

Tracking of Fascicles of Sartorius and Pectineus Nerves-A Key to Neurosurgery

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ABSTRACT

Introduction: Sartorius and pectineus are muscles of thigh innervated by Nerve to Sartorius (NS) and Nerve to Pectineus (NP) respectively. If pectineus and sartorius muscles are paralysed or weakened due to injury to NP and NS or their fascicles, these injured nerves and/or fascicles are to be repaired or stimulated to restore the functions of these muscles.

Aim: An experimental histological study of the NP and NS was designed to explore the configuration of fascicles and their identification for repair at fascicular level. This microscopic work has been done at such a close sample interval (1-2 mm) for the first time.

Materials and Methods: The slides from 24 blocks of 4/5 mm tissue of femoral nerve were observed under microscope and photographs were taken. The fascicles were identified at emergence of NS and NP and mapped/tracked cranially in presence of transformational processes in their pathways.

The group of fascicles contributing to these nerves were also mapped cranially up to the top of cropped femoral nerve.

Results: Fascicles 10, 11 and 12 were main contributors to nerve to sartorius. These fascicles underwent 49 transformational processes transforming into fascicles 298, 299, 307, 308 and 309 forming NS in S1 1. Similarly the fascicles 7, 8 and 9 constituted NP having undergone 33 transformational processes forming fascicles 220, 305 and 306 in A24 1 matching with the fascicles of P1 1. The clear picture of pathways of above mentioned fascicles from point of emergence of NS and NP upto top of femoral nerve and vice versa is presented in table Models NP and NS.

Conclusion: The pictures of composite fascicles will revolutionise not only diagnosis and treatment of injured fascicles but also interpretation of MRI along with the identification. The study will add new dimension to future diagnosis and treatment of injured nerve or fascicle.

Keywords: Fascicular mapping, Identification of fascicle, Nerve to pectineus, Nerve repair, Nerve to sartorius, Nerve stimulation

INTRODUCTION

Sartorius muscle is the muscle of anterior compartment of thigh. It is innervated by Nerve to Sartorius (NS), a branch of femoral nerve. Normally, NS originates from anterior division of femoral nerve. The pectineus muscle is the muscle of adductor compartment of thigh. Nerve to Pectineus (NP) innervates pectineus muscle [1].

Sartorius is responsible for abduction and outward rotation of the hip joint, flexion and inward rotation of knee joint. Overstraining of sartorius muscle as in jogging and swimming (breast stroking) may cause pes anserine bursitis. Pes anserine bursitis is most common in young individuals involved in sporting activities and in obese, middle-aged women [2,3]. Main function of pectineus is adduction of thigh and involved in groin strain. Groin strain often occurs in sports that require pivoting and quick changes of direction, such as soccer and hockey. The injury may range from mild pain in the adductor muscles to severe pain, tenderness, and inflammation in this area [1].

So, in case of injury to NS and NP or their Composite Fascicles (CFs), functions of sartorius and pectineus muscles are impaired. The information pertaining to impairment of functions of sartorius and pectineus muscles on account of injury to NS and NP and fascicles therein or in their extension in femoral nerve, are transmitted to and from the brain by these NS and NP through their fascicles. So, to restore the impairment by neural intervention at fascicular level [4], the configuration of pathways of CFs and distribution pattern of nerve fibers for innervations are necessary to be investigated for location, identification and longitudinal extension both ways, cranially and caudally in femoral nerve. The identification of pathways, configuration of CFs is usually done by high resolution MRI but it has limitations of resolution (accuracy of sampling interval) in recording and interpretation. This has inspired the author to carry

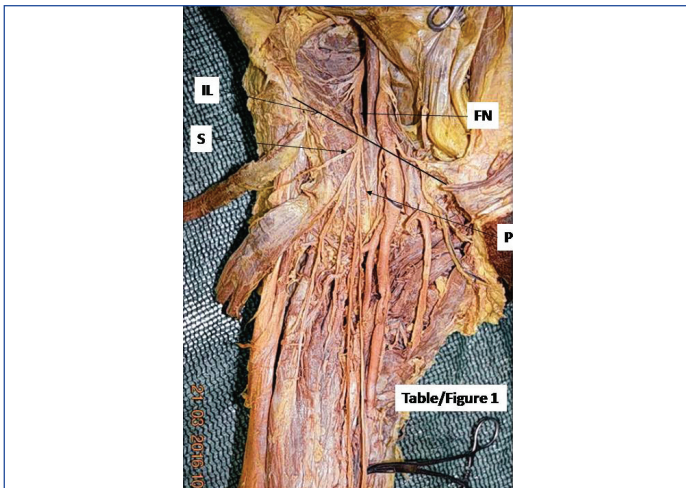
out a histological experiment to overcome these limitations of MRI. Scanty literature is available regarding intraneural, internal or external morphological set up of CFs in the femoral in general and its branches NS and NP in particular. So NS and NP have been taken up to unfold identification, configuration and extension of pathways of CFs and distribution of nerve fibers in femoral nerve up to emergence of these nerves.

Therefore, a micro-anatomical study has been designed for revealing fascicular configuration, distribution and exploration of sharing of nerve fibres through tracking, the longitudinal pathways of CFs in femoral nerve terminating into emergence of sartorius and pectineus nerve.

MATERIALS AND METHODS

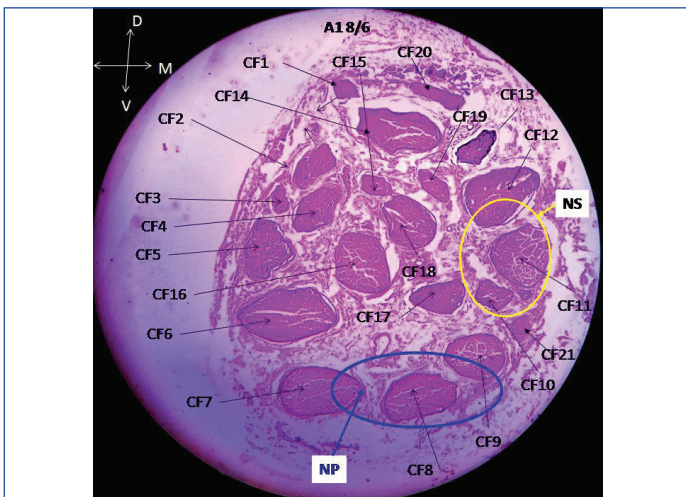
The present study was a histological or microanatomical study which was conducted as histological workup from October 2017 to January 2018. A right femoral nerve [Table/Fig-1] was harvested from a 70-year-old female cadaver in the dissection hall of Anatomy department of AIIMS, Rishikesh, Uttaranchal, India. In our case NS emerged from lateral side of femoral trunk below the inguinal ligament and NP from the medial side of trunk of femoral nerve as usual. As transformational processes consisting of split, fusion and multiplexing, due to more transient dynamics of morphological changes in fascicular pathways and their organisational setup, are more frequent in this part [5,6] so right from upper most end of the femoral nerve to the emergence of NS and NP was considered for preparing closely sampled histological slides at 1 to 2 mm sample interval to record almost all the transformational processes. The measurements of the entire length of the femoral nerve were taken from inguinal ligament, a discernible landmark, in this region and blocks of 0.4 to 0.5 cm lengths were prepared.

The cropped femoral nerve measuring 1.0 cm below and 8.9 cm above, so net 9.9 cm of femoral nerve, keeping inguinal ligament



[Table/Fig-1]: In situ image of cropped femoral nerve with identification of nerve to sartorius and pectineus; FN=femoral nerve; P=nerve to pectineus; S=nerve to Sartorius; IL= Inguinal ligament

as origin (0.0 cm) was cut into 24 pieces, sequentially from proximal to distal end of femoral nerve. So, all the 24 blocks, (A1-A24) were processed to prepare 13 to 19 slides in 3 to 4 sets from each block. Typically, each block was trimmed 1 mm from the bottom by microtome. Then first set of 3/7 slides was processed. Similarly, second and third sets of 3/7 slides was prepared after trimming 1 mm each from the bottom of the block after each set in continuation. All the slides were stained with haematoxylin and eosin. This process was repeated for all the 24 blocks. Besides these 24 blocks, we have also prepared two more blocks (S1 and P1), one each from NS and NP at their emergence. After processing and staining by haematoxylin and eosin, these slides were photographed by 16 mega pixel Sony camera under a high resolution microscope of model PZRM-26 having software "Future Winjoe". After this, the CFs were named from CFs 1 to 21 [Table/Fig-2] in the cranial most slide A1 8 correlated down to A24 1 from proximal to distal.



[Table/Fig-2]: Slide A1 8: CF=composite fascicle; CFs named from CF 1 to CF 21, NS=nerve to sartorius enveloped by yellow colour; NP=nerve to pectineus enveloped by blue colour; D=dorsal, V=ventral, M=medial

Correlation and tracking of CFs: All the CFs participating in formation of NS and NP in slides from A1 8 through A24 1 were tracked, correlated and interpreted for identification, shape, size, continuity, consistency and transformational processes of CFs [Table/Fig-3,4] in femoral nerve and in first block of NS and NP up to slides S1 1 and P1 1 at their emergence. The methodology adopted for correlation was placement of consecutive slides side by side caudally and cranially and compared all the fascicles in both sections on the basis of correlation parameters. The correlation parameters used were location (Dorsal, Ventral, Lateral and Medial; Central or peripheral), shapes (circular, oval and irregular), sizes (large, medium and small), orientation, split, fusion, migration and multiplexing.

Lateral and medial sides and dorsal and ventral faces were marked by tissue marker with colour code for lateral, medial, dorsal and ventral sides for our reference. As soon as intra-block correlation was over; the similar methodology was adopted for inter-block (A1 through A24) correlation through sequential slides. As long as the CF was found consistent and continuous during correlation in different sections in (proximal to distal) cranial to caudal direction, the same number was retained but when any CF underwent transformational processes [Table/Fig-5a,b; 6a,b; 7a,b], after crossing over the point of inflexion, a sequential new unique identity numbers were given to new CFs so formed.

Thus, all the sections were correlated and pathways of CFs were identified from top slide to the emergence of NS and NP and then the correlation of CFs in slides of branch nerves NS and NP was also done in slides S1 1 and P1 1. The correlation of fascicles was also done beginning from 1st slide S1 and P1 of NS and NP through A24 block up to the top of femoral nerve from distal (A24 block) to proximal (A1 block). The position of transformational processes including continuity and consistency were determined in the slides from inguinal ligament as all the sections were calibrated in distance from inguinal ligament.

Modelling of fascicular pathways: The data in tables present models of pathways of CFs of NS and NP. The 3D model of pathways of these contributing fascicles would have been too complex to be drawn therefore; the same CF's data would be stored in tables to provide a sense of continuous and consistent pathways of CFs interrupted by transformational processes. The intervals of consistent pathways of these CFs contributing to NS and NP in each slide would be shown. The group of CFs constituting NS and NP would also be tracked in each slide. All these data set would be shown in tables.

RESULTS

Based on physical examination, NS and NP emerged out from lateral and medial side of femoral nerve at distances of 0.5 and 0.9 cm below the inguinal ligament respectively [Table/Fig-1]. The tracking, correlation and interpretation of CFs revealed the formation of NS and NP after A23 1 [Table/Fig-8] and A24 1 [Table/Fig-9], their point of emanation was situated at 0.49975 cm and 0.89975 cm respectively below inguinal ligament. These distances established a good matching between physical measurements and computed by slide planning and preparation.

A huge data volume was generated out of the experimental research work consisting of 24 blocks from femoral nerve and two blocks, one each from NS and NP, each of length 4/5 mm. All the 26 blocks were processed for 344 slides consisting of images of 5400 CFs. These CFs were consolidated into 318 through tracking, correlating and interpreting all the 21 CFs in A1 8 passing through 209 transformational processes. The CFs, 10, 11, 12 and 7, 8, 9 fully or partially contributing to NS and NP underwent 82 transformational processes transforming into CFs, 298, 299, 307, 308, 309 forming NS at emergence in A23 1 and 220, 305, 306 forming NP at emergence in A24 1.

Correlation results: The tracking, correlation and interpretation of CFs in histological slides from A1 8 in which the CFs were named (CF1-21) to A24 1 (the point of emergence of NP) portrayed good matching of CFs in the same set of slides (such as A2 19-A2 16 etc.), barring few exceptions (A8 2-3). The correlation between two sets of slides too was found satisfactory except when it is disturbed by multiplexing. The inter-block (Block A1 1, A2 19 etc., here A1 1 is the caudal most slide of block A1 and A2 19 is the cranial most slide of block A2) tracking and correlation was also found matching with good level of confidence. Thus, the correlation of CFs was extended in all the blocks and thereby in all slides. This tracking brought out continuity, consistency and correlativity of CFs interrupted by transient dynamic processes

Level of slides (cm)		Involved sections	Participating CF's	T Process	New CF's
8.60275	8.50225	A1 6-A1 5	10+11, 12*	F	23, 22*
8.20475	8.10425	A2 10-A2 9	23	S	23.1; 23.2
8.10275	8.00225	A2 6-A2 5	23.1	S	29; 30
8.00025	7.80725	A2 1-A3 15	29, 23.2	F	31
7.60025	7.40675	A3 1-A4 14	22*, 30	F	39*
7.30275	7.20225	A4 6-A4 5	39*, 31	F	45*
7.20025	7.00575	A4 1-A5 12	45*	S	45.1, 2*, 3
6.80025	6.60725	A5 1-A6 15	45.2*	S	58*+57
			45.3, 57	F	57'
6.40025	6.20525	A6 1-A7 11	58*	S	65, 66
5.80525	5.70475	A8 11-A8 10	66	S	66a, 66b
5.20025	5.00725	A9 1-A10 15	76d, 66b , 80	F	94*
			57', 27'	F	95*
5.00525	4.90475	A10 11-A10 10	94*	S	100*, 101*
			95*, 90	M	95a, 102*
4.90275	4.80225	A10 6-A10 5	102*	S	109, 110*
4.80025	4.60675	A10 1-A11 14	95a, 106, 110*	M	112, 113*
4.60475	4.50425	A11 10-A11 9	113*, 112	F	117*, 113a
4.50275	4.40225	A11 6-A11 5	99, 100*, 101*	F	118*
			66a, 117	M	119, 120, 121, 122
4.40025	4.20725	A11 1-A12 15	122*	SC	122a*
			118*	S	129*, 130, 131
4.20525	4.10475	A12 11-A12 10	129*	S	133, 134
4.10275	4.00225	A12 6-A12 5	119+134	F	138*
4.00025	3.80675	A12 1-A13 14	122a*	S	145*, 146
			133, 138*	F	142*
3.80475	3.70425	A13 10-A13 9	145*	S	150, 151
			142*	S	142'+142''
3.70275	3.60225	A13 6-A13 5	150+142' *	M	155+156+161
3.60025	3.40675	A13 1-A14 14	155, 156, 161	F	166*
3.40475	3.30425	A14 10-A14 9	166*	S	174, 175*
3.30275	3.20225	A14 6-A14 5	167, 175*	M	180, 181, 182
3.00475	2.90425	A15 10-A15 9	157, 181	F	190*
			174	SC	193
			180, 186	F	192*
2.90225	2.80175	A15 5-A15 4	193, 194, 195	F	196*
			191, 192*	F	197*
2.80025	2.60725	A15 1-A16 15	185, 190*, 197, 199	M	204*, 205*, 206
2.60525	2.50475	A16 11-10	204*	S	207*, 208
2.50275	2.40225	A16 6-A16 5	188, 207	M	210*, 211
			205*	S	215, 216*
2.40025	2.20675	A16 1-A17 14	196*	S	217, 218*
2.10275	2.00225	A17 6-A17 5	216*	S	219, 220
			218*	S	218a+218b
			210*, 212, 200	M	221, 222, 223, 224, 225
1.70275	1.60225	A18 6-A18 5	218a+CF215	F	242
1.30275	1.20225	A19 6-A19 5	224, 242	F	254
1.00525	0.90475	A20 11-A20 10	254	S	272, 273
0.40025	0.20825	A21 1-A22 17	272, 273, 219	M	289, 290
IL0.00025	-0.29275	A22 1-A23 15	290	S	298, 299
-0.39725	-0.49975	A23 6-A23 1	289	S	307, 308, 309
-0.49975		After A23 1	307, 308, 309, 298, 299	As NS	Emerged out

[Table/Fig-3]: Correlation of CFs participating in formation of nerve to sartorius. Level of slides=Represent the distance of slide from Inguinal ligament; involved sections=Represent the consecutive section between which transformational process is taking place; participating CFs in transformation=The CFs numbers contributing fully to Nerve to sartorius are made complete bold and those contributing partly are marked by star and made partly bold; T process=Transformational process; new CFs named after transformation; S=Split; F=Fusion; M=Multiplexing; SC=Shape change

Level of slides in mm		Involved sections	Participating CF's	TP	New CF's
		A1 8-A1 1	7*, 8, 9*		
8.50025	8.30925	A1 1-A2 19	8	S	8.1+8.2
8.30775	8.20725	A2 16-A2 15	7*+ 8.1	F	26
8.20475	8.10425	A2 10-A2 9	26	S	26a*+26b
			9*+ 8.2	F	27*
7.60025	7.40675	A3 1-A4 14	34*+ 26a*	F	40
7.20025	7.00575	A4 1-A5 12	40+27*	M	40*+27'
6.80025	6.60725	A5 1-A6 15	40*	S	52*+53
5.60225	5.60025	A8 5-A8 1	83+ 52*	F	86*
5.20025	5.00725	A9 1-A10 15	86*	S	92*+93
			47a+ 27'	F	95*
5.00525	4.90475	A10 11-A10 10	95*+90	M	95a+102*
4.90275	4.80225	A10 6-A10 5	92*	S	107+ 108
			102*	S	109+110*
4.80025	4.60675	A10 1-A11 14	95a+106+ 110*	M	112+113*
			108+109	F	109a
4.60475	4.50425	A11 10-A11 9	107+ 109a	F	116*
			113*	S	113a+113b*
			113b*+112	F	117
4.50275	4.40225	A11 6-A11 5	66a+117	M	119+120+121*+122*
			116*+ 113a	M	123*+124
4.40025	4.20725	A11 1-A12 15	123*+124+121*	F	127*
			122*	SC	122a*
4.00025	3.80675	A12 1-A13 14	122a*	S	145*+146
		A13 10-A13 9	145*	S	150+151
3.70275	3.60225	A 13 6- A13 5	127*	S	158*+159+160
3.60025	3.40675	A13 1-A14 14	158*	S	168+169
			146+160	F	157
3.20025	3.00625	A14 1-A15 13	169+182+151	M	185*+186
3.00475	2.90425	A15 10-A15 9	157+181	F	190
2.80025	2.60725	A15 1-A16 15	185*+190,	M	205*+206
2.50275	2.40225	A16 6-A16 5	205*	S	215+216*
2.10275	2.00225	A17 6-A17 5	206	S	213+214
			216*	S	219+220
1.70275	1.60225	A18 6-A18 5	213+214	F	238
-0.29475	-0.39525	A23 11-A23 10	238	S	305+306
-0.89975	-0.89975	After A24 1	220+305+306	NP	Emerged out

[Table/Fig-4]: Correlation of CFs participating in formation of Nerve to Pectineus. Level of slides=Represent the distance of slide from Inguinal ligament; involved sections=Represent the consecutive section between which transformational process is taking place; participating CFs in transformation=The CFs numbers contributing fully to Nerve to pectineus are made complete bold and those contributing partly are marked by star and made partly bold.; T process=Transformational process; new CFs named after transformation; S=Split; F=Fusion; M=Multiplexing; SC=Shape change

and changes in shapes, sizes and orientations causing plexiforms [6]. The CFs correlation data pertaining to exclusively NS and NP formation in their entire pathways were presented in [Table/Fig-3,4]. Besides, the mapping of NS and NP through correlation of group of CFs contributing to NS and NP, was also carried out in all the slides [Table/Fig-10,11]. The positions of transformational processes with reference to inguinal ligament of all these histological sections were also labelled and recorded in these tables. Normally the changes in these processes were regular but in a few cases, the transformational processes occurred instantly as observed in the consecutive slides A8 2-3, at the interval of 5 micron. The consolidation of correlation, tracking and interpretation of the occurrences of these dynamic changes brought out two tabular models both for NS and NP as illustrated below to comprehend the pathways of CFs contributing to NS and NP.

Table model NS: The correlation of CFs (1-21) from top most slide A1 8 to A23 1 revealed that the CF 10, 11 and 12 located in the medial

side at periphery of femoral nerve in A1 8, were the main **{CF 10, 11 (fully) and 12 (partly)}** contributors to NS. These CFs underwent 49 transformational process (21 splits, 19 fusions and 9 multiplexings) transforming into **CFs 298, 299, 307, 308 and 309** [Table/Fig-3] forming NS ready to emerge in section, A23 1 [Table/Fig-8]. These CFs present in A23 1 were matching with those present in the slides S1 1 prepared from first block of NS. These CFs were found absent in A24 15. The continuity, consistency and correlativity of the group of CFs contributing to NS in each slide and the positions of group of CFs were also shown in [Table/Fig-3]. Furthermore, widespread individual CFs less affected by transformational processes was presented [Table/Fig-12]. For example-CF23 remained continuous, consistent and extendable between the slides A1 5-A2 9 for 3 mm. These tables together draw clear picture of pathways of CFs contributing to NS.

Table model NP: The main contributors of NP, CFs 7, 8 and 9 out of 21 CFs in cranial most slide A1 8 were located in the ventral periphery of femoral nerve. The correlation of these CFs down to A24 1 and

Slides	Group of CFs in NS	CCC between (cm)	CCC Interval (cm)
A1 8-6	10+11+12*	8.60375-8.60275	0.001
A1 5/1-A2 10	22*+23	8.50225-8.20475	0.2975
A2 9/6	23.1+23.2+22*	8.10425-8.10275	0.0015
A2 5/1	23.2+30+29+22	8.00225-8.00025	0.002
A3 15/1	31+30+22	7.80725-7.60025	0.207
A4 14/6	31+39	7.40675-7.30275	0.104
A4 5/1	45	7.20225-7.20025	0.002
A5 12/1	45.2+45.3	7.00575-6.80025	0.2055
A6 15/12	57+45.3+58	6.60725-6.60575	0.0015
A6 11/1	57'+58	6.50525-6.40025	0.105
A7 11-A8 11	66+57'	6.20525-5.80525	0.4
A8 10-A9 1	66b+66a+57'	5.70475-5.20025	0.5045
A10 15/11	94*+95*+66a	5.00725-5.00525	0.002
A10 10/6	101*+100*+95a+66a+102	4.90475-4.90275	0.002
A10 5/1	101*+100*+95a+66a+110*	4.80225-4.80025	0.002
A11 14/10	101*+100*+113*+66a	4.60675-4.60475	0.002
A11 9/6	101*+100*+117*+66a	4.50425-4.50275	0.0015
A11 5/1	122+118+119	4.40225-4.40025	0.002
A12 15/11	122a+129+119	4.20725-4.20525	0.002
A12 10/6	122a*+133+119	4.10475-4.10275	0.002
A12 5/1	122a*+133+138	4.00225-4.00025	0.002
A13 14/10	145+142*	3.80675-3.80475	0.002
A13 9/6	150+142'	3.70425-3.70275	0.0015
A13 5/1	155+156+161	3.60225-3.60025	0.002
A14 14/10	166	3.40675-3.40475	0.002
A14 9/6	175*+174	3.30425-3.30275	0.0015
A14 5- A15 10	181+180+174	3.20225-3.00475	0.1975
A15 9/5	193+192*+190*	2.90425-2.90225	0.002
A15 4/1	196*+197*+190*	2.80175-2.80025	0.0015
A16 15/11	196*+204*+205*	2.60725-2.60525	0.002
A16 10/6	196*+207*+205*	2.50475-2.50275	0.002
A16 5/1	196*+210*+215+216*	2.40225-2.40025	0.002
A17 14/6	218*+215+216*+210*	2.20675-2.10275	0.104
A17 5/1-A18 6	219+215+218a+224	2.00225-1.70275	0.2995
A18 5/1-A19 6	219, 224, 242	1.60225-1.30275	0.2995
A19 5/1-A20 11	219, 254	1.20225-1.00525	0.197
A20 10-A21 1	219, 272, 273	0.90475-0.40025	0.5045
A22 17-A22 1	290, 289	0.20825-0.00025	0.208
A23 15/6	289, 298, 299	-0.29275+0.39725	0.1045
A23 5/1	298, 299, 307, 308, 309	-0.49775+0.49975	0.002
A23 1	298, 299, 307, 308, 309	-0.49975	NS emerged out

[Table/Fig-10]: Mapping of Nerve to sartorius in the slides from A1 8 through A23 1. This table shows correlation of group of fascicles constituting NS, their continuity, consistency and correlativity. Only those slides are given between which fascicles are correlatable, consistence and continuous. CCC between (cm): levels of slides between which the group of CFs are continuous, consistent and correlatable. CCC interval (cm): interval of continuity, consistency and correlativity of group of CFs. CCC=Continuous, Consistent and Correlatable

through enveloping NS by yellow curve and NP by blue curve in [Table/Fig-2].

2. Two typical examples of multiplexing between slides, A14 6 and A14 5 [Table/Fig-6a,b].

CF 164, 165 and 170 in A14 6 after multiplexing, reorganised into CF 176, 177, 178 and 179 in A14 5. CF 167 and 175 in A14 6 multiplexed transforming into CF 180, 181 and 182 in A14 5. The constituents of NS and NP as mapped are shown here in symbolic representation as-NS=CF 175*+174; NP=151+157+169 in A14 6 and NS=CF 181+180+174; NP=157+169+151 in A14 5.

Slides	Group of CFs in NP	C CC between (cm)	C CC Interval (cm)
A1 8-A1 1	7*+8+9*	8.60375- 8.50025	0.1035
A2 19/16	7*+8.1+9*+8.2	8.30925-8.30775	0.0015
A2 15/10	26*+9*+8.2	8.20725-8.20475	0.0025
A2 9-A3 1	26a*+27*	8.10425-7.60025	0.5040
A4 14/1	40*+27*	7.40675-7.20025	0.2065
A5 12/1	40*+27'	7.00575-6.80025	0.2055
A6 15-A8 3	52*+27'	6.60725-5.60125	1.006
A8 2-A9 1	86*+27'	5.60075-5.20025	0.4005
A10 15/11	92*+95*	5.00725-5.00525	0.002
A10 10/6	92*+102*	4.90475-4.90275	0.002
A10 5/1	108+109+110*	4.80225-4.80025	0.002
A11 14/10	109a+113**	4.60675-4.60475	0.002
A11 9/6	117+116*+113a	4.50425-4.50275	0.0015
A11 5/1	122+123*+124+121*	4.40225-4.40025	0.002
A12 15/1	122a+127*	4.20725-4.00025	0.207
A13 14/10	146+127*+145*	3.80675-3.80475	0.002
A13 9/6	146+127*+151	3.70425-3.70275	0.0015
A13 5/1	146+160+158*+151	3.60225-3.60025	0.002
A14 14/1	151+157+169	3.40675-3.20025	0.2065
A15 13/10	185*+157	3.00625-3.00475	0.0015
A15 9/1	190+185*	2.90425-2.80025	0.104
A16 15/6	205+206	2.60725-2.50275	0.1045
A16 5-A17 6	216*+213+214	2.40225-2.10275	0.2995
A17 5-A18 6	220+213+214	2.00225-1.70275	0.2995
A18 5-A23 15/11	220+238	1.60225-(-0.29275)	1.895
A23 10-A24 1	220, 305, 306	-0.39525-(-89975)	0.5045
After A24 1	220, 305, 306	NP emerged out	

[Table/Fig-11]: Mapping of nerve to pectineus in the slides from A1 8 through A24 1. This table shows correlation of group of fascicles constituting NP, their continuity, consistency and correlativity. Slides=Only those slides between which fascicles are correlatable, consistence and continuous. CCC between (cm): levels of slides between which the group of CFs are continuous, consistent and correlatable. CCC interval (cm): interval of continuity, consistency and correlativity of group of CFs. CCC=continuous, consistent and correlatable

Contributing CFs	Sections/slides	CCC Interval (mm)	Placement between (cm)
CF23	A1 5-A2 9	3.98	8.50225-8.10425
CF22	A1 5-A3 1	9.02	8.50225-7.60025
CF30	A2 5-A3 1	4.02	8.00225-7.60025
CF31	A3 15-A4 6	5.045	7.80725-7.30275
CF45.3	A5 12-A6 12	4.0	7.00575-6.60575
CF57/57'	A6 15-A9 1	14.07	6.60725-5.20025
CF66/66a	A711-A11 6	17.025	6.20525-4.50275
CF100+101	A10 10-A11 6	4.02	4.90475-4.50275
CF122/122a	A11 5-A12 1	4.02	4.40225-4.00025
CF196	A15 4-A16 1	4.015	2.80175-2.40025
CF215+216	A165-A17 6	3.045	2.40225-2.10275
CF219	A17 5-A21 1	16.02	2.00225-0.40025
CF219+272+273	A20 10-A21 1	5.045	0.90475-0.40025

[Table/Fig-12]: Computation of continuity, consistency and correlativity of selected CFs of nerve to sartorius. Contributing CFs are CFs contributing to NS which are more continuous, consistent and correlatable. Slides/sections=represents the slides between which the CF is continuous, consistent and correlatable. CCC interval (mm)=represents the interval in millimetres in which CFs are continuous, consistent and correlatable. Placement between: represents the calibrated labels in distance from inguinal ligament. The stimulating electrodes can be placed within this interval. CCC=continuous, consistent and correlatable

3. The typical examples of split, fusion and multiplexing together between slides A17 1-A18 15 [Table/Fig-7a,b]:

CF 209 in A17 5 split into CF 226 and 227 in A18 15. CF 223, 218b and 211 in A17 5 fused into CF 228 in A18 15. CF 221 and 222 in A17 5 multiplexed into CF 229, 230, 231, 232, 233 and 234 in A18 15. The mapped fascicles in NS and NP symbolically can be shown as: NS=219+215+218a+224; NP=220+213+214 in A17 1 and NS=219+215+218a+224; NP=220+213+214 in A18 15.

DISCUSSION

Though the mapping of fascicles has been done by some authors on femoral nerve [7], sciatic nerve [8] and on pudendal nerve [9]. But these authors tracked the fascicles at the interval of five mm missing the complete distribution of nerve fibers through transformational processes which are very important for suitable placement of nerve cuff electrodes unlike our study. Few authors have also carried out 3D peripheral nerve fascicle reconstruction [10].

The physical measurements of positions of various important points in correlation and tracking of straight and continuous pathways of CFs, interrupted by transformational processes and point of emanation of NS and NP [Table/Fig-3,4] of femoral nerve from 'Inguinal ligament', are highly useful in localising and identifying the CFs. The phenomenon of transformational processes alters the distribution pattern of nerve fibers, their grouping into CFs, their setup or organisation and their innervation pattern. The split of CFs in one slide subdivides the nerve fibers into two or more CFs in next slide and the fusion causes mixing of the fibers from two or more CFs in one slide into one CF in next slide. The multiplexing of two or more input CFs redistribute the nerve fibers into two or more output CFs. No nerve possesses purely sensory or motor nerve fibers [11]. These nerve fibers emanating from afferent and efferent nerve roots are initially grouped into motor and sensory fascicles but the naturally, developing system to form branch nerves containing appropriate blend of both types nerve fibers to facilitate emergence of branch nerves for appropriate innervation of targeted region, causes these transformational processes. As these processes of split, fusion and multiplexing cause the changes in shape, size, orientation and location of CFs due to distribution and intermixing of nerve fibers so identification, extension of pathways constituting configuration and organisation of CFs becomes complex. However, comprehending correlation parameters and using our own skill, experience and knowledge, the clear picture of fascicular pathways of CFs during formation of NS and NP till their emergence from femoral nerve, have been brought out.

NS Model

This model reveals the complete pathways of CFs 10, 11 and 12 contributing to NS from slide A1 8 cranially to CFs, 298, 299, 307, 308 and 309 in A23 1 caudally [Table/Fig-3]. After identification through slide correlation, the group of CFs constituting NS in each and every slide were sorted out in groups to track NS in these slides. The continuity, consistency and correlativity of groups of CFs in each slide forming NS are also displayed in this table [Table/Fig-10]. The specific CFs having more continuity, consistency and correlativity were also identified to see more straight paths [Table/Fig-12].

NP Model

The CFs 7, 8 and 9 in slide A1 8 have been found main contributors to NP formed of CFs 220, 305 and 306 at emergence after A24 1 [Table/Fig-4]. The model presents very clear picture of pathways of CFs from top of femoral nerve to the emanation of NP. Not only these CFs have been identified by names in the entire length of their pathways but also the location of transformational processes may be determined with the help of calibrated slides and calibrated distances. The NP was correlated in all the slides by identifying through assigned numbers of group of CFs contributing partially or fully to NP as shown in [Table/Fig-11]. The specific CFs having more continuity, consistency and correlativity were also shown in

[Table/Fig-13]. These tables not only provide very clear picture of CF's pathways but also determine the location of transformational processes interrupting their straight pathways.

Contributing CFs	Sections/slides	Interval (mm)	Placement between (cm)
CF9*	A1 8-A2 10	0.399	8.60375-8.20475
CF26/26a	A2 15-A3 1	0.607	8.20725-7.600250
CF40/40'	A4 14-A5 1	0.6065	7.40675-6.80025
CF27*	A2 9-A9 1	2.904	8.10425-5.20025
CF86*	A8 2-A9 1	0.4005	5.60075-5.20025
CF122**/122a**	A11 5-A12 1	0.402	4.40225-4.00025
CF127*	A12 15-A13 6	0.5045	4.20725-3.70275
CF213+214	A16 5-A18 6	0.6995	2.40225-1.70275
CF220+238	A18 5-A23 11	1.895	1.60225-(-0.29275)
CF220, 305, 306	A23 10-A24 1	0.5045	-0.39525-(-89975)

[Table/Fig-13]: Computation of continuity, consistency and correlativity of selected CFs of nerve to pectineus.

Contributing CFs is CFs contributing to NP which is more continuous, consistent and correlatable. Slides/sections=represents the slides between which the CF is continuous, consistent and correlatable. COC interval (mm)=represents the interval in millimetres in which CFs are continuous, consistent and correlatable. Placement between: represents the calibrated labels in distance from inguinal ligament. The stimulating electrodes can be placed within this interval. CCC=continuous, consistent and correlatable.

This experimental work produced two table models of NS and NP, would not only facilitate to smoothen the controversy of somatotopy and plexification [12] by identification, isolation and localisation of fascicle and its relationship with the specific nerve and region of innervations by these nerve fibers but also provide shape, size and directivity of fascicle for matching of nerve fibers for repair, grafting and regeneration [4]. Our models also brought out consistency and continuity of CFs in their longitudinal pathways which is never along the whole pathway of fascicle rather it is prevalent in between transformational processes. This establishes limited somatotopic structure of fascicles.

Clinical Significance

The nerves NP and NS and also their CFs can be injured anywhere in entire fascicular pathways causing partial/full paralysis of the sartorius and pectineus muscles impairing their functions. Therefore, the diagnosis of such neural insults requires not only the location and degree of injury but also identification, isolation, orientation, shape and size of injured CFs for planning surgical repair, grafting and regeneration. As the current experimental study brought out detailed microanatomy of NS and NP at the fascicular level consisting of internal morphology of CFs, fascicular pathways and detailed configuration so this data can be of immense use in diagnosis and neurosurgical manipulation.

The lack of thorough knowledge of innervation pattern illustrating the relationship of nerve to impaired muscle or problematic sensory area hinders the diagnosis. Here the injured elements are nerve, fascicles or nerve fibers. So, it is not only nerves but also fascicles which can suffer from injury anywhere in entire fascicular pathways from origin to point of innervation producing signs and symptoms. Thus, the investigation should not be focused on nerves only rather longitudinal pathways and configuration of fascicles should also be kept in mind. Now to investigate the degree and location of injury in nerve and CF, high resolution MRI advanced neurography [13,14] is used. But this has its own limitations of recording and interpretation and as such injured CFs neither can be identified nor related to innervations pattern of the ailed sensory area with more accuracy. This generates uncertainty in diagnosis and thereby in treatment. Thus the radiologists and neurosurgeons face the impediments of pinpointing the precise position of injury and its degree, the identification and localisation of injured CFs either for intraoperative stimulation to observe muscle response or to plan neuro-therapy or to plan neurosurgical manipulation.

However, our models of NS and NP from femoral nerve may reasonably overcome this impediment of identification of expected injured CFs as these models present the complete picture of CFs contributing to these nerves coupled with innervation pattern. Methodologically, it can be done by one to one correlation between images of histological transverse section and MRI transverse section at the same position from inguinal ligament. The distance of location of injured fascicle from inguinal ligament may be computed in MRI neurography and then the calibrated histological section in the evolved models of NS and NP at the same level is compared and examined for confirmation of identification of injured fascicle. This can further be reconfirmed by the analysis of the response of ailed organ by placing the electrodes at most suitable location for external or internal electrical stimulation of specific fascicle during intraoperative stimulation on the fascicles constituting NS and NP. The successful and non-invasive stimulation will depend on finding out most suitable location for placement of electrical electrode. The most suitable site of placement of electrode may be decided on the basis of continuous and consistent fascicles contributing fully to the nerve innervating the problematic area from our models. After identification of injured fascicle, the idea of shape, size, location and orientation can be obtained from our Table models NP and NS for matching, alignment and directivity of nerve fibers for repair, grafting and regeneration. Intraoperative nerve stimulation in the awake patient is a readily available tool that can aid in this goal by identifying the CF and suitable location through our model. Careful alignment is the critical factor in the first step. The nerve ends are inspected to identify fascicular groups matching proximally and distally amenable to individual fascicular repair [4]. These detailed procedures focus on longitudinal identification of fascicles, location of injury site and matching of fascicles for repair and grafting for administering the suitable surgical treatment. This is expected to improve failure rate with minimum invasion to other anatomical structures by iatrogenic procedures.

Our models alert the neurosurgeon to avoid intraoperative damage to fascicles during neurosurgical manipulation for repair and grafting to manage regeneration. Planning of neuro-surgical manipulations from MRI neurography in association with our models will definitely enhance success to failure ratio drastically.

LIMITATION

The distribution of nerve fibers after multiplexing is complex so identification of nerve fibers or fascicle is subjective but interpretational skill and experience together with the suitability of correlation parameters minimises the ambiguity. The study has been carried out on a single cadaver. More experiments will produce more data for expanding scope of results of this study for making it statistically significant. This will take into account the variations in nerve network along with other anatomical variations affecting the dimensions of nerves and corresponding variations in fascicular configuration.

CONCLUSION

The models NS and NP portrayed a clear and complete picture consisting of detailed configuration, pattern, organisation of fascicular pathways interrupted by splits, fusions and plexiforms, shapes and sizes of CFs. All the histological slides used in the

models were calibrated with distances from inguinal ligament for locating the injury on specific fascicles. The models of NS and NP can significantly improve the interpretation of injured CFs in MRI advanced neurography. The complete fascicular picture will facilitate its repair, grafting or regeneration together with detection of most suitable site for placement of electrical electrode for stimulation of specific fascicle. This will minimise the iatrogenic invasion along with reducing the failure rate. This may also help in advancing the technology in developing the instrument or equipment, CF electrode like nerve cuff electrode to revolutionise the treatment.

RECOMMENDATIONS

1. More experiments are to be carried out on femoral nerves in general and NS and NP in particular to accommodate variations and branching pattern through fascicular mapping.
2. The ability to manipulate nerve recovery at the cellular or gene level will provide the next significant improvement in nerve recovery.
3. Stimulation by nerve cuff electrode superseded cumbersome muscle based electrode but intraoperative stimulation by development of fascicle cuff electrode (Author's innovative idea) may be another improvement over nerve cuff electrode. But this requires further research to find the dimensions of CFs contributing to innervating nerve.
4. Fascicular anatomy of the branch-free length of the femoral nerve distal to the inguinal ligament, the length of branch-free portion, its dimensions, the level and order of origin of various motor branches has direct clinical applications and should be further investigated in more number of cases.

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