsies. **24**

II.3. Analysis of the quality and quantity of the synovial tissue retrieved during synovial biopsies. **25**

II.4. Statistical analysis. **26**

III. RESULTS **26**

III.1. Patient characteristics. **26**

III.2. US guided biopsy procedure was safe and successful. **27**

III.3. Quality and quantity of the synovial tissue retrieved after US guided synovial biopsies. **29**

IV. DISCUSSION **31**

V. CONCLUSION **34**

VI. REFERENCES **35**

VII. ANNEXES **43**

RESUME **52**

TABLE DES TABLEAUX

Table 1.

Données démographiques et cliniques des patients.

Table 2.

Récapitulatif de l'analyse histologique des biopsies synoviales.

Table 3.

Données finales de la cohorte de patients après suivi.

TABLE DES FIGURES

Figure 1. A. D'après Simkin (1). Articulation interphalangienne proximale normale. On y voit : les phalanges, le cartilage et la cavité articulaire (qui apparaît ici en blanc). **B.** Tissu synovial avec ses trois couches. 1. Lining layer ou ligne bordante ou intima. 2. Sublining ou sub-intima. 3. Tissu de soutien conjonctivo-adipeux.

Figure 2. D'après Barland et al. (2). Membrane synoviale normale en microscopie électronique. Ligne bordante. 1. Cavité articulaire 2. Espace intercellulaire. 3. Matrice extra cellulaire. 4. Prolongements cellulaires.

Figure 3. D'après Krenn et al. (3). Score de synovite. Exemples de grades 0 à 3 pour les 3 composants principaux : hyperplasie de la ligne bordante, activation du stroma et infiltrat inflammatoire.

Figure 4. D'après Krenn et al. (4). Algorithme diagnostique basé sur l'analyse histologique de la membrane synoviale.

Figure 5. D'après Pitzalis et al. (5). Illustration des 3 grands phénotypes synoviaux chez les patients porteurs de PR. H&E : Hématoxyline et éosine, CD20 : immunomarquage des lymphocytes B, CD3 : immunomarquage des lymphocytes T, CD68 : immunomarquage des macrophages, CD138 : immunomarquage des plasmocytes.

Figure 6. A, B, C, D, E. Biopsies synoviales de 5 lésions histologiques spécifiques.

- **A.** Dépôts de fibrine avec infiltrats de Polynucléaires neutrophiles (astérisque) correspondant à une arthrite septique.
- **B.** Synovite villo-nodulaire. Coloration Hématoxyline et Eosine.
- **C.** Synovite villo-nodulaire avec une coloration de Perl's montrant des sidérophages (flèche).
- **D.** Infiltrat cellulaire au sein de la membrane synoviale correspondant à une localisation articulaire de lymphome.
- **E.** Dépôts amyloïdes révélés par une coloration Rouge Congo. Amylose AL
- **F.** Micro tophus entouré de cellules géantes et de lymphocytes (flèche noire) aboutissant à un diagnostic de goutte.

Figure 7. Exemple d'une analyse histologique. La ligne noire représente la mesure globale de l'échantillon ; la ligne rouge représente la mesure de la largeur de l'échantillon et la ligne blanche la longueur de l'échantillon.

LISTE DES ABBREVIATIONS

ACR : American College of Rheumatology

Anticorps anti-CCP : Anticorps anti peptides citrullinés

CD : Cluster de différentiation

DAF : Decay-accelerating factor

EULAR : European League Against Rheumatism

FLS : Fibroblasts Like Synoviocytes

FLE : Follicules Lymphoïdes Ectopiques

FR : Facteur Rhumatoïde

IRM : Imagerie par Résonance Magnétique

PCR : Polymerase Chain Reaction

MEC : Matrice Extra cellulaire

MSU : Monosodium Urate Crystals

PPC : Calcium PyroPhosphate

PR : Polyarthrite Rhumatoïde

RIC : Rhumatisme Inflammatoire Chronique

TNF alpha : Tumor Necrosis Factor alpha

US : Ultrasound

I. INTRODUCTION

I.1. Généralités sur le tissu synovial

I.1.1. Morphologie du tissu synovial normal

Le tissu synovial ou membrane synoviale est le tissu tapissant la cavité articulaire des articulations diarthrodiales, et certaines bourses et tendons de l'organisme. Ce tissu est en relations intimes avec le cartilage, les tissu osseux, musculaire, tendineux, ligamentaire, et la capsule articulaire. Son positionnement privilégié au sein de l'articulation lui confère de multiples propriétés : élaboration et sécrétion du liquide synovial, lubrification, nutrition du cartilage, fonction de défense immunitaire, régulation de la pression et la température intra-articulaire (6). Il est composé des 3 couches : la ligne bordante synoviocytaire ou intima (synovial lining layer), la couche sous-intimale ou subintima (sub-lining) et le tissu de soutien conjonctivo adipeux ou subsynoviale (Figure 1).

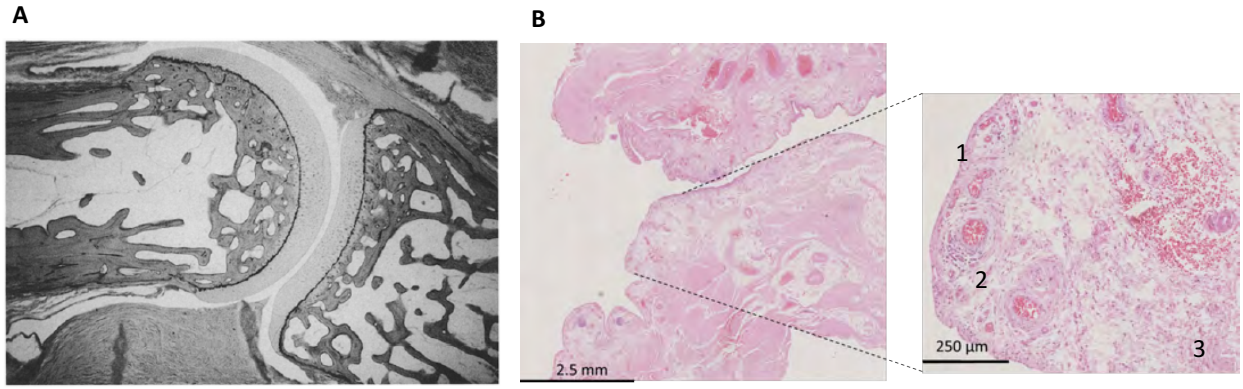

Figure 1. A. D'après Simkin (1). Articulation interphalangienne proximale normale. On y voit : les phalanges, le cartilage et la cavité articulaire (qui apparaît ici en blanc). **B.** Tissu synovial avec ses trois couches. 1. Ligne bordante (ou intima ou lining layer). 2. Sublining. 3. Tissu de soutien conjonctivo-adipeux.

I.1.1.a) Intima

L'intima ou ligne bordante est composée de 1 à 3 couches cellulaires en conditions physiologiques. Son épaisseur est 20 à 40µm (6). De façon intéressante, elle n'est pas pourvue de membrane basale pour délimitation et les cellules ne possèdent pas de jonctions serrées cellules-cellules ou de desmosomes. Ces contacts « lâches » entre cellules bordantes sont à la base des propriétés de la membrane. Ils permettent aux molécules de la matrice extra-cellulaire de diffuser au sein du liquide synovial dans la cavité articulaire et *vice-versa*. Il a été démontré en microscopie électronique l'enchevêtrement de ces cellules au sein d'une matrice extracellulaire (MEC) et l'existence de prolongements cellulaires au contact les uns des autres et au contact de la cavité articulaire. A noter qu'en certaines zones, un contact direct entre la MEC et la cavité articulaire est visualisé (2).

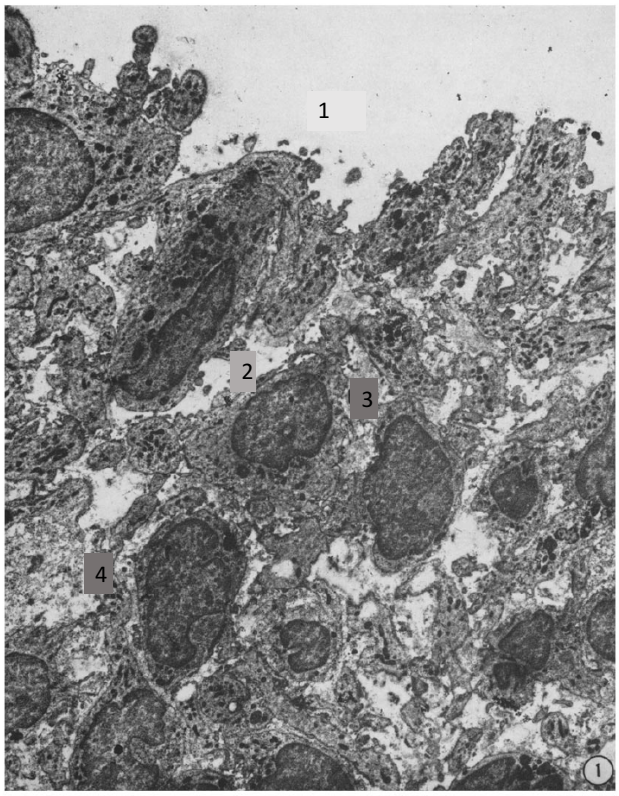

Figure 2. D'après Barland et al (2). Membrane synoviale normale en microscopie électronique. Ligne bordante. 1. Cavité articulaire 2. Espace intercellulaire. 3. Matrice extra cellulaire. 4. Prolongements cellulaires.

La ligne bordante est constituée de deux types cellulaires en conditions physiologiques : les synoviocytes ou FLS (Fibroblasts Like Synoviocytes) et les macrophages synoviaux résidents. Ces cellules ont longtemps été considérées comme des sous types cellulaires (cellules synoviales de type A et de type B, respectivement) distincts issus d'un même progéniteur (2,7). Ce concept a progressivement évolué et les études montrent qu'il s'agit bien de types cellulaires différents, qui interagissent de façon continue, et particulièrement en situation inflammatoire (8). Ces deux types de cellules présentent une origine embryonnaire différente ainsi qu'un phénotype morphologique et immunohistochimique différent. Les macrophages synoviaux résidents sont des cellules phagocytaires d'origine myéloïde. Ces cellules sont tissu-spécifiques et ainsi différentes des macrophages recrutés et différentiés à partir de monocytes circulants issus de la moelle osseuse (9–13). Elles expriment fortement le CD68 à leur surface, ce qui en fait le marqueur immunologique de choix en immunohistochimie. De façon intéressante, une étude a montré la capacité de macrophages dérivés de monocytes sanguins, à se différencier en macrophages résidents in situ, en conditions inflammatoires (14). Les synoviocytes sont des cellules fibroblastiques d'origine mésenchymateuse. Elles expriment fortement le CD55 à la surface (decay-accelerating factor [DAF]), ce qui en fait un marqueur de choix en immunohistochimie ou en cytométrie de flux (15). Ces cellules ont plusieurs rôles au sein de la membrane synoviale. D'une part un rôle mécanique : synthèse et excrétion d'acide hyaluronique et autres protéoglycanes constitutifs du liquide synovial ; mais également maintien de l'intégrité de la ligne bordante par l'expression de molécules d'adhésion (16,17). D'autre part immunitaire : reconnaissance des stimuli bactériens et rôle anti-infectieux par l'expression de Toll Like Receptors (18).

I.1.1.b) Sub-intima

La couche subintimale est constituée de tissu conjonctivo-adipeux, de capillaires et lymphatiques, et de cellules de l'immunité innée : mastocytes et macrophages. Les macrophages subintimaux pourraient présenter, selon certains auteurs, un phénotype différent de ceux de la ligne bordante notamment chez les patients porteurs de rhumatisme inflammatoires chroniques (14,19). En effet, dans le travail d'Ambarus et al. alors que les

macrophages de la ligne bordante exhibaient une polarisation plutôt de type M2, les macrophages subintimaux exprimaient à la fois des marqueurs de polarisation M1 et M2.

I.1.1.c) Tissu de soutien conjonctivo-adipeux

Le tissu de soutien, quant à lui, est constitué de tissu adipeux, de tissu conjonctif lâche, de vaisseaux sanguins et lymphatiques, ainsi que de fibres nerveuses (20,21). La répartition conjonctivo-adipeuse est variable selon les localisations articulaires. Ce tissu est en continuité avec l'appareil capsulo-ligamentaire. Ce tissu peut être le siège d'un infiltrat de cellules de l'immunité innée (lymphocytes en particulier).

I.2. Intérêt diagnostic des biopsies synoviales.

I.2.1. Places actuelle des biopsies synoviales en pratique clinique.

Le diagnostic étiologique d'une arthrite aigue ou chronique se fait généralement sur un faisceau d'arguments cliniques, paracliniques et d'imagerie. Dans ce cadre, la ponction et l'analyse du liquide articulaire permet l'analyse cytologique, bactériologique et la recherche de microcristaux et est indispensable en première intention dans cette démarche diagnostique. Malgré tout, il persiste des situations diagnostiques difficiles pour le clinicien, notamment lorsque l'analyse du liquide synovial et/ou les examens d'imagerie ne donnent pas d'information suffisantes.

Dans ces circonstances, l'analyse bactériologique, mycobactériologique, fungique, et la réalisation de PCR spécifiques de micro-organismes type Borrelia Burgdorferi pour la maladie de Lyme ou Tropheryma Whipplei pour la maladie de Whipple de la membrane synoviale peut permettre de poser le diagnostic d'arthrite infectieuse.

L'analyse histologique de la membrane synoviale, siège de l'inflammation, permet également de donner des informations importantes. Par exemple, la présence d'aspects histologiques typiques de certaines pathologies au sein de la membrane synoviale permet un diagnostic de certitude. Par exemple, l'existence de dépôts de sidérophages pour les synovites villo-nodulaires, la présence de métaplasie cartilagineuse au sein de la membrane pour l'osteochondromatose, la présence de dépôts microcristallins pour les arthropathies

microcristallines ou la présence de granulomes pour les arthrites infectieuses à mycobactéries.

Le diagnostic des rhumatismes inflammatoires chroniques (RIC), et notamment le plus fréquent d'entre eux, la polyarthrite rhumatoïde (PR), repose sur un faisceau d'arguments cliniques, biologiques (syndrome inflammatoire biologique, présence de facteurs rhumatoïdes, d'anticorps anti-CCP), radiologiques (érosions radiographiques, synovites à l'échographie ou à l'IRM). Les sociétés savantes américaines et européennes (ACR et EULAR) ont également développé des critères de classification pour la PR (22) et pour d'autres RIC. Il existe également des caractéristiques synoviales macroscopiques différentes au sein des RIC, et notamment entre PR et rhumatisme psoriasique. Les équipes pratiquant l'arthroscopie ont en effet observé et décrit un aspect tortueux caractéristique des vaisseaux, au sein des synovites de patients porteurs de rhumatismes psoriasiques et de spondylarthropathies. En revanche, les capillaires sont rectilignes dans les synovites de PR (23). Des différences ont également été observées à l'échelon microscopique histologique, avec une néovascularisation plus prononcée dans les membranes synoviales de patients porteurs de spondylarthropathies.

Au-delà de du diagnostic précis du type de rhumatisme inflammatoire, l'analyse histologique permet également d'étudier l'existence d'un infiltrat inflammatoire dans les synovites et de décrire les types cellulaires qui le composent (polynucléaires neutrophiles, macrophages, lymphocytes, plasmocytes). Un score a été développé par Krenn et al. en 2002, visant à grader les synovites en fonction de 3 paramètres principaux : l'hyperplasie de la ligne bordante, l'activation du stroma (cellules résidentes : macrophages du sublining, cellules endothéliales), l'infiltrat inflammatoire (cellules de l'immunité adaptative : lymphocytes, plasmocytes) qui sont gradés de façon semi-quantitative de 0 à 3 (24). Le score total est sur 9, et les synovites sont considérées de bas grade si le score est inférieur ou égal à 4, ou de haut grade si le score est supérieur ou égal à 5. Dans la publication originale, la sensibilité était de 61,7% avec une spécificité de de 96,1%, avec le seuil de 5 sur 9, pour différencier arthropathie mécanique et inflammatoire (3,24).

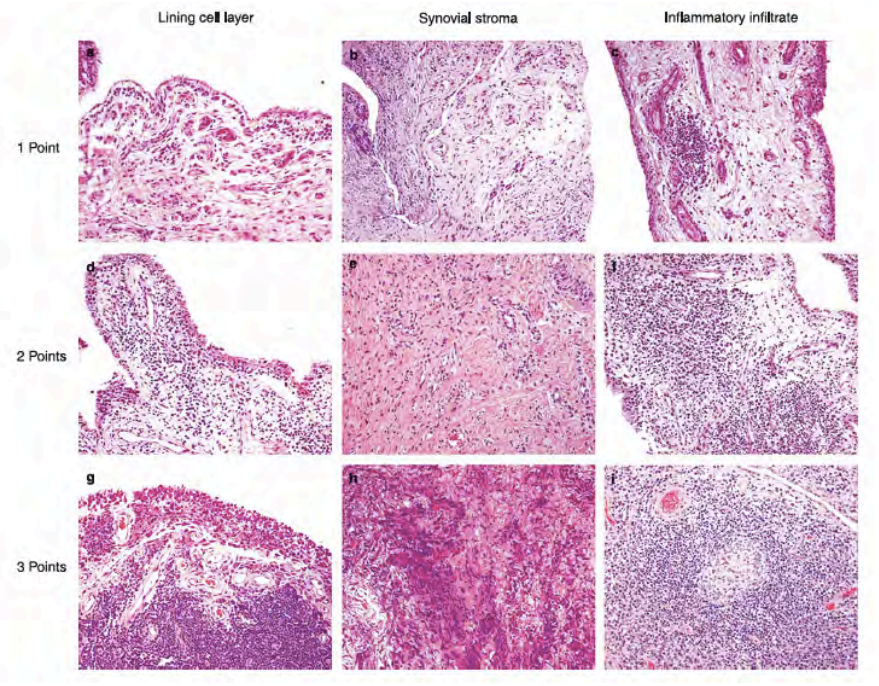

Figure 3. D'après Krenn et al. (3). Score de synovite. Exemples de grades 0 à 3 pour les 3 composants principaux : hyperplasie de la ligne bordante, activation du stroma et infiltrat inflammatoire.

De façon intéressante, un algorithme diagnostic basé sur l'analyse anatomopathologique de la membrane synoviale a été récemment publié par Krenn et al. Cet algorithme permet d'orienter le diagnostic en fonction du caractère inflammatoire ou non de la membrane synoviale, de l'existence d'aspects histologiques typiques comme granulomes ou cristaux, et du score de synovite précédemment décrit.

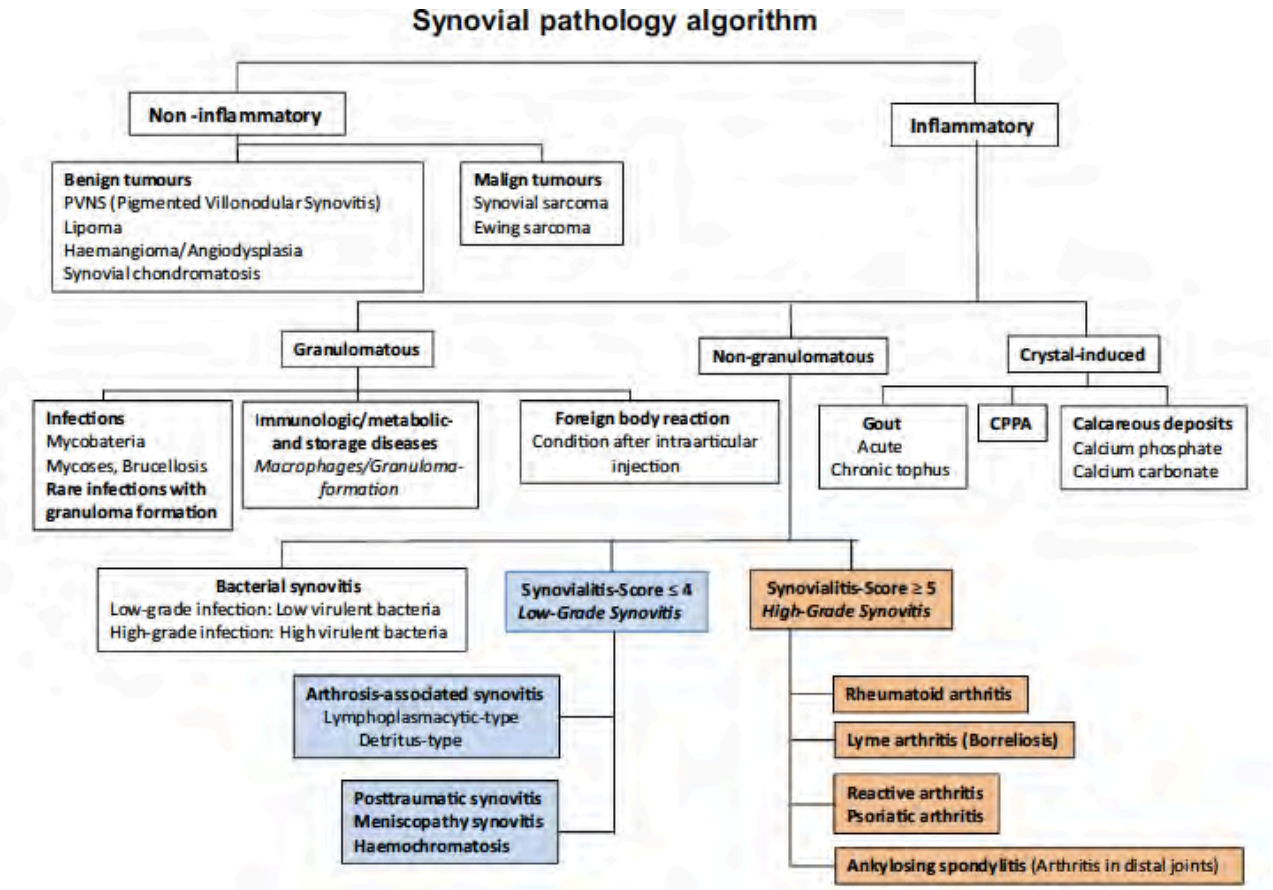

Figure 4. D'après Krenn et al. (4). Algorithme diagnostique basé sur l'analyse histologique de la membrane synoviale.

I.2.2. Place actuelle des biopsies synoviales en recherche translationnelle.

La polyarthrite rhumatoïde (PR) est une maladie hétérogène et complexe. Au cours des 15 dernières années de grandes avancées ont été faites dans la connaissance de la physiopathologie de la PR, mais certaines questions restent entières, notamment celle du pronostic, qui est variable selon les patients et fortement intriqué avec l'existence de destructions articulaires, mais aussi celle de la réponse au traitements, absente chez 30% des patients (25). Aussi, la recherche sur le tissu synovial s'est concentrée sur trois grandes questions scientifiques. Comment diagnostiquer la PR de façon précoce ? Comment prédire le pronostic (recherche de biomarqueurs membranaires) ? Comment prédire la réponse au traitement ?

I.2.2.a) Diagnostic précoce de la polyarthrite rhumatoïde chez les patients porteurs d'arthrites indifférenciées.

Concernant le diagnostic précoce des PR débutantes chez des patients porteurs d'arthrites indifférenciées, plusieurs modèles cliniques prédictifs ont été proposés, mais aucun d'entre eux n'est utilisé en pratique courante en raison d'un manque de validation sur de grandes cohortes et de performances insuffisantes en terme de sensibilité/spécificité (25–27). Afin de répondre à cette problématique, plusieurs travaux ont été réalisés, comparant l'aspect histologique et immunohistologique de la membrane synoviale de patients porteurs d'arthropathies mécaniques, de rhumatisme psoriasique, de spondylarthropathie et de polyarthite rhumatoïde. L'objectif était de mettre en évidence des aspects histologiques spécifiques qui permettraient de classer les patients porteurs d'arthrites indifférenciées à un stade précoce (28,29). Bien que quelques différences aient été mises en évidence (néovascularisation plus importante dans les spondylarthopathies, infiltrats lympho-plasmocytaire plus important dans la PR, altération des molécules d'adhésion (intégrines) dans la PR), aucun des travaux n'a mis en évidence d'aspect spécifique d'une pathologie.

Cette problématique a également été étudiée à l'échelon transcriptomique, dans un travail qui visait à étudier les signatures transcriptomiques des membranes synoviales de patients porteurs de PR, de spondylarthropathies, d'arthrose et d'arthrites indifférenciées. En raison d'un chevauchement important des expressions géniques entre PR, spondylarthropathies et arthrites indifférenciées, la précision diagnostique sur la seule base de l'analyse transcriptomique était faible (58%). En ajoutant les données cliniques, on augmentait la sensibilité de l'algorithme de prédiction diagnostic à 98% (30). Ces données nous informent donc quant au caractère insuffisant de la seule analyse du transcriptome synovial. Des travaux supplémentaires sont donc nécessaires dans ce domaine et notamment des analyses protéoiques peuvent avoir leur intérêt.

I.2.2.b) Biomarqueurs synoviaux pronostiques dans la PR.

L'étude de la membrane synoviale, dans un premier temps à l'échelon histologique et immunohistologique de patient porteurs de PR, a permis de mettre en évidence l'existence d'aggrégats cellulaires, organisés en follicules lymphoïdes ectopiques (FLE) au sein de la membrane synoviale de certains patients. Ces FLE s'organisent au sein de la membrane comme des organes lymphoïdes tertiaires avec lymphocytes B, T, macrophages et cellules dendritiques. Leur rôle dans la physiopathologie de la PR est encore discuté (32). Une équipe a montré une corrélation entre l'existence de ces FLE et l'atteinte structurale dans la PR (33), mais ces résultats n'ont pas été confirmés dans des travaux ultérieurs (34,35).

De façon intéressante, ces FLE ne sont présents que chez 30-40% des patients porteurs de PR (5). Trois grands phénotypes ont ainsi été décrits : le phénotype folliculaire dans lequel on retrouve un infiltrat majoritairement lymphocytaire avec formation de FLE, le phénotype diffus dans lequel l'infiltrat inflammatoire est également réparti au sein de la membrane et le phénotype pauci-immun, où l'infiltrat inflammatoire est discret.

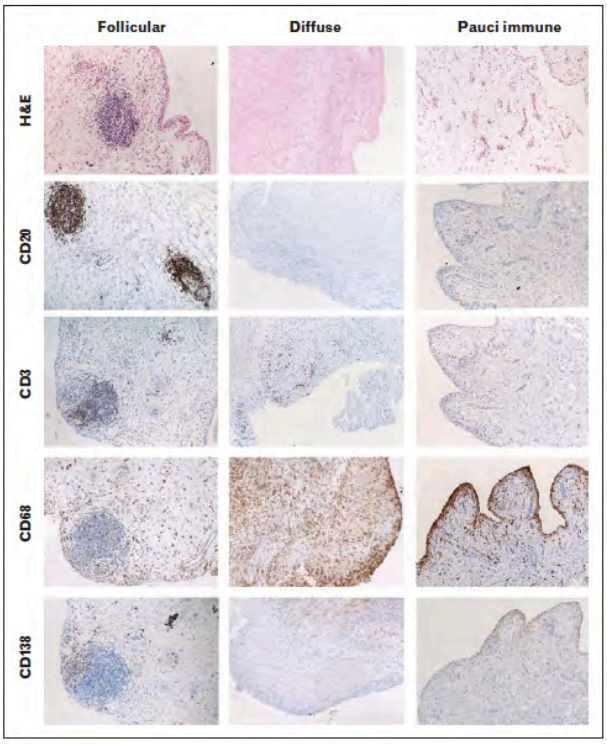

Figure 5. D'après Pitzalis et al. (5). Illustration des 3 grands phénotypes synoviaux chez les patients porteurs de PR. H&E : Hématoxyline et éosine, CD20 : immunomarquage des lymphocytes B, CD3 : immunomarquage des lymphocytes T, CD68 : immunomarquage des macrophages, CD138 : immunomarquage des plasmocytes.

Ces phénotypes histologiques, ont été secondairement associés à des signatures génomiques et transcriptomiques spécifiques : lymphocytaire pour le phénotype folliculaire, macrophagique pour le phénotype diffus et fibroblastique pour le phénotype pauci-immun (36). Actuellement, la corrélation entre ces phénotypes histologiques, ces signatures cellulaires/moléculaires spécifiques et les phénotypes cliniques (atteinte structurale, présence ou absence de facteurs rhumatoïdes ou d'anticorps anti-CCP) fait l'objet d'intense recherche auprès de plusieurs équipes. Récemment, une équipe dublinoise a montré l'existence d'un infiltrat lymphocytaire B et T plus important et l'existence de FLE plus fréquemment chez les patients porteurs de PR positifs pour les anticorps anti-CCP. De plus, les patients dont les membranes synoviales comportaient plus de lymphocytes B avaient plus fréquemment des érosions radiographiques au cours du suivi (37). D'autres travaux sont en cours et sont nécessaires pour confirmer ces données.

Par ailleurs, l'étude de protéines membranaires a montré un intérêt comme facteur pronostique. En effet, la présence de taux élevés de TNF alpha et d'Interleukine-6 -deux cytokines pro-inflammatoires jouant un rôle clé dans la PR- dans la membrane synoviale est associée à une PR active cliniquement (38).

I.2.2.c) Biomarqueurs synoviaux prédictifs de la réponse au traitement dans la PR.

Actuellement, 30 à 40% de patients porteurs de PR ne répondent pas à une première biothérapie (39). Pour le clinicien, il n'existe à l'heure actuelle pas de moyen fiable permettant de prédire la réponse d'un patient au traitement par biothérapie, et la décision reste basée sur un faisceau d'arguments cliniques, biologiques, et sur l'expérience du médecin. Cette problématique est un enjeu de taille, tant à l'échelle du patient (douleurs, atteinte structurale) qu'à l'échelle sociétale (coût des biothérapies).

Des études ont par le passé tenté d'identifier des marqueurs prédictifs de réponse aux anti-TNF alpha dans le sang circulant, plus facile d'accès que la membrane synoviale, mais les résultats se sont avérés insuffisants en terme de sensibilité et de spécificité pour être utilisés en routine (40,41).

Concernant la prédiction de réponse aux traitements par anti-TNF alpha, des études contradictoires ont été publiées. Une première équipe a montré que l'existence d'agrégats lymphocytaire au sein de la membrane synoviale était un facteur prédictif indépendant de mauvaise réponse aux anti-TNF alpha (42). A l'inverse, l'équipe de Klaasen et al. a montré que les patients porteurs de synovites avec agrégats lymphocytaires répondaient en moyenne mieux à l'infliximab (43). D'autres études seront nécessaires pour conclure à ce sujet.

Des travaux d'analyse génomique en pré- ou post- traitement, ou de signature transcriptomique entre patients répondeurs et non-répondeurs aux anti-TNF alpha ont également été réalisés. Ces études ont mis en évidence des expression différentielles de gènes pro-inflammatoires principalement, mais également de gènes codant pour des acteurs de l'activation immune et de la prolifération cellulaire entre les patients répondeurs et non répondeurs (44–46). Des travaux de validation sur de plus larges cohortes seront nécessaires.

La réponse au rituximab, un anticorps monoclonal anti-CD20, a également fait l'objet de plusieurs études de prédiction. La déplétion en lymphocytes B et en plasmocytes synoviaux semble être un marqueur prédictif de bonne réponse au traitement (47,48).

Jusqu'ici, l'ensemble des études publiées, qu'elles soient rétrospectives ou prospectives, bien qu'informatives, n'étaient pas randomisées. L'étape suivante indispensable au développement à plus large échelle de stratégies de stratification thérapeutique basées sur l'analyse phénotypique de la membrane synoviale dans la PR est la mise en place d'essais thérapeutiques randomisés. Ce type d'essai clinique de stratégie est en cours actuellement en Angleterre, et d'autres protocoles sont à venir dans d'autres pays d'Europe. Les résultats de ces travaux permettront peut-être le développement en pratique courante de stratégies thérapeutiques personnalisées basées sur l'analyse du phénotype synovial des patients porteurs de PR.

I.3. Techniques de biopsie synoviale.

De multiples techniques de biopsie synoviale sont été décrites dans la littérature.

Les premières biopsies synoviales ont été réalisées dans de grosses articulations faciles d'accès comme le genou. Les biopsies étaient réalisées à l'aide d'aiguilles à biopsies ou de punch de différents calibres introduites dans l'articulation en repérage anatomique (49–51). Progressivement les techniques ont évolué avec le développement de l'arthroscopie et d'arthroscopes de petits calibres (1-2,7mm) (52).

Les biopsies synoviales ont ainsi pu être réalisées sous contrôle visuel à l'aide d'une voie d'abord supéro et inféro latérale pour l'arthroscope et le forceps. Cette technique a été largement utilisée à partir des années 1960 et est encore actuellement utilisée en Rhumatologie à des fins diagnostiques ou de recherche scientifique avec une bonne tolérance (52). Cependant cette technique de biopsie reste invasive et difficilement accessible en pratique courante.

L'échographie s'est beaucoup développée ces dernières années en rhumatologie du fait de son caractère maniable, facile d'accès, rapide et non invasif. L'échoguidage a donc pris un essor important pour la réalisation de gestes infiltratifs, mais également pour la réalisation de biopsies synoviales. De plus, l'échoguidage permet la réalisation de biopsies synoviales au sein de petites articulations comme les metacarpo-phalangiennes et inter-phalangiennes proximales, souvent touchées dans les rhumatismes inflammatoires chroniques comme la polyarthrite rhumatoïde (53). Certaines équipes utilisent un trocard à biopsies semi-automatique avec ou sans co-axiale (53–56) alors que d'autres utilisent un système de forceps avec introducteur (57,58). La technique de biopsies synoviale échoguidée à l'aide d'un trocard à biopsies semi-automatique avec aiguille co-axiale est utilisée dans le service de Rhumatologie du CHU de Nantes depuis 2007.

I.4. Introduction de l'article.

Success rate and utility of ultrasound guided synovial biopsies in clinical practice.

Aurélie Najm[1], Carl Orr[2], Marie-Françoise Heymann[3], Géraldine Bart[1], Douglas J Veale[2], Benoît Le Goff[1].

1. Department of Rheumatology, Hôtel-Dieu Hospital, 1, place Alexis-Ricordeau, 44093 Nantes cedex 1, France.

2. University College Dublin Department of Rheumatology, St Vincent's Hospital, Dublin, Ireland

3. Department of pathology, Hôtel-Dieu Hospital, 1, place Alexis-Ricordeau, 44093 Nantes cedex 1, France.

INTRODUCTION

Synovial tissue is the principal target and end organ involved in the pathogenesis of multiple articular disease processes (59,60). Synovial tissue analysis has been widely used for basic science, translational and clinical research. Moreover, synovial assessment allows for studying many aspects of disease processes including pathogenesis (5), the identification of relevant targets clinical features (61), diagnosis, prognosis (62) as well as in assisting in assessments of response to treatment (63–65).

Histological and immunohistological synovial assessment is also used as a diagnostic tool (66). Indeed, it is especially useful for identifying arthritis of an infectious aetiology, when synovial fluid or blood analysis (Gram, Ziehl) and cultures are negative or in cases where empiric antimicrobial therapy has been commenced before it has been possible to examine the synovial fluid (67). The bacterial broad range 16S ribosomal RNA can also be tracked down by polymerase chain reaction (PCR) on synovial tissue (68). The same methods allow identification of fungal, mycobacterial, spirochetes and Tropheryma Whipplei in the joint. False negative for monosodium urate crystals (MSU) and calcium pyrophosphate (PPC)

occur frequently at microscopic examination of the synovial fluid (69), and synovial tissue assessment can be helpful with typical histological features. Finally, synovial benign tumours such as primary or secondary osteochondromatosis or villonodular synovitis can be diagnosed as well, showing specific macroscopic and histological pattern.

There are several techniques to obtain synovial tissue from the joints. Synovial biopsy was performed by Forestier in 1932 using a needle blindly introduced in the knee joint (49). Polley (50) and Parker (51) described new smaller diameter needles that have been widely used over the past years for knee synovial biopsies. Beaulé (70), Parlier and Cuau (71) then described a technique of synovial biopsy under direct visualisation under flurorscopy with a semi-automatic Tru-cut needle. This technique allows performing multi-sites biopsies such as hips, shoulders, elbows, ankles and wrists. Synovial biopsies were later performed under direct vision using 2 portals via an arthroscope (72). Although this technique is usually well tolerated (66), it remains invasive, expensive and not yet widely available. Moreover, it has been shown that microscopic measurements of synovial inflammation does not differ between biopsies taken blindly or under guided vision (73).

More recently, ultrasound guided synovial biopsies have been developed. Musculoskeletal ultrasound (US) is very commonly used nowadays, especially for guiding interventional procedures (74,75). This technique has the benefit of being low cost, rapidly and easily performed without the need for exposing the patient to ionising radiation, and is widely available (76). It is more practical than arthroscopy for biopsying small joints and allows guidance to the thickest synovial zones. Moreover, Kelly et al (53), reported that increasing synovial thickness on ultrasound correlated with increasing grades of synovitis on histological examination. However, few studies have reported on synovial biopsies performed in routine clinical practice (54,55). It is unknown if the success and the quality of the biopsy are the same as the one performed in a research setting. Finally, their clinical utility is still a matter of debate.

The aims of our study are (a) to describe the indications for US guided synovial biopsies in the clinical setting, (b) to determine the rate of success in acquiring synovial tissue using this approach and to report the complications**,** (c) to determine how frequently the synovial biopsy

can lead to a clear diagnosis and (d) to assess the quality of the synovial tissue obtained using this technique.

II. MATERIAL AND METHODS

II.1. Patients and histological diagnosis.

We included all patients who underwent a US guided synovial biopsy between February 2007 and December 2014 in Nantes University hospital for arthritis without definite diagnosis based on the history, clinical examination or imaging. Ethics approval was not required in accordance with the policy of our institution. During this service evaluation study, we collected epidemiological (age, sex) and clinical data (clinical presentation, indication, biopsied joint, complications) using a standardized form. Final histological diagnosis was reported by 3 pathologists who had an expertise in assessing synovial tissue in a formal report based on a Hematoxylin and Eosin staining. Patients were followed to determine the clinical course of their symptoms.

II.2. US guided synovial biopsies.

Synovial biopsies were performed under US guidance using a Philips HD11 XE ultrasound machine and a 7-13MHz transducer from Philips Healthcare. They were performed in an outpatient and inpatient setting depending on the patient's presentation. All patients underwent a thorough assessment of the joint to be biopsied. Vascular and nervous structures nearby were identified and synovial thickness was assessed.

All the biopsy procedures were performed by one operator (BLG) who had an expertise in US examination, under sterile technique (wearing gown, sterile gloves, mask and a surgical cap). Skin disinfection was processed with a 5 steps protocol using Iodine polyvidone or Hibiscrub if the patient had Iodine past history of allergy. The joint was draped and a sterile field thus generated. The transducer was covered with sterile gel and sterile sheath. Anaesthesia was performed injecting 5 to 10 ml of lidocaine 2% in the subcutaneous tissue and up to the joint capsule. If an effusion was present, synovial fluid was withdrawn and sent to the laboratory for cell count, crystal microscopy, bacteriological, mycobacteriological

and/or fungal analysis depending on the patient clinical history and features. A semi-automatic guillotine biopsy “Tru-cut®” needle from TEMNOS has been used for all the biopsies. The calibre used was 16 Gauge (G) for small and intermediate joints or 14G for large joints such as hips, shoulders and knees. Coaxial needle was inserted under US guidance through the skin until reaching the articular cavity. The coaxial needle was positioned in intimate contact with the synovium. The semi-automatic guillotine biopsy “Tru-cut®” needle was then inserted through the cannula of the co-axial needle, still under US guidance. Once positioned within the zone of interest of the synovial tissue, the Tru-cut® needle was triggered collecting a piece of synovial tissue according to the size of the joint. This Tru-cut® needle was repeatedly inserted through the co-axial needle and triggered to obtain the appropriate number of samples. Then, these two needles were removed and a classical bandage was applied. Patients were recommended to have 48 hours rest after the procedure.
Depending on the indication of the biopsy and the size of the joint, 3 to 8 biopsies were performed per procedure and sent for bacteriological, mycobacteriological and/or fungal examination in appropriate laboratories. At least 1 sample was fixed in formalin 4%, embedded in paraffin and sent to the pathology laboratory. When the clinical history was relevant extra samples were sent for universal bacterial polymerase chain reaction (PCR) (ARN 16S), universal fungal PCR (ARN 18S) and Trophyrema Whipplei or Lyme PCR.

II.3. Analysis of the quality and quantity of the synovial tissue retrieved during synovial biopsies.

All the synovial biopsies were blindly read by one rheumatologist (AN). The number of samples per patient, the presence or absence of synovial tissue, the presence or absence of a synovial lining layer, the length and the width, the total area of the biopsy (mm2), the area of proper synovial tissue (mm2), was assessed in standardized manner with the NDP viewer® software. These findings were compared to the histological findings described on the pathologist reports which were the gold standard. In case of disagreement between rheumatologist and pathologist, an expert reader (DV) was responsible for final decision. We considered the biopsy successful when synovial tissue was seen at the histological examination. Good quality was defined as: sufficient size (>0,5 mm^2) (77), preserved tissue allowing assessment by pathologists and presence of lining layer.

II.4. Statistical analysis.

Mean and median were used to describe quantitative data according to their Gaussian distribution. Number and percentage were used to report qualitative data. Fisher test has been used to compare percentage. Kappa coefficient calculation was used to assess the interobserver reliability for histological analysis. $p<0.05$ was considered as statistically significant. All statistics were made through GraphPad Prism 6.0® software.

III. RESULTS

III.1. Patient characteristics.

Seventy-four patients underwent 76 US guided synovial biopsy procedures. Demographic and clinical features of patients included in the study are shown in Table 1. Mean age was 57 years (Range 13-86 years) and there were 39 (52.7%) men. Most of the patients presented with an undifferentiated chronic monoarthritis (54.1%, n=40). The biopsied joints were reparsed as followed: 46 knees (60.5%), 6 ankles (8%), 6 wrists (8%), 5 shoulders (7%), 4 hips (5%), 2 elbows, 2 sternoclavicular joints, 2 metatarso-phalangeal joints and one pubic symphysis, one acromio-clavicular joint and one peroneal tenosynovitis. Patients were mainly referred to rule out the diagnosis of septic arthritis (82.4%, n=61).

Table 1. Demographic and clinical features of the patients. Values are n (%) unless otherwise specified.

Variables	Values
Female	35 (47.3)
Male	39 (52.7)
Age, yrs, mean (range)	57 (13–86)
Indications	
Undifferentiated chronic monoarthritis	40 (54.1)
Acute monoarthritis	18 (24.0)
Chronic undifferentiated oligoarthritis	7 (9.3)
Chronic polyarthritis	6 (8.0)
Chronic bursitis	1 (1.3)
Chronic tenosynovitis	1 (1.3)
Acute polyarthritis	1 (1.3)

Table 1. Données démographiques et cliniques des patients.

III.2. US guided biopsy procedure was safe and successful.

Overall, 62 of the 76 biopsies (81.6%) yielded synovial tissue according to the pathologists' analysis. Within these 62 biopsies, the main histological finding was a non-specific inflammatory mononuclear cell infiltrate (lymphocyte, monocytes and plasma cells) (81%, n=50). A mild neutrophil infiltrate was seen in 24 (50%) of these biopsies. 8 (13%) biopsies showed specific histological lesions (Figure 1). A major neutrophil cell infiltrate consistent with a septic arthritis was found in 2 cases. 2 biopsies showed a synovial infiltration of positive Perls' siderophages (villo-nodular synovitis). 1 biopsy showed vascular and interstitial deposits of Sirius red staining protein consistent with amyloidosis AL. 1 biopsy contained tophi surrounded by lymphocytes and giant cells. 1 biopsy found dystrophic cartilage inside the synovial tissue; consistent with synovial osteochondromatosis. Finally, 1 biopsy showed an articular localisation of lymphoma. Four biopsies retrieved normal synovial tissue without any inflammatory cell infiltrate (Table 2).

Table 2. Histopathological analysis.

Histopathological Findings	No. Biopsies
Normal synovium	4
Inflamed synovium	50
Cell infiltrate:	
Lymphocytes	50
Plasma cells	22
Neutrophils	24
Specific lesions	8
Villonodular synovitis, shoulder and knee	2
Infectious arthritis*	2
Amyloid arthritis, knee	1
Articular localization of mantle B cell lymphoma, ankle	1
Gout, first MTP	1
Osteochondromatosis, knee	1
Failure	14

* Two infectious arthritis sites (hip, ankle) treated on typical histological aspect with no relapse after 6 weeks of empiric antibiotics. MTP: metatarsophalangeal.

Table 2. Récapitulatif de l'analyse histologique des biopsies synoviales.

The 14 failed biopsies occurred in both small and large joints. Percentages of failed biopsies per joint were as follows: Glenohumeral joints n=3/5 (60%), ankle n=3/6 (50%), hip n=2/4 (50%), wrist n=2/6 (38.3%), elbow n=1/2 (50%), sternoclavicular joint n=1/2 (50%), knees n=2/46 (4.3%). In case of failure, histological analysis showed mainly connective and adipose tissue in 10 cases, fibrin and leucocytes in 3 cases, tendon in 1 case. Tolerance per procedure was excellent. One patient taking acetyl salicylic acid at the time of the biopsy presented with a haemarthrosis 48 hours after the procedure, which resolved following arthrocentesis within one week.

Overall, 10 (16.2%) definitive diagnoses were made based only on synovial tissue histological or PCR analysis.

Long term follow-up (mean 34.9 months (Range; <1 month-96 months) and final diagnosis were available for 66 of the 74 patients (Table 3). No patient has since been diagnosed with an infectious arthritis or villo-nodular synovitis or developed any complication of the biopsy procedure. In three of the cases where the diagnosis remained unclear despite the US guided biopsy and in two case of failed biopsy, patients underwent secondary procedures. One of them had an arthroscopic examination after the US guided biopsy and four of them had an open synovectomy. One of those synovectomy allowed a diagnosis of chondrocalcinosis on pathological examination.

Table 3. Overall final diagnosis after followup. Values are n (%).

Final Diagnosis	Values
Rheumatoid arthritis	7 (9.5)
Ankylosing spondylitis	2 (2.7)
Psoriatic arthritis	5 (6.8)
Degenerative arthropathy	12 (16.2)
Crystal arthropathy	4 (5.4)
Chondrocalcinosis	2 (2.7)
Gout	3 (4.1)
Villonodular synovitis	2 (2.7)
Osteochondromatosis	1 (1.4)
Giant cell arthritis	1 (1.4)
Behçet disease	1 (1.4)
Latent infectious arthritis	4 (5.4)
Others	2 (2.7)
Undifferentiated arthritis	21 (28.4)
Lost to followup	7 (9.5)
Total	74 (100)

Table 3. Données finales de la cohorte de patients après suivi.

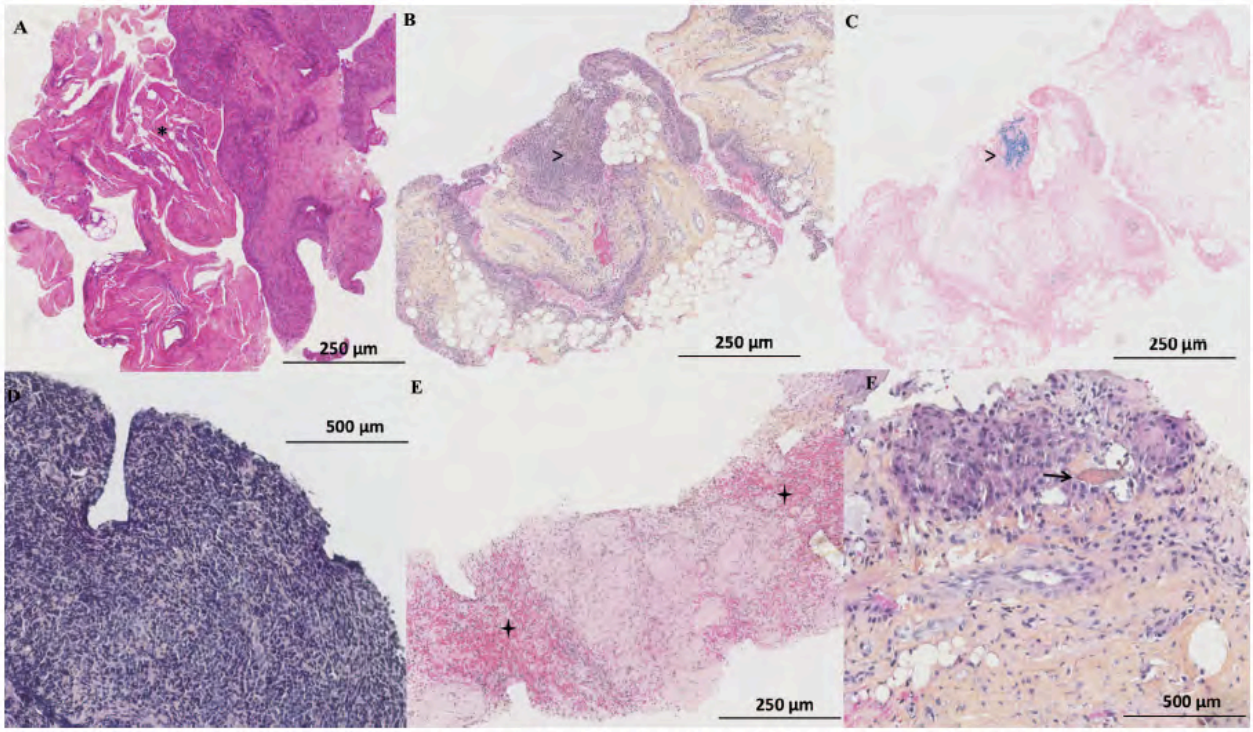

Figure 6. A, B, C, D, E. Biopsies synoviales de 5 lésions histologiques spécifiques.

- **A.** Dépôts de fibrine avec infiltrats de Polynucléaires neutrophiles (astérisque) correspondant à une arthrite septique.
- **B.** Synovite villo-nodulaire. Coloration Hématoxyline et Eosine.
- **C.** Synovite villo-nodulaire avec une coloration de Perl's montrant des sidérophages (flèche).
- **D.** Infiltrat cellulaire au sein de la membrane synoviale correspondant à une localisation articulaire de lymphome.
- **E.** Dépôts amyloïdes révélés par une coloration Rouge Congo. Amylose AL
- **F.** Micro tophus entouré de cellules géantes et de lymphocytes (flèche noire) aboutissant à un diagnostic de goutte.

III.3. Quality and quantity of the synovial tissue retrieved after US guided synovial biopsies.

Finally, the synovial tissue retrieved was assessed for quality and quantity. For this purpose, we analysed the histological characteristics per sample retrieved during the procedure (Figure 2). The median number of sample taken per patient was 1 (IQR 1-3) leading to a total of 125 samples available for analysis. Mean length and width of the biopsy samples were 6.34

millimetres (mm) (+/- 3.60) and 1.70 mm (+/- 0.77) respectively. The mean total area of the samples was 8.77mm^2.

Biopsies showed synovial tissue at the histological examination in 102 samples (80.1%). The average area of synovial tissue is these samples was 6.36 mm^2 corresponding to 72.5% of the total area of biopsied tissue. The other type of tissue present on these biopsies were connective tissue in 101 cases (80.8%), adipose tissue in 42 cases (33.6%), tendon in 14 cases (11.2%) and fibrin in 24 cases (19.2%). The 23 samples retrieving no synovial tissue were composed of fibrin in 15 cases (12%), conjunctive and adipose tissue in 17 cases (13.6%), tendinous tissue in 3 cases (3.15%), cartilage in 3 cases (3.15%) and muscle in one case (0.8%).

Synovial lining layer was found in 92.6% of the successful biopsies.

We finally compared our histological final finding regarding presence or absence of synovial tissue with the ones given by the pathologist and found 97.1% of agreement. Interobserver reliability for presence/absence of synovial tissue was high with a kappa coefficient of 0.90 (95% CI = 0.763 to 1).

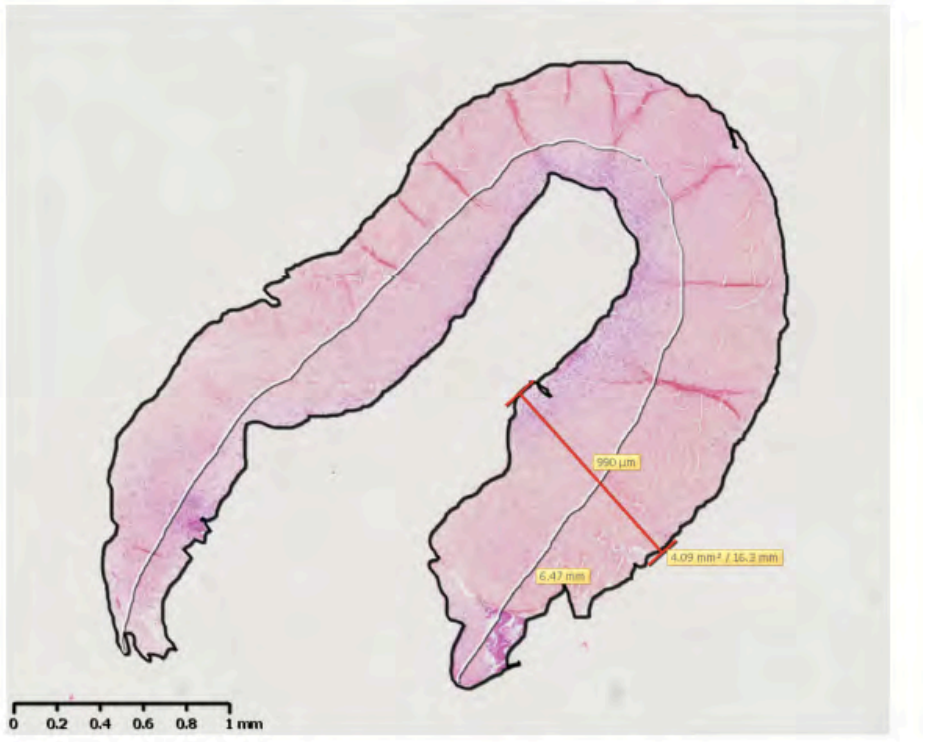

Figure 7. Exemple d'une analyse histologique. La ligne noire représente la mesure globale de l'échantillon ; la ligne rouge représente la mesure de la largeur de l'échantillon et la ligne blanche la longueur de l'échantillon.

IV. DISCUSSION

Given the fact that synovial tissue analysis has been mostly used for research purposes, our study highlights the potential diagnostic role of synovial biopsy in routine clinical practice. In order to develop this technique in clinical practice, the patient needs to be offered a well-tolerated technique with an acceptable rate of success.

To date, two different techniques of US guided synovial biopsies have been described. Both have been shown to be safe, and well tolerated by the patients (76). The first method requires a single portal with a flexible or rigid biopsy forceps. The portal is directly introduced inside of the joint to perform biopsies (57). The second technique as outlined above, requires an empty co-axial needle that is inserted inside of the joint and a semi-automatic guillotine-type needle that is inserted through the co-axial. The procedure is not painful after the local anaesthesia and once the co-axial needle is settled and this technique allows retrieving several biopsies during the same procedure without moving the co-axial needle. To our knowledge, five other studies, reporting their experience of US guided synovial biopsies, have been published to date. Two reported their experience using the first technique (57,58), one of them a technique using semi-automatic guillotine-type needle without co-axial needle (53) and two of them using the second technique outlined above (54,55).

The success rates in retrieving synovial tissue described by other authors vary from 89% to 100% (53,55–58). Although, the rate of success in our cohort was slightly lower, for which there are several potential reasons. Our patients comprised a heterogeneous group regarding clinical features and the joints that have been biopsied among those studies and there were also minor differences in techniques in 2 of the studies referenced above. Moreover, no biopsies have been done prior 2007 in our centre and 43% of the failures occurred within the first 18 months (6 on 14), especially in more challenging joints such as ankles, wrists, hips or shoulders. This might correspond to the operator learning curve. However, our success rate remains equivalent to the highest rates described for synovial biopsies with blind needle (48 to 85%) (78).

In our study, patients were referred mostly by their GPs or their rheumatologist with no clear diagnosis despite multiple punctures with synovial fluid analysis and imaging consisting in computed tomography scanner (CT-scan) or magnetic resonance imaging (MRI). Given the fact that low-grade infection often evolves in chronic arthritis with joint destruction, it is very important to pursue atypical germs such as tuberculosis, fungi, Tropheryma Whipplei, Borrelia Burgdorferi. Moreover, some of the more common bacteria can be responsible of low-grade infection in some rheumatic patients because of immunosuppression. In all these situations, the biopsy allows a quick bacteriological examination with Gram staining, then later culture and PCR analysis for atypical organism. Indeed, 2 patients were diagnosed with Lyme and articular Whipple disease by PCR analysis. Interestingly, the Whipple PCR that was performed on the synovial fluid collected during procedure was negative. There is one previously reported similar cases where synovial fluid PCR failed to demonstrate the presence of Tropheryma Whipplei but the synovial tissue PCR was positive (79).

Bacterial culture in both synovial fluid and synovial membrane is a key examination for septic arthritis diagnosis. However, using those methods, infectious agents was isolated in only 41,2% of the patients (38.7 % of synovial fluid and 23.5 % of synovial membrane positive cultures) (80).Therefore, histological synovial cell infiltrate analysis is also relevant for septic arthritis assessment. A neutrophilic cellular infiltrate, has been showed to be highly associated with septic arthritis (81). Their presence inside of the synovial tissue is considered as a sufficient evidence for the diagnosis of septic arthritis. Regarding the data we present, the diagnosis of septic arthritis was established following the histological examination of 2 patients. Interestingly, after empiric antimicrobial therapy was commenced in these 2 patients, no relapse occurred within at least 6 years' follow-up for both. This analysis can also be useful in fibrocartilagenous joints (acromio-clavicular, pubic symphysis) where fluid is rarely found even in case of inflammation. Furthermore, we can conclude from our data, that no patient of our cohort has been further diagnosed with infectious arthritis. This technique can therefore be considered as reliable to rule out septic arthritis assessment, permitting thus for local treatments such as steroids injections.

More rarely, synovial biopsy can be performed for synovial tumour assessment, especially villo nodular synovitis or osteochondromatosis. The 2 patients in our cohort diagnosed with

villo-nodular synovitis underwent surgical synovectomy. The histological examination of the tissue confirmed those findings.

For the biopsy to be useful in clinical practice, the quality of the biopsies retrieved has to be good. Quality of a synovial biopsy has been defined for research recently (53). But no definition has been given for the clinical setting yet. In our study, we defined good quality as: sufficient size defined by synovial tissue area > 0,5mm^2, preserved tissue allowing assessment by pathologists and presence of lining layer. In our cohort, the quality was good enough to allow a histological examination in all biopsies retrieving synovial tissue. Lining layer was found in 92.2% of the cases. In some instances, the lining layer could be identified but was not connected to the main biopsy, which may have occurred during tissue processing or may represent separation due to fibrin deposition in case of ulcerative synovitis.

No study has thus far demonstrated a predictive clinical value for histological findings in identifying those with early arthritis or those that will go on to have an aggressive disease course (63,66,67). Indeed, multiple studies tried to determine histological cell infiltrates patterns matching with different rheumatologic conditions. There is undeniable differences between RA and Psoriatic arthritis (29,82), RA and Ankylosing Spondylitis (AS) (83) and RA and osteoarthritis (OA) (23,84). OA synovial membrane is known to show less inflammatory infiltrate and less vascularity than their inflammatory counterparts (RA, PsA, AS). RA synovium has been described to show a higher number of B cells and more rarely ectopic follicles, helping in the diagnosis. The high grade synovitis features are more consistent with RA (3). However, despite those differences, no algorithm is able to predict the evolution in early arthritis (81).

Given this, the histopathologist was rarely able to determine the type of inflammatory arthritis. However, by ruling out or confirming infectious arthritis or synovial tumour, it is clear enough that US guided synovial biopsy is helpful on patients with remaining unknown diagnosis despite synovial fluid analysis, X-ray, CT scan and/or MRI examinations. In our setting, synovial biopsies allowed to treat some patients by achieving a definite diagnosis, or to give systemic immunosuppressive or local therapies such as intraarticular steroid injections. We acknowledge that our work has limitations. One limitation is the monocentric design of our study. The biopsies were performed by a trained investigator and the

pathologists in our centre have an expertise in biopsy assessment. This could be a limit for the generalization of those results. Although all patients had 3 to 8 biopsies taken, 55% of them had a single fragment sent to pathology department. This might be another limitation.

Finally, one of the main concerns about any procedure is its tolerance. In our cohort, one patient treated with salicylic acid presented with knee haemarthrosis 48 hours after the procedure. Overall, in our cohort, the adverse effects rate was 1.35% (IC 95 -1.3-4) (1/74) and no severe adverse event (life-threatening, leading to patient admission in hospital or with a risk of sequelae) occurred. The arthroscopic biopsies have the advantage to be retrieved under direct vision and therefore allow a histological analysis of the inflamed areas within the joint. However, this procedure is more invasive and has multiple adverse effects (joint infection; wound infection; haemarthrosis; deep venous thrombosis; neurological damage, thrombophlebitis) (52).

V. CONCLUSION

Our study highlights the potential diagnostic role of synovial biopsy. To our knowledge, it is the first study describing indications, tolerability, rate of success, diagnosis role and quality of ultrasound guided synovial biopsy in the clinical setting. Ultrasound guided synovial biopsy is performed in clinical practice in a heterogeneous population with variant clinical features. The success rate of the procedure remains high with only rare and minor complications. 13.3% achieved a definitive diagnosis leading to a specific treatment. In other patients, we could rule out the diagnosis of septic arthritis. Therefore, this procedure should not only be used for research purposes, but may also be used routinely in undifferentiated arthritis.

VI. REFERENCES

1. Simkin PA. Physiology of normal and abnormal synovium. Semin Arthritis Rheum. déc 1991;21(3):179‑83.

2. Barland P, Novikoff AB, Hamerman D. Electron microscopy of the human synovial membrane. J Cell Biol. 1962;14(2):207–220.

3. Krenn V, Morawietz L, Burmester G-R, Kinne RW, Mueller-Ladner U, Muller B, et al. Synovitis score: discrimination between chronic low-grade and high-grade synovitis. Histopathology. oct 2006;49(4):358‑64.

4. Krenn V, Morawietz L, Häupl T, Neidel J, Petersen I, König A. 15 years of the histopathological synovitis score, further development and review: A diagnostic score for rheumatology and orthopaedics. Pathol - Res Pract. janv 2002;198(5):317‑25.

5. Pitzalis C, Kelly S, Humby F. New learnings on the pathophysiology of RA from synovial biopsies: Curr Opin Rheumatol. mai 2013;25(3):334‑44.

6. Ea H.-K., Bazille C., Lioté F. Histologie et physiologie de la membrane synoviale. In: EMC (Elsevier Masson SAS, Paris), Appareil Locomoteur, 14-004-A-10,2008.

7. Ghadially FN. Fine structure of synovial joints. In: Butterworths, London, pp 1-41. (1983).

8. Kennedy A, Fearon U, Veale DJ, Godson C. Macrophages in synovial inflammation. Front Immunol. 2011;2:52.

9. Revell PA. Synovial lining cells. Rheumatol Int. 1989;9(2):49–51.

10. Athanasou NA, Quinn J. Immunocytochemical analysis of human synovial lining cells: phenotypic relation to other marrow derived cells. Ann Rheum Dis. mai 1991;50(5):311‑5.

11. Edwards JCW, Blades S, Cambridge G. Restricted expression of FcγRIII (CD16) in synovium and dermis: implications for tissue targeting in rheumatoid arthritis (RA). Clin Exp Immunol. juin 1997;108(3):401‑6.

12. Hamann J, Wishaupt JO, Van Lier RA, Smeets TJ, Breedveld FC, Tak PP. Expression of the activation antigen CD 97 and its ligand CD 55 in rheumatoid synovial tissue. Arthritis Rheum. 1999;42(4):650–658.

13. Tanaka M, Nagai T, Tsuneyoshi Y, Sunahara N, Matsuda T, Nakamura T, et al. Expansion of a unique macrophage subset in rheumatoid arthritis synovial lining layer. Clin Exp Immunol. oct 2008;154(1):38‑47.

14. Xu H, Manivannan A, Dawson R, Crane IJ, Mack M, Sharp P, et al. Differentiation to the CCR2+ inflammatory phenotype in vivo is a constitutive, time-limited property of blood monocytes and is independent of local inflammatory mediators. J Immunol. 2005;175(10):6915–6923.

15. Hamann J, Wishaupt JO, van Lier RA, Smeets TJ, Breedveld FC, Tak PP. Expression of the activation antigen CD97 and its ligand CD55 in rheumatoid synovial tissue. Arthritis Rheum. avr 1999;42(4):650‑8.

16. Li P, Sanz I, O'Keefe RJ, Schwarz EM. NF-kappa B regulates VCAM-1 expression on fibroblast-like synoviocytes. J Immunol Baltim Md 1950. 1 juin 2000;164(11):5990‑7.

17. Lee DM, Kiener HP, Agarwal SK, Noss EH, Watts GFM, Chisaka O, et al. Cadherin-11 in synovial lining formation and pathology in arthritis. Science. 16 févr 2007;315(5814):1006‑10.

18. Elshabrawy HA, Essani AE, Szekanecz Z, Fox DA, Shahrara S. TLRs, future potential therapeutic targets for RA. Autoimmun Rev. 2017 Feb;16(2):103-113.

19. Ambarus CA, Noordenbos T, de Hair MJ, Tak PP, Baeten DL. Intimal lining layer macrophages but not synovial sublining macrophages display an IL-10 polarized-like phenotype in chronic synovitis. Arthritis Res Ther. 2012;14(2):1.

20. Haywood L, Walsh DA. Vasculature of the normal and arthritic synovial joint. Histol Histopathol. janv 2001;16(1):277‑84.

21. Wilkinson LS, Edwards JC. Demonstration of lymphatics in human synovial tissue. Rheumatol Int. 1991;11(4‑5):151‑5.

22. Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, Bingham CO 3rd, Birnbaum NS, Burmester GR, Bykerk VP, Cohen MD, Combe B, Costenbader KH, Dougados M, Emery P, Ferraccioli G, Hazes JM, Hobbs K, Huizinga TW, Kavanaugh A, Kay J, Kvien TK, Laing T, Mease P, Ménard HA, Moreland LW, Naden RL, Pincus T, Smolen JS, Stanislawska-Biernat E, Symmons D, Tak PP, Upchurch KS, Vencovsky J, Wolfe F, Hawker G. 2010 rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism collaborative initiative. Ann Rheum Dis 2010 Sep;69(9):1580-8 Erratum in: Ann Rheum Dis 2010 Oct;69(10):1892.

23. Baeten D, Demetter P, Cuvelier C, et al. Comparative study of the synovial histology in rheumatoid arthritis, spondyloarthropathy, and osteoarthritis: influence of disease duration and activity. Ann Rheum Dis. 2000;59(12):945–953.

24. Krenn V, Morawietz L, Häupl T, Neidel J, Petersen I, König A. Grading of chronic synovitis—a histopathological grading system for molecular and diagnostic pathology. Pathol-Res Pract. 2002;198(5):317–325.

25. van der Helm-van Mil AH, le Cessie S, van Dongen H, Breedveld FC, Toes RE, Huizinga TW. A prediction rule for disease outcome in patients with recent-onset undifferentiated arthritis: how to guide individual treatment decisions. Arthritis Rheum 2007 Feb;56(2):433-40.

26. Visser H, le Cessie S, Vos K, Breedveld FC, Hazes JM. How to diagnose rheumatoid arthritis early: a prediction model for persistent (erosive) arthritis. Arthritis Rheum. 2002 Feb;46(2):357-65.

27. Alves C, Luime JJ, van Zeben D, Huisman AM, Weel AE, Barendregt PJ, Hazes JM. Diagnostic performance of the ACR/EULAR 2010 criteria for rheumatoid arthritis and two diagnostic algorithms in an early arthritis clinic (REACH). Ann Rheum Dis 2011 Sep;70(9):1645-7.

28. Baeten D, Demetter P, Cuvelier C, Van den Bosch F, Kruithof E, Van Damme N, et al. Comparative study of the synovial histology in rheumatoid arthritis, spondyloarthropathy, and osteoarthritis: influence of disease duration and activity. Ann Rheum Dis. déc 2000;59(12):945‑53.

29. Kruithof E, Baeten D, De Rycke L, et al. Synovial histopathology of psoriatic arthritis, both oligo-and polyarticular, resembles spondyloarthropathy more than it does rheumatoid arthritis. Arthritis Res Ther. 2005;7(3):R569–R580.

30. Lauwerys BR, Hernández-Lobato D, Gramme P, Ducreux J, Dessy A, Focant I, Ambroise J, Bearzatto B, Nzeusseu Toukap A, Van den Eynde BJ, Elewaut D, Gala JL, Durez P, Houssiau FA, Helleputte T, Dupont P. Heterogeneity of synovial molecular patterns in patients with arthritis. PLoS One 2015 Apr 30;10(4):e0122104

31. Orr C, Sousa E, Boyle DL, Buch MH, Buckley CD, Cañete JD, et al. Synovial tissue research: a state-of-the-art review. Nat Rev Rheumatol. août 2017;13(8):463‑75.

32. van de Sande MGH, Thurlings RM, Boumans MJH, Wijbrandts CA, Modesti MG, Gerlag DM, et al. Presence of lymphocyte aggregates in the synovium of patients with early arthritis in relationship to diagnosis and outcome: is it a constant feature over time? Ann Rheum Dis. avr 2011;70(4):700‑3.

33. Klimiuk PA, Sierakowski S, Latosiewicz R, Skowronski J, Cylwik JP, Cylwik B, et al. Histological patterns of synovitis and serum chemokines in patients with rheumatoid arthritis. J Rheumatol. sept 2005;32(9):1666‑72.

34. Thurlings RM, Wijbrandts CA, Mebius RE, Cantaert T, Dinant HJ, van der Pouw-Kraan TCTM, et al. Synovial lymphoid neogenesis does not define a specific clinical rheumatoid arthritis phenotype. Arthritis Rheum. juin 2008;58(6):1582‑9.

35. Cañete JD, Celis R, Moll C, Izquierdo E, Marsal S, Sanmartí R, et al. Clinical

significance of synovial lymphoid neogenesis and its reversal after anti-tumour necrosis factor alpha therapy in rheumatoid arthritis. Ann Rheum Dis. mai 2009;68(5):751‑6.

36. Townsend MJ. Molecular and cellular heterogeneity in the Rheumatoid Arthritis synovium: clinical correlates of synovitis. Best Pract Res Clin Rheumatol. 2014 Aug;28(4):539-49.

37. Orr C, Najm A, Biniecka M, McGarry T, Ng C-T, Young F, et al. Synovial Immunophenotype and Anti-Citrullinated Protein Antibodies in RA patients: Relationship to treatment response and radiological prognosis. Arthritis Rheumatol Doi 101002art40218 Epub Ahead Print. 21 juill 2017;

38. Ng CT, Biniecka M, Kennedy A, McCormick J, Fitzgerald O, Bresnihan B, et al. Synovial tissue hypoxia and inflammation in vivo. Ann Rheum Dis. juill 2010;69(7):1389‑95.

39. Bergman GJD, Hochberg MC, Boers M, Wintfeld N, Kielhorn A, Jansen JP. Indirect comparison of tocilizumab and other biologic agents in patients with rheumatoid arthritis and inadequate response to disease-modifying antirheumatic drugs. Semin Arthritis Rheum. juin 2010;39(6):425‑41.

40. Lequerré T, Gauthier-Jauneau A-C, Bansard C, Derambure C, Hiron M, Vittecoq O, et al. Gene profiling in white blood cells predicts infliximab responsiveness in rheumatoid arthritis. Arthritis Res Ther. 2006;8(4):R105.

41. Sekiguchi N, Kawauchi S, Furuya T, Inaba N, Matsuda K, Ando S, et al. Messenger ribonucleic acid expression profile in peripheral blood cells from RA patients following treatment with an anti-TNF-alpha monoclonal antibody, infliximab. Rheumatol Oxf . juin 2008;47(6):780‑8.

42. Cañete JD, Celis R, Moll C, Izquierdo E, Marsal S, Sanmartí R, et al. Clinical significance of synovial lymphoid neogenesis and its reversal after anti-tumour necrosis factor alpha therapy in rheumatoid arthritis. Ann Rheum Dis. mai 2009;68(5):751‑6.

43. Klaasen R, Thurlings RM, Wijbrandts CA, van Kuijk AW, Baeten D, Gerlag DM, et al. The relationship between synovial lymphocyte aggregates and the clinical response to infliximab in rheumatoid arthritis: a prospective study. Arthritis Rheum. nov 2009;60(11):3217‑24.

44. van der Pouw Kraan TC, Wijbrandts CA, van Baarsen LG, Rustenburg F, Baggen JM, Verweij CL, et al. Responsiveness to anti-tumour necrosis factor alpha therapy is related to pre-treatment tissue inflammation levels in rheumatoid arthritis patients. Ann Rheum Dis. avr 2008;67(4):563‑6.

45. Badot V, Galant C, Nzeusseu Toukap A, Theate I, Maudoux A-L, Van den Eynde

BJ, et al. Gene expression profiling in the synovium identifies a predictive signature of absence of response to adalimumab therapy in rheumatoid arthritis. Arthritis Res Ther. 2009;11(2):R57.

46. Lindberg J, Wijbrandts CA, van Baarsen LG, Nader G, Klareskog L, Catrina A, et al. The gene expression profile in the synovium as a predictor of the clinical response to infliximab treatment in rheumatoid arthritis. PloS One. 25 juin 2010;5(6):e11310.

47. Kavanaugh A, Rosengren S, Lee SJ, Hammaker D, Firestein GS, Kalunian K, et al. Assessment of rituximab's immunomodulatory synovial effects (ARISE trial). 1: clinical and synovial biomarker results. Ann Rheum Dis. mars 2008;67(3):402‑8.

48. Thurlings RM, Vos K, Wijbrandts CA, Zwinderman AH, Gerlag DM, Tak PP. Synovial tissue response to rituximab: mechanism of action and identification of biomarkers of response. Ann Rheum Dis. juill 2008;67(7):917‑25.

49. Forestier J. Instrumentation pour biopsie médicale. In: C.R. Séances Soc. Biol. Filiales. Paris, 1932;110:186.

50. Polley HF, Bickel WH. Punch biopsy of synovial membrane. Ann Rheum Dis. 1951;10(3):277.

51. Parker RH, Pearson CM. A simplified synovial biopsy needle. Arthritis Rheum. 1963;6(2):172–176.

52. Kane D, Veale DJ, FitzGerald O, Reece R. Survey of arthroscopy performed by rheumatologists. Rheumatol Oxf. 2002;41(2):210-5.

53. Kelly S, Humby F, Filer A, et al. Ultrasound-guided synovial biopsy: a safe, well-tolerated and reliable technique for obtaining high-quality synovial tissue from both large and small joints in early arthritis patients. Ann Rheum Dis. 2015;74(3):611‑7.

54. Marin F, Lasbleiz J, Albert JD, et al. Technique et évaluation du guidage échographique pour la réalisation de biopsies synoviales. J Radiol 2006;87:561-5.

55. Van Vugt RM, Van Dalen A, Bijlsma JWJ. Ultrasound guided synovial biopsy of the wrist. Scand J Rheumatol. 1997;26(3):212–214.

56. Gonçalves B, Ambrosio C, Serra S, Alves F, Gil-Agostinho A, Caseiro-Alves F. US-guided interventional joint procedures in patients with rheumatic diseases—When and how we do it? Eur J Radiol. sept 2011;79(3):407‑14.

57. Koski JM. Ultrasound guided synovial biopsy using portal and forceps. Ann Rheum Dis. 2005;64(6):926‑9.

58. Scirè CA, Epis O, Codullo V, et al. Immunohistological assessment of the synovial

tissue in small joints in rheumatoid arthritis: validation of a minimally invasive ultrasound-guided synovial biopsy procedure. Arthritis Res Ther. 2007;9(5):R101.

59. Man GS, Mologhianu G. Osteoarthritis pathogenesis-a complex process that involves the entire joint. J Med Life. 2014;7(1):37.

60. McInnes IB, Schett G. The pathogenesis of rheumatoid arthritis. N Engl J Med. 2011;365(23):2205-19.

61. Rooney M, Whelan A, Feighery C, Bresnihan B. Changes in lymphocyte infiltration of the synovial membrane and the clinical course of rheumatoid arthritis. Arthritis Rheum. 1989;32(4):361-9.

62. Bresnihan B, Tak PP. Synovial tissue analysis in rheumatoid arthritis. Baillieres Best Pract Res Clin Rheumatol. 1999;13(4):645-59.

63. Tak PP. Analysis of synovial biopsy samples: opportunities and challenges. Ann Rheum Dis. 2000;59(12):929–930.

64. Tak PP. Lessons learnt from the synovial tissue response to anti-rheumatic treatment. Rheumatol Oxf. 2000;39(8):817–820.

65. Tak PP, Van Der Lubbe PA, Cauli A, Daha MR, Smeets TJ, Kluin PM, et al. Reduction of synovial inflammation after anti-CD4 monoclonal antibody treatment in early rheumatoid arthritis. Arthritis Rheum. 1995;38(10):1457-65.

66. Bresnihan B. Are synovial biopsies of diagnostic value? Arthritis Res Ther. 2003;5(6):271–278.

67. Gerlag DM, Tak PP. How to perform and analyse synovial biopsies. Best Pract Res Clin Rheumatol. 2013;27(2):195‑207.

68. van der Heijden IM, Wilbrink B, Vije AEM, Shouls LM, Breedveld FC, Tak PP. Detection of bacterial DNA in serial synovial samples obtained during antibiotic treatment from patients with septic arthritis. Arthritis Rheum. 1999;42(10):2198–2203.

69. Graf SW, Buchbinder R, Zochling J, Whittle SL. The accuracy of methods for urate crystal detection in synovial fluid and the effect of sample handling: A systematic review. Clin Rheumatol. 2013;32(2):225‑32.

70. Beaulé V, Larédo JD, Cywiner C, Bard M, Tubiana JM. Synovial membrane: percutaneous biopsy. Radiology. 1990;177(2):581-5.

71. Parlier-Cuau V, Hamzé.B, Bellaïche L, Wybier M, Laredo JD. Biopsie percutanée de la synoviale : technique. Feuillet de Radiologie 1999;39:225-30.

72. Altman RD, Gray R. Diagnostic and therapeutic uses of the arthroscope in

rheumatoid arthritis and osteoarthritis. Am J Med. 1983 Oct 31;75(4B):50-5.

73. Youssef PP, Smeets TJ, Bresnihan B, et al. Microscopic measurement of cellular infiltration in the rheumatoid arthritis synovial membrane: a comparison of semiquantitative and quantitative analysis. Br J Rheumatol. 1998;37(9):1003-7.

74. Jacob D, Cyteval C, Moinard M. [Interventional sonography]. J Radiol. 2005;86(12 Pt 2):1911-23.

75. Cardinal E, Chhem RK, Beauregard CG. Ultrasound-guided interventional procedures in the musculoskeletal system. Radiol Clin North Am. 1998;36(3):597-604.

76. Lazarou I, D'Agostino M-A, Naredo E, Humby F, Filer A, Kelly SG. Ultrasound-guided synovial biopsy: a systematic review according to the OMERACT filter and recommendations for minimal reporting standards in clinical studies. Rheumatol Oxf. 2015 Oct;54(10):1867-75.

77. Bresnihan B, Cunnane G, Youssef P, Yanni G, Fitzgerald O, Mulherin D. Microscopic measurement of synovial membrane inflammation in rheumatoid arthritis: proposals for the evaluation of tissue samples by quantitative analysis. Rheumatol Oxf. 1998;37(6):636–642.

78. van de Sande MGH, Gerlag DM, Lodde BM, et al. Evaluating antirheumatic treatments using synovial biopsy: a recommendation for standardisation to be used in clinical trials. Ann Rheum Dis. 2011;70(3):423‑7.

79. O'Duffy JD, Griffing WL, Li CY, Abdelmalek MF, Persing DH. Direct Detection of Tropheryma whippelii in Synovial Fluid and Tissue. Arthritis Rheum. 1999 Apr;42(4):812-7.

80. Madruga Dias J, Costa MM, Pereira da Silva JA, Viana de Queiroz M. Septic arthritis: patients with or without isolated infectious agents have similar characteristics. Infection. avr 2014;42(2):385‑91.

81. Della Beffa C, Slansky E, Pommerenke C et al. C. The Relative Composition of the Inflammatory Infiltrate as an Additional Tool for Synovial Tissue Classification. PLoS ONE. 8 août 2013;8(8):e72494.

82. van Kuijk AWR, Tak PP. Synovitis in Psoriatic Arthritis: Immunohistochemistry, Comparisons With Rheumatoid Arthritis, and Effects of Therapy. Curr Rheumatol Rep. 2011;13(4):353‑9.

83. Kidd BL, Moore K, Walters MT, Smith JL and Cawley MID. Immunohistological features of synovitis in ankylosing spondylitis : a comparison with rheumatoid arthritis. Annals of the Rheumatic Diseases, 1989; 48, 92-98.

84. Pessler F, Dai L, Diaz-Torne C et al. The synovitis of ‘‘non-inflammatory’’ orthopaedic arthropathies: a quantitative histological and immunohistochemical analysis. Ann Rheum Dis. 2008 Aug;67(8):1184-7.

VII. ANNEXES

The Journal of Rheumatology

The Journal of Rheumatology

Success Rate and Utility of Ultrasound-guided Synovial Biopsies in Clinical Practice

Aurélie Najm, Carl Orr, Marie-Françoise Heymann, Géraldine Bart, Douglas J. Veale and Benoît Le Goff

DOI: 10.3899/jrheum.151441
http://www.jrheum.org/content/early/2016/10/06/jrheum.151441

Success Rate and Utility of Ultrasound-guided Synovial Biopsies in Clinical Practice

Aurélie Najm, Carl Orr, Marie-Françoise Heymann, Géraldine Bart, Douglas J. Veale, and Benoît Le Goff

ABSTRACT. ***Objective.*** The utility of synovial biopsy in increasing our understanding of the pathogenesis of inflammatory arthropathies, as well as in evaluating treatments, is well established. Ultrasound (US) allows synovial assessment and therefore assists in biopsying synovial tissue in a safe and well-tolerated manner. This study's objectives were to (1) determine the rate of success in retrieving synovial tissue using US guidance, (2) describe the indications for US-guided synovial biopsies in the clinical setting, (3) determine how frequently the synovial biopsy can lead to a clear diagnosis, and (4) assess the quality of the synovial tissue obtained using this technique.

Methods. Synovial biopsies of small and large joints were performed under US guidance between February 2007 and December 2014 using a semiautomatic core biopsy needle. The biopsy procedure was considered successful if synovial tissue was found at histological examination.

Results. Seventy-four patients with undifferentiated arthritis underwent 76 synovial biopsies. The success rate in retrieving synovial tissue was 81.6% (62/76). One patient taking acetyl salicylic acid at 75 mg at the time of the biopsy presented with hemarthrosis 48 h after the procedure, which resolved following simple arthrocentesis. A definitive diagnosis was achieved in 16% of the patients where synovial tissue was sampled successfully.

Conclusion. US-guided synovial biopsies in clinical practice can be performed safely on patients with undifferentiated arthritis and with heterogeneous presentations. The rate of success in acquiring synovial tissue is high. The procedure usually retrieves quality tissue and leads to a definite diagnosis in a significant minority of patients. (J Rheumatol First Release October 15 2016; doi:10.3899/jrheum.151441)

Key Indexing Items:

BIOPSY SYNOVIAL MEMBRANE ULTRASONOGRAPHY
DIAGNOSIS EARLY ARTHRITIS

Synovial tissue is the principal target and end organ involved in the pathogenesis of multiple articular disease processes[1,2]. Synovial tissue analysis has been widely used for basic science and translational and clinical research. Moreover, synovial assessment allows for the study of many aspects of disease processes including pathogenesis[3], the identification of the relevant target's clinical features[4], diagnosis, and prognosis[5], as well as assisting in assessments of response to treatment[6,7,8].

Histological and immunohistological synovial assessment is also used as a diagnostic tool[9]. Indeed, it is especially useful for identifying arthritis of an infectious etiology when synovial fluid (SF) or blood analysis (Gram, Ziehl) and cultures are negative or in cases where empiric antimicrobial therapy has been commenced before it has been possible to examine the SF[10]. The bacterial broad range 16S ribosomal RNA can also be tracked down by PCR on synovial tissue[11]. The same methods allow identification of fungal, mycobacterial, spirochetes, and *Tropheryma whipplei* in the joint. False negatives for monosodium urate crystals and calcium pyrophosphate occur frequently at microscopic examination of the SF[12], and synovial tissue assessment can be helpful with typical histological features. Finally, synovial benign tumors such as primary or secondary osteochondromatosis or villonodular synovitis can be diagnosed as well, showing specific macroscopic and histological pattern.

There are several techniques to obtain synovial tissue from the joints. Synovial biopsy was performed by Forestier using a needle blindly introduced into the knee joint[13]. Polley and Bickel[14] and Parker and Pearson[15] described new smaller-diameter needles that have been widely used over the years for knee synovial biopsies. Beaulé, *et al*[16] and Parlier-Cuau, *et al*[17] then described a technique of synovial biopsy under

From the Department of Rheumatology, and Department of Pathology, Hôtel-Dieu Hospital, Nantes, France; University College Dublin Department of Rheumatology, St. Vincent's Hospital, Dublin, Ireland.

A. Najm, MD, Resident, Department of Rheumatology, Hôtel-Dieu Hospital; C. Orr, MD, University College Dublin Department of Rheumatology, St. Vincent's Hospital; M.F. Heymann, MD, Department of Pathology, Hôtel-Dieu Hospital; G. Bart, MD, Resident, Department of Rheumatology, Hôtel-Dieu Hospital; D.J. Veale, MD, PhD, Professor, University College Dublin Department of Rheumatology, St. Vincent's Hospital; B. Le Goff, PhD, Assistant Professor, Department of Rheumatology, Hôtel-Dieu Hospital.

Address correspondence to Prof. B. Le Goff, Hôtel-Dieu Hospital, 1 place Alexis-Ricordeau, 44093 Nantes cedex 1, France.
E-mail: benoit.legoff@chu-nantes.fr

Accepted for publication August 16, 2016.

direct visualization under fluoroscopy with a semiautomatic Tru-Cut needle. This technique allows the performance of multisite biopsies such as in the hips, shoulders, elbows, ankles, and wrists. Synovial biopsies were later performed under direct vision using 2 portals through an arthroscope[18]. Although this technique is usually well tolerated[9], it remains invasive, expensive, and not yet widely available. Moreover, it has been shown that microscopic measurements of synovial inflammation do not differ between biopsies taken blindly or under guided vision[19].

More recently, ultrasound (US)-guided synovial biopsies have been developed. Musculoskeletal US is very commonly used today, especially for guiding interventional procedures[20,21]. This technique has the benefit of being low-cost, rapidly and easily performed without the need for exposing the patient to ionizing radiation, and widely available[22]. It is more practical than arthroscopy for biopsying small joints and allows guidance to the thickest synovial zones. Moreover, Kelly, *et al*[23] reported that increasing synovial thickness on US correlated with increasing grades of synovitis on histological examination. However, few studies have reported on synovial biopsies performed in routine clinical practice[24,25]. It is unknown whether the success and the quality of the biopsy are the same as the one performed in a research setting. Finally, their clinical utility is still a matter of debate.

The aims of our study were to (1) determine the rate of success in retrieving synovial tissue using US guidance, (2) describe the indications for US-guided synovial biopsies in the clinical setting, (3) determine how frequently the synovial biopsy can lead to a clear diagnosis, and (4) assess the quality of the synovial tissue obtained using this technique.

MATERIALS AND METHODS

Patients and histological diagnosis. We included all patients who underwent a US-guided synovial biopsy between February 2007 and December 2014 in Nantes University Hospital for arthritis without a definite diagnosis based on the history, clinical examination, or imaging. Ethics approval was not required in accordance with the policy of our institution. During this service evaluation study, we collected epidemiological (age, sex) and clinical data (clinical presentation, indication, biopsied joint, complications) using a standardized form. Final histological diagnosis was reported by 3 pathologists who had an expertise in assessing synovial tissue in a formal report based on H&E staining. Patients were followed to determine the clinical course of their symptoms.

US-guided synovial biopsies. Synovial biopsies were performed under US guidance using a Philips HD11 XE US machine and a 7–13 MHz transducer from Philips Healthcare. They were performed in an outpatient and inpatient setting depending on the patient's presentation. All patients underwent a thorough assessment of the joint to be biopsied. Vascular and nervous structures nearby were identified and synovial thickness was assessed.

All the biopsy procedures were performed by 1 operator (BLG) who had an expertise in US examination under sterile technique (wearing gown, sterile gloves, mask, and a surgical cap). Skin disinfection was processed with a 5-step protocol using iodine polyvidone or antibacterial cleanser if the patient had a history of iodine allergy. The joint was draped and a sterile field was generated. The transducer was covered with sterile gel and sterile sheath. Anesthesia was performed, injecting 5 to 10 ml of lidocaine 2% in the subcutaneous tissue and up to the joint capsule. If an effusion were present, SF was withdrawn and sent to the laboratory for cell count, crystal microscopy, bacteriological, mycobacteriological, and/or fungal analysis depending on the patient's clinical history and features. A semiautomatic guillotine biopsy Tru-Cut needle from Temnos was used for all the biopsies. The caliber used was 16-gauge for small and intermediate joints or 14-gauge for large joints such as the hips, shoulders, and knees. Coaxial needle was inserted under US guidance through the skin until it reached the articular cavity. The coaxial needle was positioned in intimate contact with the synovium. The semiautomatic guillotine biopsy Tru-Cut needle was then inserted through the cannula of the coaxial needle, still under US guidance. Once positioned within the zone of interest of the synovial tissue, the Tru-Cut needle was triggered, collecting a piece of synovial tissue according to the size of the joint. This Tru-Cut needle was repeatedly inserted through the coaxial needle and triggered to obtain the appropriate number of samples. Then, these 2 needles were removed and a bandage was applied. Patients were recommended to have 48 h of rest after the procedure.

Depending on the indication of the biopsy and the size of the joint, 3 to 8 biopsies were performed per procedure and sent for bacteriological, mycobacteriological, and/or fungal examination in appropriate laboratories. At least 1 sample was fixed in formalin 4%, embedded in paraffin, and sent to the pathology laboratory. When the clinical history was relevant, extra samples were sent for universal bacterial PCR (ARN 16S), universal fungal PCR (ARN 18S), and *T. whipplei* or Lyme PCR.

Analysis of the quality and quantity of the synovial tissue retrieved during synovial biopsies. All the synovial biopsies were blindly read by 1 rheumatologist (AN). These characteristics were assessed in a standardized manner with NDP viewer software: the number of samples per patient; the presence or absence of synovial tissue; the presence or absence of a synovial lining layer; the length, width, and total area of the biopsy (mm^2); and the area of proper synovial tissue (mm^2). These findings were compared with the histological findings described in the pathology reports, which were the gold standard. In case of disagreement between rheumatologist and pathologist, an expert reader (DV) was responsible for the final decision. We considered the biopsy successful when synovial tissue was seen at the histological examination. Good quality was defined as the following: sufficiently sized (> 0.5 mm^2)[26] preserved tissue allowing assessment by pathologists and presence of lining layer.

Statistical analysis. Mean and median were used to describe quantitative data according to their Gaussian distribution. Number and percentage were used to report qualitative data. Fisher's exact test had been used to compare percentage. coefficient calculation was used to assess the interobserver reliability for histological analysis. A p value < 0.05 was considered as statistically significant. All statistics were made through GraphPad Prism 6.0 software.

RESULTS

Patient characteristics. Seventy-four patients underwent 76 US-guided synovial biopsy procedures. Demographic and clinical features of patients included in our study are shown in Table 1. Mean age was 57 years (range 13–86 yrs) and there were 39 men (52.7%). Most of the patients presented with an undifferentiated chronic monoarthritis (54.1, n = 40). The number of joints and their percentages among the patients were as follows: 46 knees (60.5%), 6 ankles (8%), 6 wrists (8%), 5 shoulders (7%), 4 hips (5%), 2 elbows, 2 sternoclavicular joints, 2 metatarsophalangeal joints, 1 pubic symphysis, 1 acromioclavicular joint, and 1 peroneal tenosynovitis. Patients were mainly referred to rule out the diagnosis of septic arthritis (82.4%, n = 61).

Table 1. Demographic and clinical features of the patients. Values are n (%) unless otherwise specified.

Variables	Values
Female	35 (47.3)
Male	39 (52.7)
Age, yrs, mean (range)	57 (13–86)
Indications	
Undifferentiated chronic monoarthritis	40 (54.1)
Acute monoarthritis	18 (24.0)
Chronic undifferentiated oligoarthritis	7 (9.3)
Chronic polyarthritis	6 (8.0)
Chronic bursitis	1 (1.3)
Chronic tenosynovitis	1 (1.3)
Acute polyarthritis	1 (1.3)

US-guided biopsy procedure was safe and successful. Overall, 62 of the 76 biopsies (81.6%) yielded synovial tissue according to the pathologists' analysis. Within these 62 biopsies, the main histological finding was a nonspecific inflammatory mononuclear cell infiltrate (lymphocyte, monocytes, and plasma cells; 81%, n = 50). A mild neutrophil infiltrate was seen in 24 (50%) of these biopsies. Eight (13%) biopsies showed specific histological lesions (Figure 1). A major neutrophil cell infiltrate consistent with a septic arthritis was found in 2 cases. Two biopsies showed a synovial infiltration of positive Perl's siderophages (villonodular synovitis). One biopsy showed vascular and interstitial deposits of Sirius red staining protein consistent with amyloid light-chain amyloidosis. One biopsy contained tophi surrounded by lymphocytes and giant cells. One biopsy found dystrophic cartilage inside the synovial tissue, consistent with synovial osteochondromatosis. One biopsy showed an articular localization of lymphoma. Four biopsies retrieved normal synovial tissue without any inflammatory cell infiltrate (Table 2).

The 14 failed biopsies occurred in both small and large joints. Percentages of failed biopsies per joint were as follows: glenohumeral joints (n = 3/5, 60%), ankle (n = 3/6, 50%), hip (n = 2/4, 50%), wrist (n = 2/6, 33.3%), elbow (n = 1/2, 50%), sternoclavicular joint (n = 1/2, 50%), and knees (n = 2/46, 4.3%). In case of failure, histological analysis showed mainly connective and adipose tissue in 10 cases, fibrin and leukocytes in 3 cases, and tendon in 1 case. Tolerance per procedure was excellent. One patient taking acetyl salicylic acid at the time of the biopsy presented with a hemarthrosis 48 h after the procedure, which resolved following arthrocentesis within 1 week.

Overall, 10 (16.2%) definitive diagnoses were made based only on histological or PCR analysis of synovial tissue.

Longterm followup (mean 34.9 mos, range < 1 mo to 96 mos) and final diagnosis were available for 67 of the 74 patients, and 7 were lost to followup (Table 3). No patient has since been diagnosed with an infectious arthritis or villonodular synovitis or developed any complication of the biopsy procedure. In 3 of the cases where the diagnosis remained unclear despite the US-guided biopsy and in 2 cases of failed biopsy, patients underwent secondary procedures. One of them had an arthroscopic examination after the US-guided biopsy and 4 of them had an open synovectomy. One synovectomy allowed a diagnosis of chondrocalcinosis on pathological examination.

Quality and quantity of the synovial tissue retrieved after US-guided synovial biopsies. Finally, the synovial tissue retrieved was assessed for quality and quantity. For this purpose, we analyzed the histological characteristics per sample retrieved during the procedure (Figure 2). The median number of samples taken per patient was 1 (interquartile range 1–3), leading to a total of 125 samples available for analysis. Mean length and width of the biopsy samples were 6.34 mm (± 3.60) and 1.70 mm (± 0.77), respectively. The mean total area of the samples was 8.77 mm^2.

Biopsies showed synovial tissue at the histological examination in 102 samples (80.1%). The average area of synovial tissue in these samples was 6.36 mm^2, corresponding to 72.5% of the total area of biopsied tissue. The other types of tissue present on these biopsies were connective tissue in 101 cases (80.8%), adipose tissue in 42 cases (33.6%), tendon in 14 cases (11.2%), and fibrin in 24 cases (19.2%). The 23 samples retrieving no synovial tissue were composed of fibrin in 15 cases (12%), conjunctive and adipose tissue in 17 cases (13.6%), tendinous tissue in 3 cases (3.15%), cartilage in 3 cases (3.15%), and muscle in 1 case (0.8%).

Synovial lining layer was found in 92.6% of the successful biopsies.

We compared our histological final findings regarding presence or absence of synovial tissue with those of the pathologist and found 97.1% of agreement. Interobserver reliability for presence/absence of synovial tissue was high, with a | coefficient of 0.90 (95% CI 0.763–1.000).

DISCUSSION

Because synovial tissue analysis has been mostly used for research purposes, our study highlights the potential diagnostic involvement of synovial biopsy in routine clinical practice. To develop this technique in clinical practice, the patient needs to be offered a well-tolerated technique with an acceptable rate of success.

To date, 2 different techniques of US-guided synovial biopsies have been described. Both have been shown to be safe and well tolerated by the patients[22]. The first method requires a single portal with a flexible or rigid biopsy forceps. The portal is directly introduced inside the joint to perform biopsies[27]. The second technique, as outlined above, requires an empty coaxial needle that is inserted inside the joint and a semiautomatic guillotine-type needle that is inserted through the coaxial. The procedure is not painful after local anesthesia and once the coaxial needle is settled, and this technique

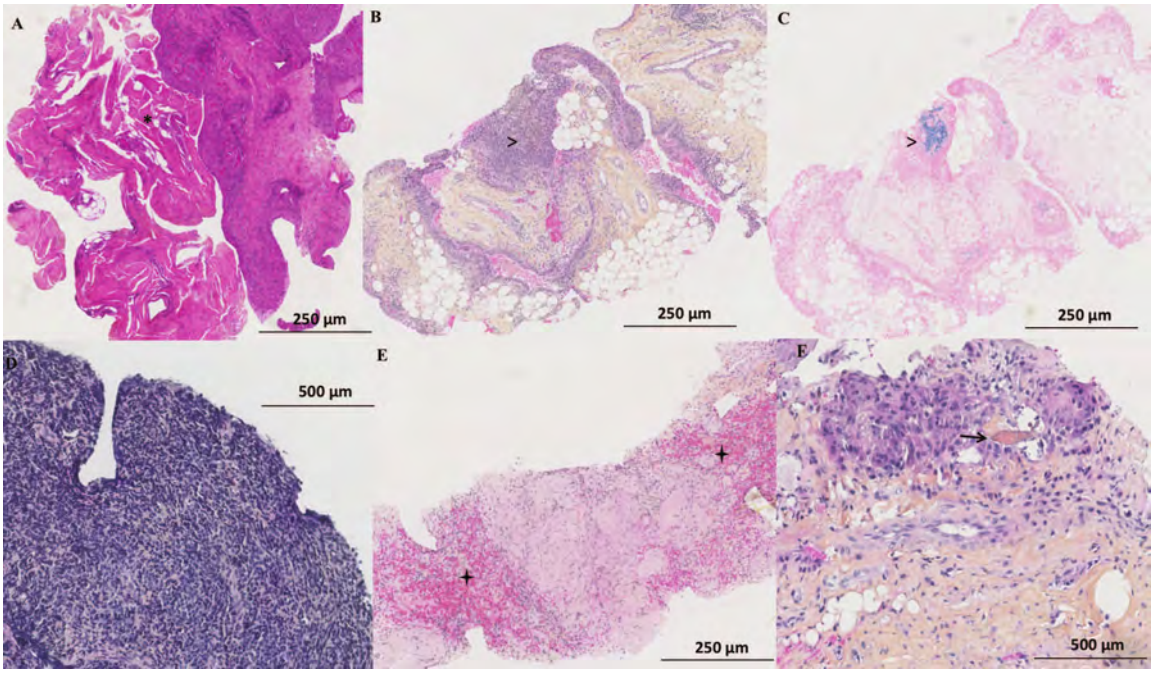

Figure 1. Synovial biopsies of 5 specific histological lesions. (A) Fibrin deposits with neutrophils infiltrate (asterisk). Septic arthritis. (B) Villonodular synovitis. H&E staining showing siderophages (arrowhead). (C) Villonodular synovitis with Perl's staining showing siderophages (arrowhead). (D) Cell infiltrate within synovial tissue in an articular lymphoma. (E) Amyloids (crosses) revealed by Sirius red staining. Amyloid light-chain amyloidosis. (F) Micro tophi surrounded by giant cells and lymphocytes (arrow) leading to gout diagnosis.

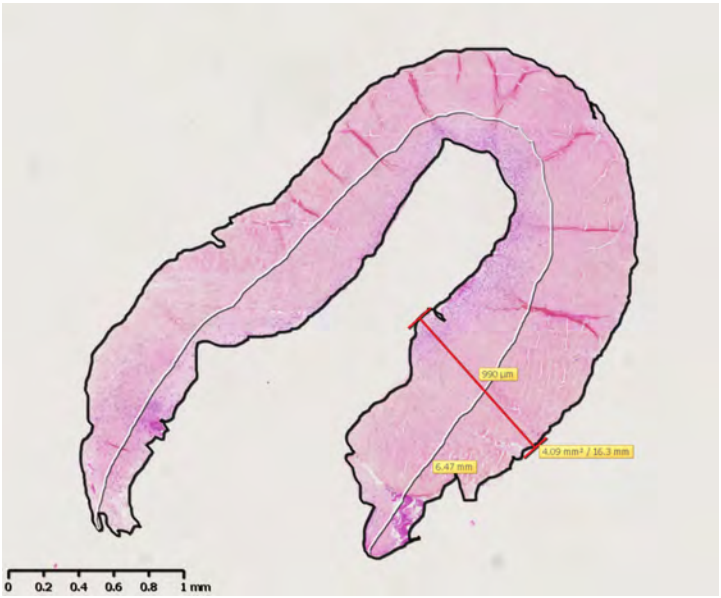

Figure 2. Example of the sample histological analysis. Black line is the global area measurement, red line is the width measurement, and white line in the length measurement.

Table 2. Histopathological analysis.

Histopathological Findings	No. Biopsies
Normal synovium	4
Inflamed synovium	50
Cell infiltrate:	
Lymphocytes	50
Plasma cells	22
Neutrophils	24
Specific lesions	8
Villonodular synovitis, shoulder and knee	2
Infectious arthritis*	2
Amyloid arthritis, knee	1
Articular localization of mantle B cell lymphoma, ankle	1
Gout, first MTP	1
Osteochondromatosis, knee	1
Failure	14

* Two infectious arthritis sites (hip, ankle) treated on typical histological aspect with no relapse after 6 weeks of empiric antibiotics. MTP: metatarsophalangeal.

Table 3. Overall final diagnosis after followup. Values are n (%).

Final Diagnosis	Values
Rheumatoid arthritis	7 (9.5)
Ankylosing spondylitis	2 (2.7)
Psoriatic arthritis	5 (6.8)
Degenerative arthropathy	12 (16.2)
Crystal arthropathy	4 (5.4)
Chondrocalcinosis	2 (2.7)
Gout	3 (4.1)
Villonodular synovitis	2 (2.7)
Osteochondromatosis	1 (1.4)
Giant cell arthritis	1 (1.4)
Behçet disease	1 (1.4)
Latent infectious arthritis	4 (5.4)
Others	2 (2.7)
Undifferentiated arthritis	21 (28.4)
Lost to followup	7 (9.5)
Total	74 (100)

allows retrieving several biopsies during the same procedure without moving the coaxial needle. To our knowledge, 5 other studies reporting their experience of US-guided synovial biopsies have been published to date. Two reported their experience using the first technique[27,28], 1 a technique using semiautomatic guillotine-type needle without coaxial needle[23], and 2 using the second technique outlined above[24,25].

The success rates in retrieving synovial tissue described by other authors vary from 89% to 100%[23,25,27,28,29]. The rate of success in our cohort was slightly lower, for several possible reasons. Our patients comprised a heterogeneous group regarding clinical features and the joints that were biopsied. There were also minor differences in techniques in 2 of the studies referended above. Moreover, no biopsies were done prior to 2007 in our center and 43% of the failures occurred within the first 18 months (6 of the 14 total failed biopsies), especially in more challenging joints such as the ankles, wrists, hips, or shoulders. This might correspond to the operator learning curve. However, our success rate remains equivalent to the highest rates described for synovial biopsies with blind needle (48%–85%)[30].

In our study, patients were referred mostly by their general practitioner or their rheumatologist with no clear diagnosis despite multiple punctures for SF analysis and computed tomography (CT) scans or magnetic resonance imaging (MRI). Because low-grade infection often evolves in chronic arthritis with joint destruction, it is very important to pursue atypical germs such as tuberculosis, fungi, *T. whipplei*, and *Borrelia burgdorferi*. Moreover, some of the more common bacteria can be responsible of low-grade infection in some rheumatic patients because of immunosuppression. In all these situations, the biopsy allows a quick bacteriological examination with Gram staining, then later culture and PCR analysis for atypical organism. Indeed, 2 patients were diagnosed with Lyme and articular Whipple disease by PCR analysis. Interestingly, the Whipple PCR that was performed on the SF collected during procedure was negative. There is 1 previously reported similar case in which SF PCR failed to demonstrate the presence of *T. whipplei*, but the synovial tissue PCR was positive[31].

Bacterial culture in both SF and synovial membrane is a key method for septic arthritis diagnosis. However, using those methods, infectious agents were isolated in only 41.2% of the patients (38.7% of SF and 23.5% of synovial membrane positive cultures)[32]. Therefore, histological synovial cell infiltrate analysis is also relevant for septic arthritis assessment. A neutrophilic cellular infiltrate has been shown to be highly associated with septic arthritis[33]. That presence inside the synovial tissue is considered sufficient for the diagnosis of septic arthritis. Regarding our data, the diagnosis of septic arthritis was established following the histological examination of 2 patients. Interestingly, after empiric antimicrobial therapy began in these 2 patients, no relapse occurred within at least 6 years of followup for both. Our analysis can also be useful in fibrocartilaginous joints (acromioclavicular, pubic symphysis), in which fluid is rarely found even in cases of inflammation. Further, we can conclude from our data that no patient of our cohort has been further diagnosed with infectious arthritis. This technique can, therefore, be considered reliable to rule out septic arthritis assessment, thus permitting local treatments such as steroid injections.

More rarely, synovial biopsy can be performed for synovial tumor assessment, especially villonodular synovitis or osteochondromatosis. The 2 patients in our cohort diagnosed with villonodular synovitis underwent surgical synovectomy. The histological examination of the tissue confirmed those findings.

For the biopsy to be useful in clinical practice, the quality

of the biopsies retrieved has to be good. Quality of a synovial biopsy has been defined for research recently[23]; however, no definition has yet been given for the clinical setting. In our study, we defined good quality as the following: sufficient size defined by synovial tissue area > 0.5 mm^2, preserved tissue allowing assessment by pathologists, and presence of lining layer. In our cohort, the quality was good enough to allow a histological examination in all biopsies retrieving synovial tissue. Lining layer was found in 92.2% of the cases. In some instances, the lining layer could be identified but was not connected to the main biopsy. This separation may have occurred during tissue processing or may represent separation due to fibrin deposition in case of ulcerative synovitis.

No study has thus far demonstrated a predictive clinical value for histological findings in identifying those with early arthritis or those who will go on to have an aggressive disease course[6,9,10]. Indeed, multiple studies tried to match histological cell infiltrate patterns with different rheumatologic conditions. There are undeniable differences between rheumatoid arthritis (RA) and psoriatic arthritis (PsA)[34,35], RA and ankylosing spondylitis (AS)[36], and RA and osteoarthritis (OA)[37,38]. OA synovial membrane is known to show less inflammatory infiltrate and less vascularity than its inflammatory counterparts in RA, PsA, and AS. RA synovium has shown a higher number of B cells and more rarely ectopic follicles, helping in the diagnosis. The high-grade synovitis features are more consistent with RA[39]. However, despite those differences, no algorithm is able to predict the evolution in early arthritis[33].

Given this, the histopathologist was rarely able to determine the type of inflammatory arthritis. However, by ruling out or confirming infectious arthritis or synovial tumor, it is clear that US-guided synovial biopsy is helpful for patients with remaining unknown diagnosis despite SF analysis, radiograph, CT scan, and/or MRI examinations. In our setting, synovial biopsies allowed the treatment of some patients by providing a definite diagnosis. They also indicated the use of systemic immunosuppressive or local therapies such as intraarticular steroid injections.

We acknowledge that our work has limitations, such as the monocentric design of our study. The biopsies were performed by a trained investigator, and the pathologists in our center had expertise in biopsy assessment. This could be a limit for the generalization of those results. Although all patients had 3 to 8 biopsies taken, 55% of them had a single fragment sent to the pathology department. This might be another limitation.

Finally, one of the main concerns about any procedure is its tolerance. In our cohort, 1 patient treated with salicylic acid presented with knee hemarthrosis 48 h after the procedure. Overall, in our cohort, the adverse effects rate was 1.35% (95% CI –1.3 to 4.0, 1/74) and no severe adverse event occurred (life-threatening, leading to patient admission to hospital, or with a risk of sequelae). The arthroscopic biopsies have the advantage to be retrieved under direct vision and therefore allow a histological analysis of the inflamed areas within the joint. However, this procedure is more invasive and has multiple adverse effects (joint infection, wound infection, hemarthrosis, deep venous thrombosis, neurological damage, thrombophlebitis)[40].

Our study highlights the potential diagnostic involvement of synovial biopsy. To our knowledge, ours is the first study describing indications, tolerability, rate of success, diagnosis involvement, and quality of US-guided synovial biopsy in a clinical setting. US-guided synovial biopsy was performed in clinical practice in a heterogeneous population with variant clinical features. The success rate of the procedure remains high, with only rare and minor complications; 13.3% achieved a definitive diagnosis leading to a specific treatment. In other patients, we could rule out the diagnosis of septic arthritis. Therefore, this procedure should be used not only for research purposes but also routinely in undifferentiated arthritis.

REFERENCES

1. Man GS, Mologhianu G. Osteoarthritis pathogenesis - a complex process that involves the entire joint. J Med Life 2014;7:37-41.
2. McInnes IB, Schett G. The pathogenesis of rheumatoid arthritis. N Engl J Med 2011;365:2205-19.
3. Pitzalis C, Kelly S, Humby F. New learnings on the pathophysiology of RA from synovial biopsies. Curr Opin Rheumatol 2013;25:334-44.
4. Rooney M, Whelan A, Feighery C, Bresnihan B. Changes in lymphocyte infiltration of the synovial membrane and the clinical course of rheumatoid arthritis. Arthritis Rheum 1989;32:361-9.
5. Bresnihan B, Tak PP. Synovial tissue analysis in rheumatoid arthritis. Baillieres Best Pract Res Clin Rheumatol 1999;13:645-59.
6. Tak PP. Analysis of synovial biopsy samples: opportunities and challenges. Ann Rheum Dis 2000;59:929-30.
7. Tak PP. Lessons learnt from the synovial tissue response to anti-rheumatic treatment. Rheumatology 2000;39:817-20.
8. Tak PP, van der Lubbe PA, Cauli A, Daha MR, Smeets TJ, Kluin PM, et al. Reduction of synovial inflammation after anti-CD4 monoclonal antibody treatment in early rheumatoid arthritis. Arthritis Rheum 1995;38:1457-65.
9. Bresnihan B. Are synovial biopsies of diagnostic value? Arthritis Res Ther 2003;5:271-8.
10. Gerlag DM, Tak PP. How to perform and analyse synovial biopsies. Best Pract Res Clin Rheumatol 2013;27:195-207.
11. van der Heijden IM, Wilbrink B, Vije AE, Schouls LM, Breedveld FC, Tak PP. Detection of bacterial DNA in serial synovial samples obtained during antibiotic treatment from patients with septic arthritis. Arthritis Rheum 1999;42:2198-203.
12. Graf SW, Buchbinder R, Zochling J, Whittle SL. The accuracy of methods for urate crystal detection in synovial fluid and the effect of sample handling: a systematic review. Clin Rheumatol 2013; 32:225-32.
13. Forestier J. Instrumentation pour biopsie médicale. C R Séances Soc Biol Fil 1932;110:186.
14. Polley HF, Bickel WH. Punch biopsy of synovial membrane. Ann Rheum Dis 1951;10:277-87.
15. Parker RH, Pearson CM. A simplified synovial biopsy needle. Arthritis Rheum 1963;6:172-6.
16. Beaulé V, Larédo JD, Cywiner C, Bard M, Tubiana JM. Synovial membrane: percutaneous biopsy. Radiology 1990;177:581-5.

17. Parlier-Cuau V, Hamzé B, Bellaïche L, Wybier M, Laredo JD. [Percutaneous biopsy of synovium: techniques]. [Article in French] Feuillet de Radiologie 1999;39:225-30.

18. Altman RD, Gray R. Diagnostic and therapeutic uses of the arthroscope in rheumatoid arthritis and osteoarthritis. Am J Med 1983;75:50-5.

19. Youssef PP, Smeets TJ, Bresnihan B, Cunnane G, Fitzgerald O, Breedveld F, et al. Microscopic measurement of cellular infiltration in the rheumatoid arthritis synovial membrane: a comparison of semiquantitative and quantitative analysis. Br J Rheumatol 1998;37:1003-7.

20. Jacob D, Cyteval C, Moinard M. [Interventional sonography]. [Article in French] J Radiol 2005;86:1911-23.

21. Cardinal E, Chhem RK, Beauregard CG. Ultrasound-guided interventional procedures in the musculoskeletal system. Radiol Clin North Am 1998;36:597-604.

22. Lazarou I, D'Agostino MA, Naredo E, Humby F, Filer A, Kelly SG. Ultrasound-guided synovial biopsy: a systematic review according to the OMERACT filter and recommendations for minimal reporting standards in clinical studies. Rheumatology 2015;54:1867-75.

23. Kelly S, Humby F, Filer A, Ng N, Di Cicco M, Hands RE, et al. Ultrasound-guided synovial biopsy: a safe, well-tolerated and reliable technique for obtaining high-quality synovial tissue from both large and small joints in early arthritis patients. Ann Rheum Dis 2015;74:611-7.

24. Marin F, Lasbleiz J, Albert JD, Askri A, Werner-Leyval S, Duval H, et al. [Synovial biopsy under US guidance: technical considerations and results]. [Article in French] J Radiol 2006;87:561-5.

25. van Vugt RM, van Dalen A, Bijlsma JW. Ultrasound guided synovial biopsy of the wrist. Scand J Rheumatol 1997;26:212-4.

26. Bresnihan B, Cunnane G, Youssef P, Yanni G, Fitzgerald O, Mulherin D. Microscopic measurement of synovial membrane inflammation in rheumatoid arthritis: proposals for the evaluation of tissue samples by quantitative analysis. Br J Rheumatol 1998;37:636-42.

27. Koski JM, Helle M. Ultrasound guided synovial biopsy using portal and forceps. Ann Rheum Dis 2005;64:926-9.

28. Scirè CA, Epis O, Codullo V, Humby F, Morbini P, Manzo A, et al. Immunohistological assessment of the synovial tissue in small joints in rheumatoid arthritis: validation of a minimally invasive ultrasound-guided synovial biopsy procedure. Arthritis Res Ther 2007;9:R101.

patients with rheumatic diseases—when and how we do it? Eur J Radiol 2011;79:407-14.

30. van de Sande MG, Gerlag DM, Lodde BM, van Baarsen LG, Alivernini S, Codullo V, et al. Evaluating antirheumatic treatments using synovial biopsy: a recommendation for standardisation to be used in clinical trials. Ann Rheum Dis 2011;70:423-7.

31. O'Duffy JD, Griffing WL, Li CY, Abdelmalek MF, Persing DH. Whipple's arthritis: direct detection of Tropheryma whippelii in synovial fluid and tissue. Arthritis Rheum 1999;42:812-7.

32. Madruga Dias J, Costa MM, Pereira da Silva JA, Viana de Queiroz M. Septic arthritis: patients with or without isolated infectious agents have similar characteristics. Infection 2014;42:385-91.

33. Della Beffa C, Slansky E, Pommerenke C, Klawonn F, Li J, Dai L, et al. The relative composition of the inflammatory infiltrate as an additional tool for synovial tissue classification. PLoS ONE 2013;8:e72494.

34. Kruithof E, Baeten D, De Rycke L, Vandooren B, Foell D, Roth J, et al. Synovial histopathology of psoriatic arthritis, both oligo- and polyarticular, resembles spondyloarthropathy more than it does rheumatoid arthritis. Arthritis Res Ther 2005;7:R569-80.

35. van Kuijk AW, Tak PP. Synovitis in psoriatic arthritis: immunohistochemistry, comparisons with rheumatoid arthritis, and effects of therapy. Curr Rheumatol Rep 2011;13:353-9.

36. Kidd BL, Moore K, Walters MT, Smith JL, Cawley MI. Immunohistological features of synovitis in ankylosing spondylitis: a comparison with rheumatoid arthritis. Ann Rheum Dis 1989;48:92-8.

37. Pessler F, Dai L, Diaz-Torne C, Gomez-Vaquero C, Paessler ME, Zheng DH, et al. The synovitis of "non-inflammatory" orthopaedic arthropathies: a quantitative histological and immunohistochemical analysis. Ann Rheum Dis 2008;67:1184-7.

38. Baeten D, Demetter P, Cuvelier C, Van Den Bosch F, Kruithof E, Van Damme N, et al. Comparative study of the synovial histology in rheumatoid arthritis, spondyloarthropathy, and osteoarthritis: influence of disease duration and activity. Ann Rheum Dis 2000;59:945-53.

39. Krenn V, Morawietz L, Burmester GR, Kinne RW, Mueller-Ladner U, Muller B, et al. Synovitis score: discrimination between chronic low-grade and high-grade synovitis. Histopathology 2006; 49:358-64.

40. Kane D, Veale DJ, FitzGerald O, Reece R. Survey of arthroscopy performed by rheumatologists. Rheumatology 2002;41:210-5.

VIII. SERMENT D'HIPPOCRATE

Au moment d’être admise à exercer la médecine, je promets et je jure d’être fidèle aux lois de l’honneur et de la probité.
Mon premier souci sera de rétablir, de préserver ou de promouvoir la santé dans tous ses éléments, physiques et mentaux, individuels et sociaux.
Je respecterai toutes les personnes, leur autonomie et leur volonté, sans aucune discrimination selon leur état ou leurs convictions. J’interviendrai pour les protéger si elles sont affaiblies, vulnérables ou menacées dans leur intégrité ou leur dignité. Même sous la contrainte, je ne ferai pas usage de mes connaissances contre les lois de l’humanité.
J’informerai les patients des décisions envisagées, de leurs raisons et de leurs conséquences.
Je ne tromperai jamais leur confiance et n’exploiterai pas le pouvoir hérité des circonstances pour forcer les consciences.
Je donnerai mes soins à l’indigent et à quiconque me les demandera. Je ne me laisserai pas influencer par la soif du gain ou la recherche de la gloire.
Admise dans l’intimité des personnes, je tairai les secrets qui me seront confiés. Reçue à l’intérieur des maisons, je respecterai les secrets des foyers et ma conduite ne servira pas à corrompre les mœurs.
Je ferai tout pour soulager les souffrances. Je ne prolongerai pas abusivement les agonies. Je ne provoquerai jamais la mort délibérément.
Je préserverai l’indépendance nécessaire à l’accomplissement de ma mission. Je n’entreprendrai rien qui dépasse mes compétences. Je les entretiendrai et les perfectionnerai pour assurer au mieux les services qui me seront demandés.
J’apporterai mon aide à mes confrères ainsi qu’à leurs familles dans l’adversité.
Que les hommes et mes confrères m’accordent leur estime si je suis fidèle à mes promesses ; que je sois déshonorée et méprisée si j’y manque.

NOM : NAJM Prénom : Aurélie

Titre de la thèse : **Qualité et utilité des biopsies synoviales échoguidées réalisées en pratique courante.**

RESUME

Objectifs : L'étude histologique de la membrane synoviale est utile non seulement en physiopathologie mais aussi pour l'évaluation de la réponse thérapeutique dans les rhumatismes inflammatoires chroniques. L'échographie est un outil largement utilisé qui permet un guidage précis lors des procédures de biopsies notamment de la membrane synoviale. Les objectifs de notre étude étaient : de déterminer le taux de succès de la technique de biopsie synoviale échoguidée, de décrire les indications de ces biopsies synoviales en pratique courante, de déterminer les situations cliniques dans lesquelles la réalisation de biopsies synoviales échoguidée était la plus rentable, et enfin de déterminer la qualité de tissu synovial obtenu à l'aide de cette technique.

Méthodes : Des biopsies synoviales échoguidées ont été réalisées au sein du service de Rhumatologie du CHU de Nantes de février 2007 à Décembre 2014 en utilisant un trocard à biopsie de type guillotine semi-automatique avec coaxiale. La procédure était considérée comme réussie lorsque du tissu synovial était visualisé à l'examen anatomopathologique.

Résultats : Soixante-seize procédures concernant soixante-quatorze patients présentant un tableau de mono ou polyarthrite sans étiologie retrouvée malgré les examens complémentaires ont été analysées.

Le taux de succès de la procédure de biopsie synoviale échoguidée était de 81.6% (62/76). En terme de tolérance, un patient traité par acide acétyl-salicylique a présenté une hémarthrose 48 heures après la procédure, résolutive après ponction évacuatrice. La procédure de biopsie synoviale échoguidée a permis d'obtenir un diagnostic définitif chez 16% des patients.

Conclusion : La procédure de biopsie synoviale échoguidée est globalement sûre et bien tolérée en pratique courante, avec un taux de succès élevé. Dans la grande majorité des cas, le tissu synovial prélevé est de qualité satisfaisante, permettant une analyse histopathologique. Chez une minorité significative de patients porteurs d'une mono ou polyarthrite d'étiologie inconnue malgré les examens complémentaires, la biopsie synoviale permet un diagnostic de certitude conduisant à un traitement adapté.

MOTS-CLES

Biopsie synoviale, Membrane synoviale, Synovite, Echographie, Echoguidage, Diagnostic, Arthrite indifférenciée.