Interfacing a next generation sequencing bioinformatic pipeline with a laboratory information management system for use with high resolution typing results and reporting

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Introduction

- NGS being implemented widely
- Challenge: Handling of high resolution results in a clinical laboratory
- Manual data processing has significant risk of human error
- Important to implement an appropriate LIMS for HLA laboratories
 - LIMS emphasis on interoperability (fully-connected LIMS)
 - LIMS vendor commitment to interface with multiple NGS filetypes (JSON, HML, XML, etc.)
- Seamless integration of different workflows
 - Both NGS & LIMS vendor must support flexible & adaptive workflows
 - The study confirmed lab's current workflow can be achieved with mTilda & Holotype
- LIMS validation
 - 2 levels of validation: procedural validation (SOP-driven workflow validation) and data integrity validation (validation of discrete-data elements for high resolution typing as well as valid nomenclature)

Study Design Diagram



Patient registration module

🔊 mTilda Main Menu - [1 - Patient M	laintenance: Patien	t Demographics]	
5. Log In/Out Patients Providers Scoring Test Review Inquiry Dat	ta Entry Reagents Ti	rays <u>M</u> onthly Screening	<u>W</u> orklists <u>A</u> dmin Pri <u>n</u> t Wind <u>o</u> w <u> </u>
N Number Last / Alias First / Alias	s	Middle / Alias	Search Patients
Category BMR Bone Marrow Recipient	▼ R		E <u>x</u> it Screen
Demographics Events Provide	ers Re	lations Ad	ldress Cadaver
Date of Birth 2/22/2004 V Age: 15 Ethnic Group HIS Hispanic or Latino V ABO/Rh A V O Pos Neg Unknown	 ○ Female ● Male ○ Unknown 	Wait List Status: WUNOS Date: 3	V ▼ ▼ 3/27/2019 ▼
Ty FN SSN Medicare Number Sc Medical Record 123456 Xm	Status () In Patient () Out Patient () Unknown	User Defined Fields 1 NGS Test Storage Box Freezer 1	2
Li ICD 9 ◯ 10 Diagnosis S31.1095		Account # 1234567	8
Unspecified open wound of abdominal wall, unspecified quadra	nt witho 🔻	Account # Date 37	27/2019 🔹 12:00:00 AM 🗘
Comments Patient comment			
New Patient Archive Cancel Edit/A	Add	<u>D</u> elete Patient	Save Edit/Add
User: Director - mTilda			

Sample Management

55		Log Patient	Samples - Double (click on any	y sample t	o load that	sample fo	or	editing.		x
P. F P	henotype	Name Mtilda, Omi: A*01:01:01:01,68:0	xon)2:01:01; B*53:01:01,08:0	1:01:01; C*04:	01:01:01,07:0	ов <u>2/22/200</u> 1:01:01; DRB1	14 Cat Bl 1*03:01:01:01,	MR 15:	аво (03:01:0	Search <u>P</u> atier E <u>x</u> it Screer	nts n
\square	Log S	amples	List Samples	S	ample Inve	ntory	DNA Ex	tra	octions Patient /	Sample Dat	a
Γ	Sample #	Draw Date	Log Date / Type	Logged By / # Tubes	Diagnosis	Discard Date	Description	E	Comments		Ā
	2	3/3/2019	3/4/2019 11:02:00 AM	D	S31.109S			Y	Sample comment		
	1	2/21/2019	2/22/2019 9:16:00 AM ACD	D			Confirmatory	Y	NGS Sample Comment		

HLA Test Order Entry

ii ii	Log Samples: Log Tests – 🗖 🔼 🗙
Patient Name Mtilda, Omixon Phenotype A*01:01:01:01,68:02:01 Log Test	DOB 2/22/2004 Category BMR ABO Pos 1:01; B*53:01:01,08:01:01:01; C*04:01:01:01;07:01:01:01; DRB1*03:01:01:01;15; Exit Screen List Test Requests Additional Patient/Test Data
Sequencing DNA SBT CI I II SBT CI I II SBT CL I SBT A Locus (new) SBT B Locus (new) SBT DR Locus (new) SET DR Locus (new) SBT DR Locus (new) SST DR LOCUT (Noninex/DE ST Crossmatch Auto CDC/DTT SOT Crossmatch Auto	Stat Test Expedite Test Type NGS Class I and II Request Date 2/22/2019 Study Test User Field 2 Billing Billing Billing Search Physicians I Dr. Augusta Bob DOB DOB ABO

Holotype HLA NGS Workflow



Result Review and Allele Assignment in HLA Twin

Genotyping Result of 1 analysis			Renata Santos@FAS	19% [memory usage]	🖹 No tasks running 😽 🕂	a (p)
Correction Genotyping analys	Sis result Analysis na	ame(s) JM-20190214S-33_S	9_L001_R1_001_2019-02-20_21-10-34)?
Sample Browse Alignment Setup Loci Filters Setup Only State Workflow State Genotyp	Assignment Comments	pprove Reject/Revoke Approval	HPRIM Export LD on/off		Displaying 11 loci c Displaying best matc	out of 11. ches only.
Sample Allele HLA-A	HLA-B	HLA-C	HLA-DPA1	HLA-DPB1	HLA-DQA1	
.001_R1_001_2019-02-20_21-10-34 Allele 1 HLA-A*25:01:01	✓ ● HLA-B*18:01:01 ① ▼ < ●	HLA-C*03:02:02	✓ ● HLA-DPA1*01:03:01 ①	HLA-DPB1*01:01:01		0
.001_R1_001_2019-02-20_21-10-34 Allele 2 🗸 🌢 HLA-A*33:03:01 🛛 🗨	✓ ● HLA-B*58:01:01 ① ▼ < ●	HLA-C*12:03:01 0	✓ ● HLA-DPA1*02:02:02 ①	HLA-DPB1*02:01:02	2 ① ▼ ● HLA-DQA1*01:02:01	0
	HLA-B					
	✓ ● HLA-B*18:01:0	01 🔘 👅				
	✓ ● HLA-B*58:01:0	01 0				
	o Accignment etc					

Data Export and File Structure in HLA Twin

JSON Export Wizard in HLA Twin

port format ?	HML	PDF	JSC	N	
utput folder	Users/Holoytpe-NGS/Desktop			+	•

JSON file format

Manuar Track
viewer lext
Paste Copy Format Remove white space Clear Load JSON data
{
"sampleName": "1-1-DATE-1",
"analysisRunName": "PM-201902145-27_53_L001_R1_001_2019-02-20_22-08-23",
"analysisDate": "Feb 20, 2019 10:08:30 PM",
"executedBy": "Renata Santos@FAS",
"approvedBy": "Not approved",
"sampleComment": "No comment available",
"genotypes": [
ί "σene", "ΗΙΔ-Δ"
"genotype": "01:01:01:01:68:02:01:01"
"genotypeGGroup": "01:01:01G+68:02:01G".
"genotypePGroup": "01:01P+68:02P"
h
{
"gene": "HLA-B",
"genotype": "08:01:01:01+53:01:01\n08:01:01:02+53:01:01",
"genotypeGGroup": "08:01:01G+53:01:01G\n08:01:01G+53:01:01G",
"genotyperGroup": "08:01P+53:01P\n08:01P+53:01P"
"gene": "HLA-C".
"genotype": "04:01:01:01+07:01:01\n04:01:01:01+07:01:01:08\n04:01:01:01+07:
"genotypeGGroup": "04:01:01G+07:01:01G\n04:01:01G+07:01:01G\n04:01:01G+07:01:0
"genotypePGroup": "04:01P+07:01P\n04:01P+07:01P\n04:01P+07:01P\n04:01P+07:01P\
},
{
"gene": "HLA-UPA1", "geneture": "H01.03.01.02.02.02.04"
genotype: 01:03:01:02+02:02:02:02:04 ,
"genotypeBGroup": "01:03P+02:02P"
}.
- (
"gene": "HLA-DPB1",
"genotype": "01:01:01:04+04:01:01",
"genotypeGGroup": "01:01:01G+04:01:01G",
"genotypePGroup": "01:01P+04:01P"

NGS Interface



Structured Data Processing

₩ mTilda		-	×
V m tilda	mTilda NGS	S	
File Type:			
Omixon	T		
File:			
PM-20190214S-2	7 •		
Process Data			
FIOCESS Data			

Typing Summary

\$ 5.			Typin	g Hist	ory			×
Ļ	Test Type	Sample Date Comp	letion S	itatus				<u>^</u>
	NGS Class I and I	2/21/2019 2/2//	2013 10:5 0	, <u> </u>	Aron:on:on:on:ar68:02:01:01 >>> Allele 1 A*01:01:01:01 B*53:01:01 C*04:01:01:01 DRB1*03:01:01:01 DRB5*01:01:01 DRB3*01:01:02:01 DQA1*01:02:01:03 DQB1*02:01:01 DPA1*01:03:01:02 DPB1*04:01:01:01	Allele 2 A*68:02:01:01 B*08:01:01:01 C*07:01:01:01 DRB1*15:03:01:01 DRB5*01:01:02 DQA1*05:01:01:02 DQB1*06:02:01:01 DPA1*02:02:02:04 DPB1*01:01:01:04		
		_⊻	iew Excludec	d/Txp Ai	ntigens <u>P</u> rint As Report	<u>P</u> rint	E <u>:</u>	git

Test Result Details

80	1 - Test Inquiry			×
Search Criteria	Test List	Ĭ	Test	Detail
Patient: Mtilda, Omixon] 1	E <u>x</u> it Screen
Category: BMR SSN:	Medical Record: 123456			
Supervisor				
Recipient/Donor			Order # 0	Test # 1
Test Type NGS Class I and II	NGSC12 Su	pervisor	Director D	Tech D
Reg Physician Dr. Augusta	1 Re	quest Date 2/22	2/2019	Ву D
User Field 1 User 2	Te	ch Forward 2/27	7/2019 10:56:24 AM	Status
Results	An	proval Date		Complete
Allele 1 Allele 2	<u>^</u>			
A*01:01:01:01 A*68:02:01:01 B*53:01:01 B*08:01:01:01	≡ Co	mpletion Date 2/27	7/2019 10:56:24 AM	Billing Flag
C*04:01:01:01 C*07:01:01:01	Ch	arge Date		B
DR81^03;01:01:01 DR81^15;03;01:01 DR85*01+01+01 DR85*01+01+01				
	He	port Date		Lesting Reagents
Sample Information	Test Comments			
Draw Date / # 2/21/2019C 1				
Description Confirmatory				
Log Date/Time 2/22/2019 9:16:00 AM				
Log Tech D				
				~
Change Tech Change Status Change Billing	Report Comments	<u>V</u> iew Patient	View S <u>c</u> oring Detail	View S <u>a</u> mple Detail

Patient Phenotype Management

Patient 1 Name Mtilda, Omixon				D	ов 2/22/2004	Cat BMR	АВО /Р	s	earch <u>P</u> atients
Phenotype A*01:01:01:01,68:0	02:01:01; B*53:01:01,08:01:0	01:01; C*04:01:01:01,07	:01:01:01; D	RB1*03:01	:01:01,15:03:01	:01; DRB5*01:01:	01,01:01:01;		E <u>x</u> it Screen
Typing History		Date Tested	3/27/2019	9 T					
Append Results	Test Results			Currrent	Phenotype		Report P	henotype	
to Current	Locus Allele 1	Allele 2	^	A*	01:01:01:01	68:02:01:01	A*	01:01	68:02
to Current			=	B*	53:01:01	08:01:01:01	B*	53:01	08:01
to Both				C*	04:01:01:01	07:01:01:01	C*	04:01	07:01
				DRB1*	03:01:01:01	15:03:01:01	DRB1*	03:01	15:03
Sero Equiv to Report				DRB5*	01:01:01	01:01:01	DRB5*	01:01	01:01
				DRB3*	01:01:02:01	01:01:02:02	DRB3*	01:01	01:01
Truncata Reculta				DQA1*	01:02:01:03	05:01:01:02	DQA1*	01:02	05:01
Funcate Results				DQB1*	02:01:01	06:02:01:01	DQB1×	02:01	06:02
Truncate to Current				DPA1*	01:03:01:02	02:02:02:04	DPA1*	01:03	02:02
Truncate to Report				DPB1*	04:01:01:01	01:01:01:04	DPB1*	04:01	01:01
Truncate: 01									
○ 2			~						
Comments									
NGS validation project									
	Curata Phone /Co	notros							
Pheno Wizard	Curate Pheno/Ge	enotype			Sort Curren	t		Sort Re	ported
Report Comments	Convert From	NMDP Convert to	NMDP						-
Copy to Exclude Pheno	Copy from	n Current to Report	Convert	: Current to	Report as Sero B	q Delet	te Locus	Undo Cl	hanges
Current Report	◯ Selected	IIA ()	0	Selected	() All	Inse	rt Locus	Save Phe	notypes

Test Results Management

200110						_	_			
	Name Mtilda, O	mixon					DOB 2/2	2/2004 Cat BMR	Search <u>P</u> atients E <u>x</u>	it Screen
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юподро	A.01:01:01:01:01	(U2:U1:U1) Br03:U1:U1,U6	(U1:U1:U1; C*U4:U1:U1:	01,07	:01:01:01; D	וסח	-03:01:01:0	1,15:03:01:01; DHB5*01:0	(1:01,01:01:01) DHB3*01:01:0.	
	Scoring		Results				Analys	is Detail	Test Detail	
Doculto I	ah Amaluaia					D		•		
				_		יש	NA Lest Hi	esults		
Locus	Allele I	Allele 2		<u> </u>		Ļ	Locus	Allele 1	Allele 2	^
▶ A"	01:01:01:01	58:02:01:01				₽	A^ D×	■ 01:01:01:01	68:02:01:01	
D×	00:01:01:01	53:01:01		-		⊢	B ⁿ	53:01:01	08:01:01:01	
D×	08:01:010	53:01:01G 53:01D		-		⊢		04:01:01:01	15:02:01:01	
	04.01.01.01	07.01.01.01		- 🗸		L-	DRB1"	03:01:01:01	15:03:01:01	
11	11141111111	110/111/11/11				L-	DRB5"	01:01:01	01:01:02:02	
Locus	Allele 1	Allele 2		^		⊢	DRB3" DOA1×	01:01:02:01	01:01:02:02	
► A*	01:01:01:01	68:02:01:01				⊢	DUAT" DOB1×	01:02:01:03	05:01:01:02	
B×	08:01:01:01	53:01:01				H-		02.01.01	00.02.01.01	
B×	08:01:01:01	53:01:01		≡		⊢	DDD1×	01.03.01.02	01:01:01:04	
B×	08:01:01:02	53:01:01				-		04.01.01.01	01.01.01.04	
C*	04:01:01:01	07:01:01:01				-				
C*	04:01:01:01	07:01:01:01								
C*	04:01:01:01	07:01:01:08			Show					
C*	04:01:01:01	07:01:01:16		_	Alleles					
C×	04:01:01:11	07:01:01:01		_		Ļ				•
C×	04:01:01:11	07:01:01:08		_				Constants Allali	Convert to NIME	n cada
C*	04:01:01:11	07:01:01:16		_	0	- 10		Concatenate Allei	Convert to NML	P Code
L LC*	04:01:01:14	07:01:01:01			Antiger	n w	Izaro			