

Morphometric formula for field determination of the sex of Steppe Eagle (*Aquila nipalensis*) nestlings in Altai-Sayan region



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The measurements we used

- **DK** wing length
- **DP** 5th flight feather length
- **DH** tail length
- **DC** tarsus length
- **SHC1** transverse tarsus diameter
- **SHC2** longitudinal tarsus diameter
- **KN** outer claw length
- **KS** middle claw length
- **KV** inner claw length
- **KZ** rear claw length
- **DKL** beak length from the tip to the forehead
- **DKV** beak length from the tip to the cere
- **DKN** beak length from the tip to the anterior edge of the nostrils
- **VK** beak depth
- **RR** gape's size

C-052 C-055 C-056 C-777 C-918

♀ ♂ ♂ ♀ ♀

1kb -

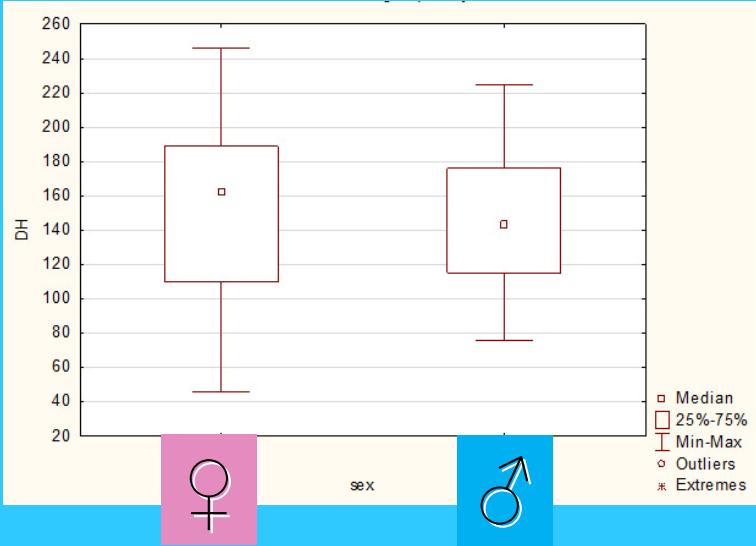
Genetic sex determination

CHD1 gene intron

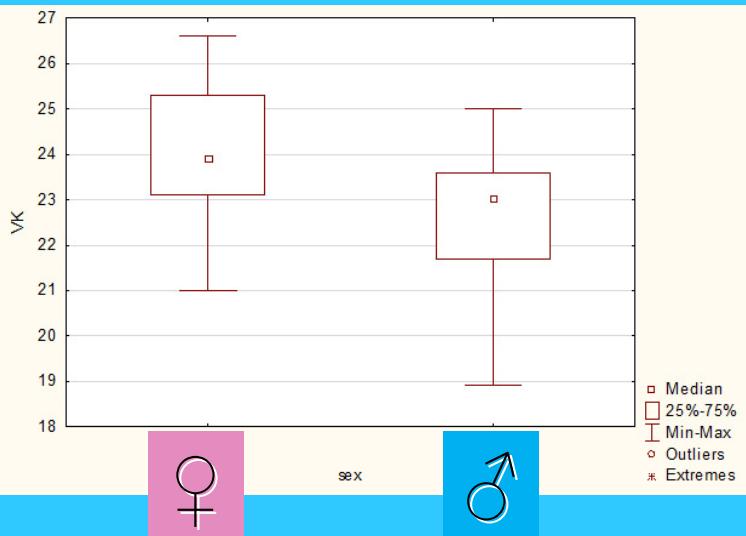
Z chromosome
W chromosome

Box Plots grouped by sex

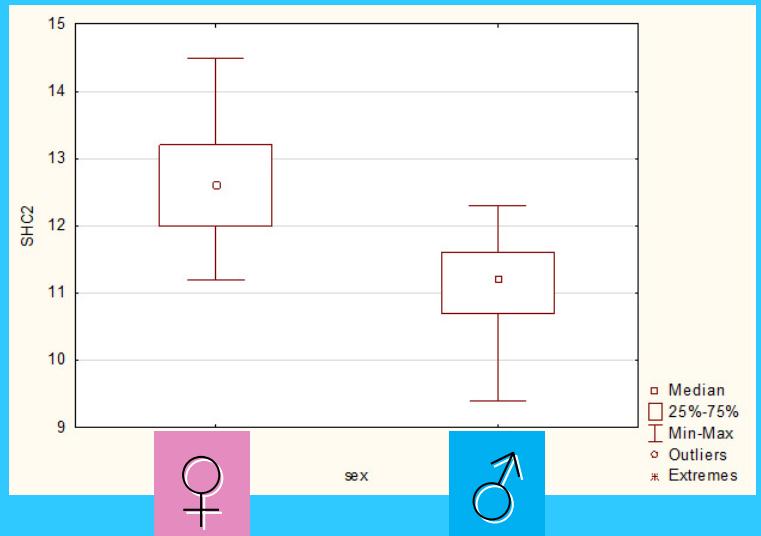
tail length



beak depth



longitudinal tarsus diameter



Correlation matrix

	feathers	tarsus				claws				beak						
	sex	DK	DP	DH	DC	SHC1	SHC2	KN	KS	KV	KZ	DKL	DKV	DKN	VK	RR
sex		7%	4%	6%	29%	73%	73%	53%	49%	55%	61%	53%	42%	50%	43%	36%
DK	7%		98%	96%	72%	25%	2%	55%	60%	67%	65%	78%	81%	78%	70%	82%
DP	4%	98%		95%	63%	22%	-1%	50%	57%	63%	60%	72%	77%	75%	63%	76%
DH	6%	96%	95%		69%	26%	8%	57%	63%	68%	64%	76%	83%	79%	75%	80%
DC	29%	72%	63%	69%		46%	26%	71%	71%	77%	74%	76%	78%	77%	77%	82%
SHC1	73%	25%	22%	26%	46%		81%	50%	50%	57%	62%	52%	47%	58%	57%	50%
SHC2	73%	2%	-1%	8%	26%	81%		44%	42%	46%	48%	37%	32%	43%	49%	30%
KN	53%	55%	50%	57%	71%	50%	44%		95%	93%	93%	76%	80%	81%	70%	70%
KS	49%	60%	57%	63%	71%	50%	42%	95%		96%	94%	75%	80%	81%	70%	71%
KV	55%	67%	63%	68%	77%	57%	46%	93%	96%		97%	83%	86%	86%	79%	79%
KZ	61%	65%	60%	64%	74%	62%	48%	93%	94%	97%		86%	85%	87%	78%	80%
DKL	53%	78%	72%	76%	76%	52%	37%	76%	75%	83%	86%		93%	92%	84%	92%
DKV	42%	81%	77%	83%	78%	47%	32%	80%	80%	86%	85%	93%		96%	86%	91%
DKN	50%	78%	75%	79%	77%	58%	43%	81%	81%	86%	87%	92%	96%		84%	88%
VK	43%	70%	63%	75%	77%	57%	49%	70%	70%	79%	78%	84%	86%	84%		86%
RR	36%	82%	76%	80%	82%	50%	30%	70%	71%	79%	80%	92%	91%	88%	86%	



Correlation matrix

	feathers				tarsus				claws				beak				
	sex	DK	DP	DH	DC	SHC1	SHC2	KN	KS	KV	KZ	DKL	DKV	DKN	VK	RR	
sex		7%	4%	6%	29%	73%	73%	53%	49%	55%	61%	53%	42%	50%	43%	36%	
DK		7%		98%	96%	72%	25%	2%	55%	60%	67%	65%	78%	81%	78%	70%	82%
DP		4%	98%		95%	63%	22%	-1%	50%	57%	63%	60%	72%	77%	75%	63%	76%
DH		6%	96%	95%		69%	26%	8%	57%	63%	68%	64%	76%	83%	79%	75%	80%
DC		29%	72%	63%	69%		46%	26%	71%	71%	77%	74%	76%	78%	77%	77%	82%
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SHC2		73%	2%	-1%	8%	26%	81%		44%	42%	46%	48%	37%	32%	43%	49%	30%
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KS		49%	60%	57%	63%	71%	50%	42%	95%		96%	94%	75%	80%	81%	70%	71%
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KZ		61%	65%	60%	64%	74%	62%	48%	93%	94%	97%		86%	85%	87%	78%	80%
DKL		53%	78%	72%	76%	76%	52%	37%	76%	75%	83%	86%		93%	92%	84%	92%
DKV		42%	81%	77%	83%	78%	47%	32%	80%	80%	86%	85%	93%		96%	86%	91%
DKN		50%	78%	75%	79%	77%	58%	43%	81%	81%	86%	87%	92%	96%		84%	88%
VK		43%	70%	63%	75%	77%	57%	49%	70%	70%	79%	78%	84%	86%	84%		86%
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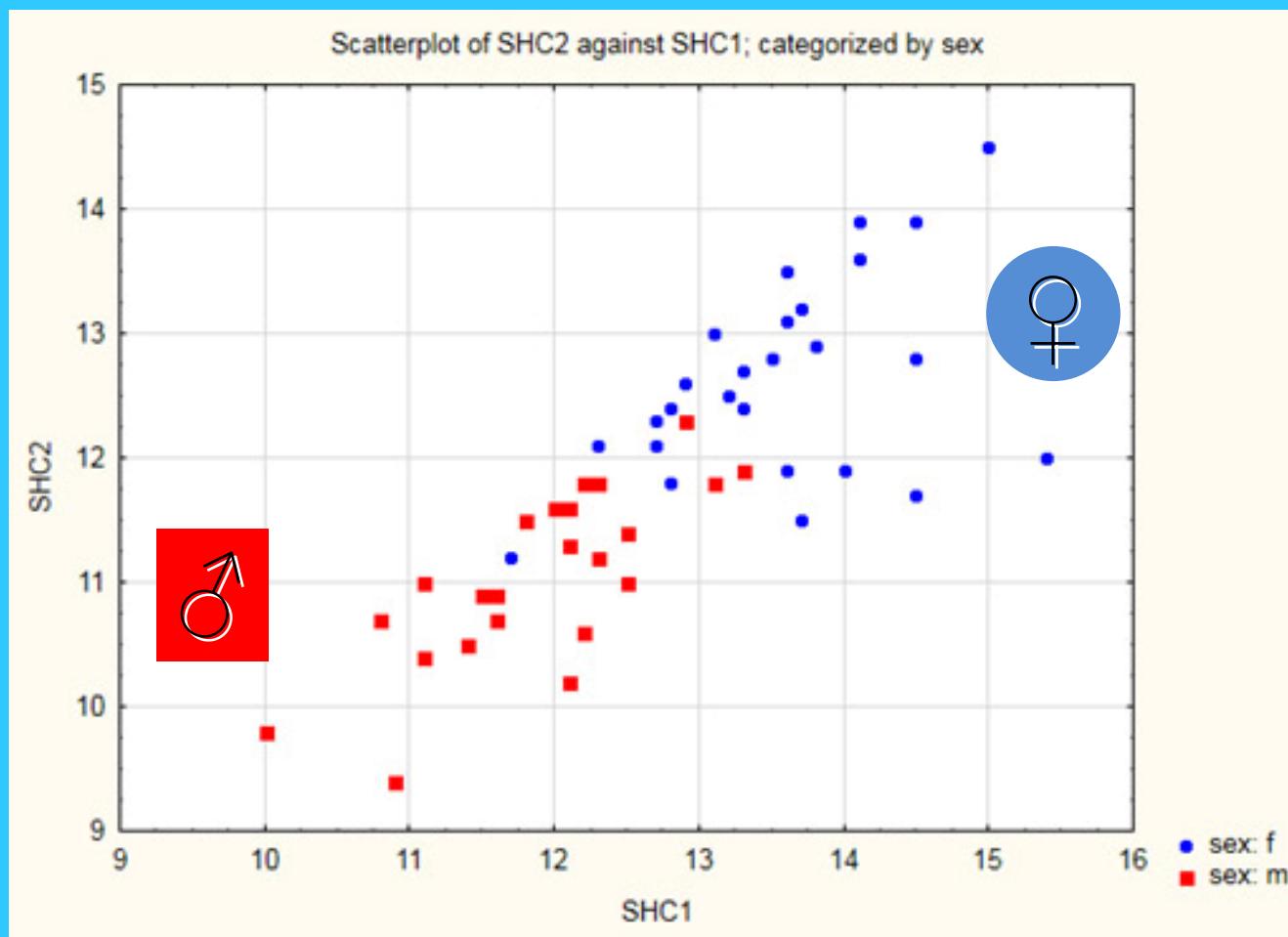


Correlation matrix

	feathers	tarsus			claws			beak								
	DK	DP	DH	DC	SHC1	SHC2	KN	KS	KV	KZ	DKL	DKV	DKN	VK	RR	
sex		7%	4%	6%	29%	73%	73%	53%	49%	55%	61%	53%	42%	50%	43%	36%
DK	7%		98%	96%	72%	25%	2%	55%	60%	67%	65%	78%	81%	78%	70%	82%
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SHC1	73%	25%	22%	26%	46%		81%	50%	50%	57%	62%	52%	47%	58%	57%	50%
SHC2	73%	2%	-1%	8%	26%	81%		44%	42%	46%	48%	37%	32%	43%	49%	30%
KN	53%	55%	50%	57%	71%	50%	44%		95%	93%	93%	76%	80%	81%	70%	70%
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KZ	61%	65%	60%	64%	74%	62%	48%	93%	94%	97%		86%	85%	87%	78%	80%
DKL	53%	78%	72%	76%	76%	52%	37%	76%	75%	83%	86%		93%	92%	84%	92%
DKV	42%	81%	77%	83%	78%	47%	32%	80%	80%	86%	85%	93%		96%	86%	91%
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VK	43%	70%	63%	75%	77%	57%	49%	70%	70%	79%	78%	84%	86%	84%		86%
RR	36%	82%	76%	80%	82%	50%	30%	70%	71%	79%	80%	92%	91%	88%	86%	



SCH1 / SCH2 transverse / longitudinal tarsus diameters



Forward stepwise discriminant function analysis

Measurements that contribute the most to division on sex:

N=52	Wilks'	Partial
SHC1	0,203022	0,881685
SHC2	0,188045	0,951911
KZ	0,179575	0,996809
DH	0,241944	0,739849
DKL	0,242545	0,738014
RR	0,197664	0,905587
DC	0,185943	0,962670
KV	0,190732	0,938499
KS	0,185720	0,963826

Classification Functions

Variable	coefficient
SHC1	-3,1286168
SHC2	-1,7891307
KZ	-0,5181316
DH	0,1088256
DKL	-2,013826
RR	0,7963207
DC	0,2784553
KV	-2,3011411
KS	1,6286852
Constant	107,7284009

$F < -1$ ♀

$F > 1$ ♂



feathers tarsus claws beak

	sex	DK	DP	DH	DC	SHC1	SHC2	KN	KS	KV	KZ	DKL	DKV	DKN	VK	RR
sex		7%	4%	6%	29%	73%	73%	53%	49%	55%	61%	53%	42%	50%	43%	36%
DK	7%		98%	96%	72%	25%	2%	55%	60%	67%	65%	78%	81%	78%	70%	82%
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DC	29%	72%	63%	69%		46%	26%	71%	71%	77%	74%	76%	78%	77%	77%	82%
SHC1	73%	25%	22%	26%	46%		81%	50%	50%	57%	62%	52%	47%	58%	57%	50%
SHC2	73%	2%	-1%	8%	26%	81%		44%	42%	46%	48%	37%	32%	43%	49%	30%
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KS	49%	60%	57%	63%	71%	50%	42%	95%		96%	94%	75%	80%	81%	70%	71%
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KZ	61%	65%	60%	64%	74%	62%	48%	93%	94%	97%		86%	85%	87%	78%	80%
DKL	53%	78%	72%	76%	76%	52%	37%	76%	75%	83%	86%		93%	92%	84%	92%
DKV	42%	81%	77%	83%	78%	47%	32%	80%	80%	80%	85%	93%		90%	88%	31%
DKN	50%	78%	75%	79%	77%	58%	43%	81%	81%	86%	87%	92%	96%		84%	88%
VK	43%	70%	63%	75%	77%	57%	49%	70%	70%	79%	78%	84%	86%	84%		86%
RR	36%	82%	76%	80%	82%	50%	30%	70%	71%	79%	80%	92%	91%	88%		86%

- We tried a lot of different combinations of variables:

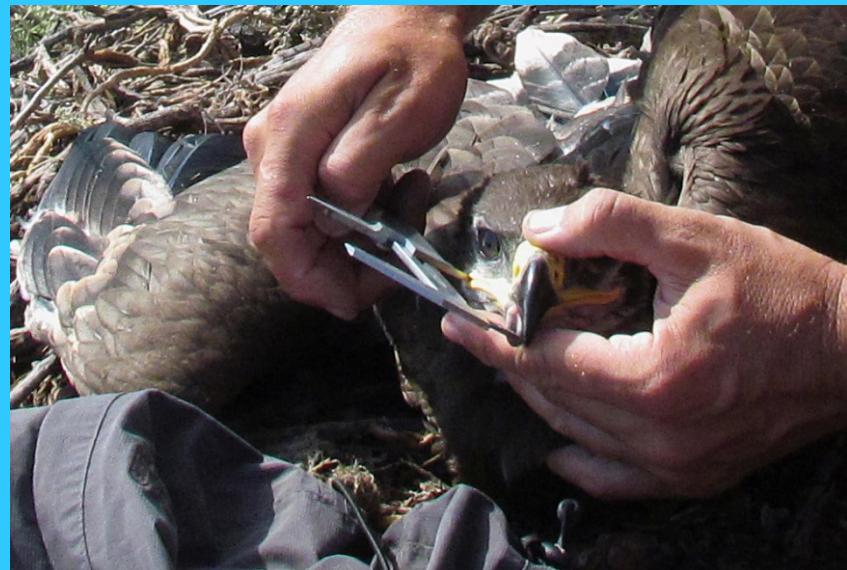
1. Removed from the analysis redundant variables that are highly correlated with others.
2. Added ratios of variables to each other as "normalization for age".
3. Removed feather length.
4. Removed a suspicious sample, which was most likely misclassified.



N=52	Wilks'	Partial
SHC1	0,216155	0,772331
RR/DKL	0,173042	0,964753
KN/DH	0,249323	0,669584
DKL	0,207680	0,803845
VK/SHC2	0,188285	0,886648
VK/SHC1	0,176706	0,944746

Testing of our formulas

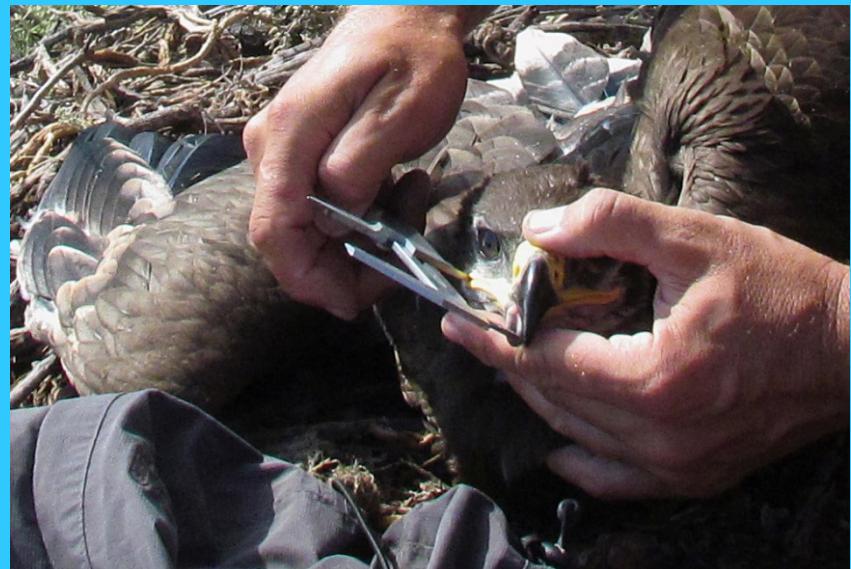
	Training sample 2019 (52=27f+25m):		Validation sample 1 2021 (33=12f+21m):		Validation sample 2 2022 (rachitics) (42=25f+17m):	
	misclassified	Not classified	misclassified	Not classified	misclassified	Not classified
F-16	1 (2%)	0	1 (3%)	1 (3%)	6 (14%)	4 (10%)
F-1	0	1 (2%)	0	1 (3%)	6 (14%)	1 (2%)
F-3	0	1 (2%)	0	1 (3%)	6 (14%)	3 (7%)
F-28	0	1 (2%)	1 (3%)	0	3 (7%)	2 (5%)



Testing of our formulas

	Training sample 2019 (52=27f+25m):		Validation sample 1 2021 (33=12f+21m):		Validation sample 2 2022 (rachitics) (42=25f+17m):	
	misclassified	Not classified	misclassified	Not classified	misclassified	Not classified
F-16	1 (2%)	0	1 (3%)	1 (3%)	6 (14%)	4 (10%)
F-1	0	1 (2%)	0	1 (3%)	6 (14%)	1 (2%)
F-3	0	1 (2%)	0	1 (3%)	6 (14%)	3 (7%)
F-28	0	1 (2%)	1 (3%)	0	3 (7%)	2 (5%)

13 of 25 females
were misclassified by
at least one of these
formulas



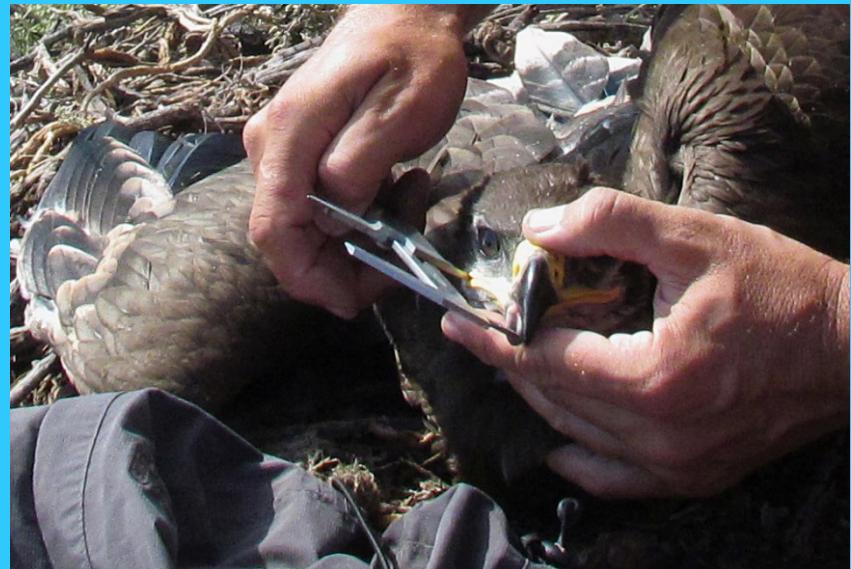
Testing of our formulas

	Training sample 2019 (52=27f+25m):		Validation sample 1 2021 (33=12f+21m):		Validation sample 2 2022 (rachitics) (42=25f+17m):	
	misclassified	Not classified	misclassified	Not classified	misclassified	Not classified
F-16	1 (2%)	0	1 (3%)	1 (3%)	6 (14%)	4 (10%)
F-1	0	1 (2%)	0	1 (3%)	6 (14%)	1 (2%)
F-3	0	1 (2%)	0	1 (3%)	6 (14%)	3 (7%)
F-28	0	1 (2%)	1 (3%)	0	3 (7%)	2 (5%)

13 of 25 females were misclassified by at least one of these formulas

Variables contributing significantly to sex discrimination for the 2022 sample (rachitics):

N=42	Wilks'	Partial
SHC2	0,338702	0,753697
KZ	0,266853	0,956626
DC	0,332003	0,768904
DH	0,296442	0,861141
KS	0,282599	0,903323
KV	0,279564	0,913133
VK	0,272383	0,937206
DKV	0,263927	0,967232

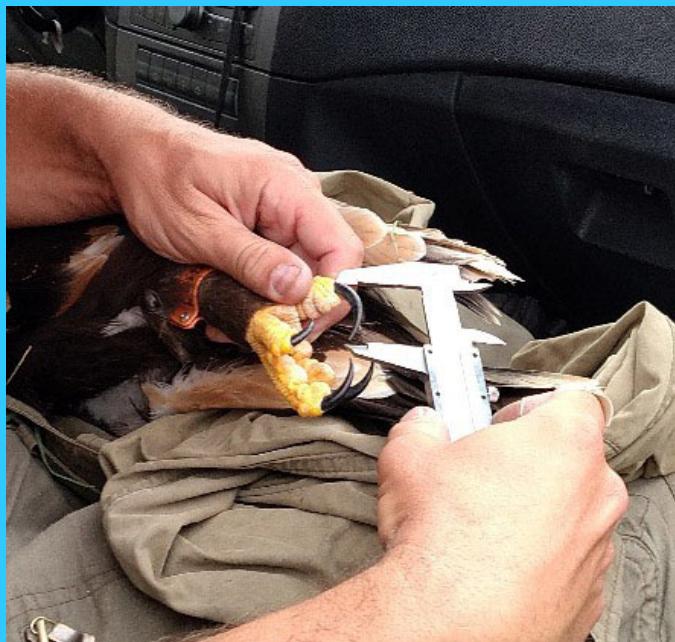


Proportion of misidentified and not unidentified individuals:

	failures (%) (2019+2021, 85 samples)	failures (%) (2022, 42 samples)
F-1	2,4%	17%
F-3	2,4%	21%
F-28	2,4%	12%
F-34	12,9%	2%
F-35	7,1%	2%
F-8	32,9%	45,2%
F-9	36,5%	45,2%

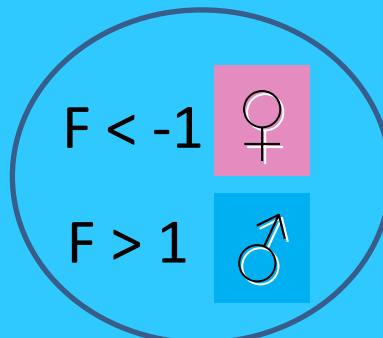
- Formulas obtained using the sample of normal nestlings
- Formulas obtained using the sample of underfed nestlings
- Formulas obtained in 2017
-without claw length
-with average tarsus diameter

(Karyakin I.V., Zinevich L.S., Shnayder E.P. A Possibility of Morphometrical Determining of Sex of Steppe Eagle Nestlings from Western and Eastern Populations? – Raptors Conservation. 2017. 35: 194–218.



Needed:

- DH tail length
- DC tarsus length
- SHC1 transverse tarsus diameter
- SHC2 longitudinal tarsus diameter
- KN outer claw length
- KS middle claw length
- KV inner claw length
- KZ rear claw length
- DKL beak length from the tip to the forehead
- DKN beak length from the tip to the anterior edge of nostrils
- VK beak depth
- RR gape's size



Normal nestlings:

F-1

Variable	coefficient
SHC1	-5,1122774
RR/DKL	23,961914
KN/DH	-106,289352
DKL	-1,3013396
VK/SHC2	16,3629746
KV/SHC1	-10,851755
Constant	103,7428332

F-3

Variable	coefficient
SHC1	-4,5290584
RR/DKL	22,116452
KZ/DH	-149,583703
DKL	-1,3072951
VK/SHC2	16,8208038
SHC1/DH	199,5877861
Constant	72,5314095

F-28

Variable	coefficient
SHC1	-4,9974374
RR/DKL	34,5081015
KZ/DC	-89,329643
DKL/DH	-185,60443
KV	-3,1990834
KS/DH	-82,4135428
KS/SHC2	38,6243393
SHC2/DH	672,1133476
Constant	81,1494196

Nestlings with weight deficiency:

F-34

Variable	coefficient
KZ/DC	-130,292548
SHC2	-8,4384238
DC/DH	-19,5329671
KV/SHC2	-22,0043082
VK/DKN	-32,710061
Constant	236,3751645

F-35

Variable	coefficient
KZ/DC	-118,181176
SHC2	-8,61708
DC/DH	-19,3085922
KV/KS	-77,9847816
KV/SHC2	-19,3150855
Constant	294,9180811

! have not been tested on an independent sample

Igor Karyakin
Elena Shnayder



Natalya Andreyenkova
Maria Shalnova