Journal of Threatened Taxa | www.threatenedtaxa.org | 26 August 2015 | 7(10): 7668–7675

New distribution records of *Epiophlebia laidlawi* Tillyard, 1921 (Insecta: Odonata) in Bhutan

ISSN 0974-7907 (Online)
ISSN 0974-7893 (Print)

OPEN ACCESS

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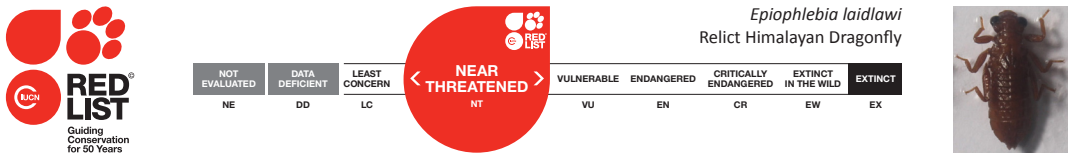
Abstract: An opportunistic survey for *Epiophlebia laidlawi* larvae was carried out within five districts in Western and Central Bhutan from 2012 to 2014. The study recorded a total of 21 individuals from five districts and also recorded F0, F3 and F8 instars larvae for the first time in Bhutan. The study adds December and February as possible months to record F0 instars within its range. The record of *E. laidlawi* from Bumthang District extends its range to the eastern most part of the Himalayas, and it also extends its range from Chhukha District to its southern most range within Bhutan. A record from Punakha District fills the gap between the previous and current record of *E. laidlawi* from Wangchhu basin in Western Bhutan and Drangmechhu basin spanning central and eastern Bhutan with Punatshangchhu basin in between. The record from Trongsa District emphasises the importance of the study area as *E. laidlawi*'s habitat. The extent range of *E. laidlawi* within Bhutan is now extended to six districts, viz., Haa, Thimphu, Chhukha, Punakha, Trongsa and Bumthang.

Keywords: Anisozygoptera, Bhutan, distribution, *Epiophlebia laidlawi*, larva.

Epiophlebia laidlawi Tillyard, 1921 is one of the four species belonging to the order Odonata, suborder Anisozygoptera, family Epiophlebiidae. It is recorded from Nepal, India and Bhutan (Carle 2012). Species belonging to the genus *Epiophlebia* are considered relict species since they display features of both damselflies (Zygoptera) and dragonflies (Anisoptera) (Büsse et al. 2012). They resemble dragonflies in their general body outline, while they resemble damselflies in having similarly shaped and petiolate fore and hindwings (Büsse et al. 2012). Out of the four extant species of *Epiophlebia*, *E. sinensis* (Li et al. 2012) and *E. diana* (Carle 2012) were recently described from Heilongjiang and Sichuan provinces respectively in China, while *E. laidlawi* and *E. superstes* were described from India and Japan respectively (Brockhaus & Hartmann 2009; Carle 2012;



Epiophlebia laidlawi
Relict Himalayan Dragonfly

DOI: http://dx.doi.org/10.11609/JoTT.o4092.7668-75 | **ZooBank:** urn:lsid:zoobank.org:pub:44355FD9-F7A9-435B-B911-46EAAB27D4E0

Editor: K.A. Subramanian, Zoological Survey of India, Kolkata, India. **Date of publication:** 26 August 2015 (online & print)

Manuscript details: Ms # o4092 | Received 08 July 2014 | Final received 08 June 2015 | Finally accepted 28 July 2015

Citation: Dorji, T. (2015). New distribution records of *Epiophlebia laidlawi* Tillyard, 1921 (Insecta: Odonata) in Bhutan. *Journal of Threatened Taxa* 7(10): 7668–7675; http://dx.doi.org/10.11609/JoTT.o4092.7668-75

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Funding: Rufford Small Grants Foundation and Royal University of Bhutan.

Competing interests: The author declares no competing interests.

Acknowledgements: I would like to thank Ruffords Small Grants Foundation and Royal University of Bhutan for partially funding the current work. I would like to also thank the Ugyen Wangchuk Institute for Conservation and Environment (UWICE) for letting me participate in the freshwater biodiversity training workshop and workshop participants for dragonfly larvae. My special thanks are to Professor Anne Hartmann, Associate Porfessor Winsor Lowe and Associate Professor Amit Mitra for sharing their articles and / or resources, and students of the B.Sc. Sustainable Development 2nd Batch, Diploma in Forestry, 22nd Batch, Assistant Professor D.B. Gurung, College of Natural Resources for the field work in Trongsa, and other students who helped me in numerous field visits. I would also like to thank Mr. Ugyen Thinley, CNR for helping me generate GIS maps for the study area and results. Finally, I would like to thank the anonymous reviewer for their critical review and suggestions that enhanced the quality of the paper.

Li et al. 2012). Out of the latter two species *E. laidlawi* is not well studied (Brockhaus & Hartmann 2009; Büsse et al. 2012; Carle 2012; Li et al. 2012). It is assessed as Near Threatened based on its restricted range, insufficient data on its distribution and population size, and it being a relict species (Clausnitzer 2006).

E. laidlawi was recorded from Bhutan in 2006 (Brockhaus & Hartmann 2009) from three districts, viz., Haa and Thimphu in western Bhutan, and Trongsa in central Bhutan from larval studies. The adult dragonfly studies in eastern Bhutan by Mitra (2002, 2006) and Mitra & Thinley (2006), and in southeastern Bhutan by Mitra et al. (2012) failed to record adult *E. laidlawi,* and thus adult *E. laidlawi* is yet to be recorded from Bhutan (Brockhaus & Hartmann 2009). Besides, larval development studies have been scanty for *E. laidlawi*, and it is very difficult to collect F0 or the ultimate larva of the species, as it probably moves out of streams to live semi-terrestrially by April/May until it emerges like that of *E. superstes.* Previous studies have recorded F0 of *E. laidlawi* only in March and October samples (Brockhaus & Hartmann 2009). Therefore, with the difficulty of recording adults due to their short flight period (Brockhaus & Hartmann 2009; Carle 2012) it is important to conduct exploratory larval distribution studies within its possible ranges. The current study aims at presenting the larval distribution of *E. laidlawi* in western and central Bhutan, and also to describe the larval development stage.

Materials and Methods

Study area

The study area spans across five districts in Bhutan, viz.: Punakha, Thimphu and Chhukha in western Bhutan, Trongsa and Bumthang in central Bhutan. The streams sampled within Punakha are the headwater streams of Punatshangchhu basin, the streams within Thimphu and Chhukha districts form the part of Wangchhu basin, while the rivers and streams sampled within Trongsa and Bumthang form the part of Drangmechhu basin that spreads from central to eastern Bhutan (Wangchuk 2011) (Image 1). The details including district, a stream's name, reach notation, coordinates and altitude for all the sampled streams, rivers and reaches are given in Appendix 1. Different models of Garmin Handheld GPS were used to record coordinates and altitude in the different sampling areas (Images 2–4).

Larval sampling

The sampling for the larva was done as part of five different activities and D-frame nets (25x25 cm) were used at all the reaches, except in Bumthang where kick nets (30x30 cm) were used. The number of samples within each reach, and the number of reaches within each stream also varied from district to district. In Bumthang District sampling was done as a part of the training workshop conducted by the Ugyen Wangchuk Institute of Conservation and Environment on aquatic biodiversity, while in Trongsa District the Nikachhu

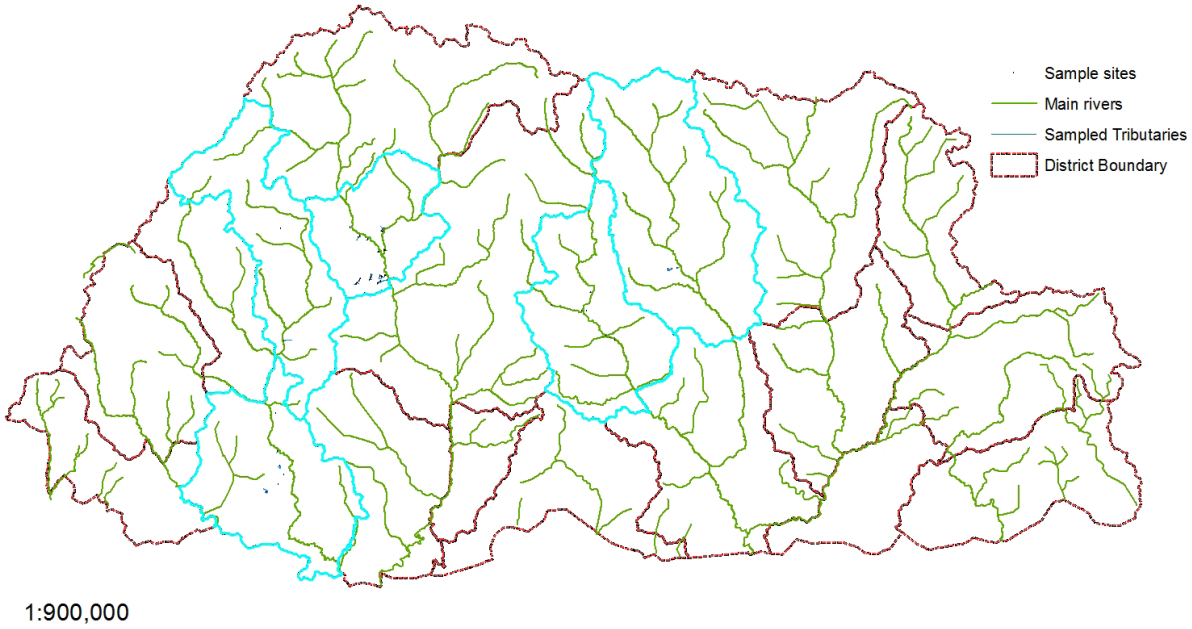

Image 1. Drainage basin of Bhutan with sample sites and highlighted sampled districts.

Image 2. Sampling reaches (a) Stream 1 (inset S1) in Bumthang, (b) Mangduechhu S4 in Trongsa.

Image 3. Sampling reaches D1 and D7 in Dorokna Stream (pre monsoon season).

and Mangdechhu rivers were sampled as part of an aquatic biodiversity assessment from 1–5 December 2012. In Punakha the samplings were done as part of a macroinvertebrate research project and Odonata larval sampling work from October 2012 to March 2014, and other nine streams along Phochhu and Mochhu rivers were sampled as part of a field work by the students of the College of Natural Resources under the guidance of the author. In Thimphu and Chhukha districts nine streams along the Thimphu-Phuntsholing highway were sampled by the author between 2–6 February 2014 as an opportunistic survey. The larvae collected from the sampling sites were stored in glass jars or polyethylene bottles containing 70% ethanol. The larvae are currently deposited in the laboratory of the College of Natural Resources and voucher samples will be deposited in National Biodiversity Centre once proper storage facilities are developed within the centre. The larvae were indentified following Nesemann et al. (2011).

Larval development stage

A detailed measurement of body length from head to abdominal appendages and head capsule width from eye to eye were done using a digital vernier calliper. The reach of wing pad and sex of the larvae were determined for the collections from Bumthang, Thimphu and Chhukha following the work of Tabaru (1984), Brokhaus & Hartmann (2009) and Nesemann et al. (2011). The larvae from Trongsa and Punakha districts were not studied in detail as they were discarded after taking photographs as the field works within the districts were done only to record their occurence.

Results

During the study period a total of 21 *E. laidlawi* larvae were recorded (Table 1). Out of six larvae from Bumthang three were female and one male, while the sex of the other two could not be determined as they were young larvae with inconspicous ovipositor (Table

Image 4. Sampling reaches in Jichulum Stream: J2.1 (post monsoon) and J2.2(pre monsoon).

2; Images 5 & 6). Even though larvae from other study areas were not studied in detail one larva from Nikachhu reach N2 is in F0 stage if judged from the image (Image 7).

Discussion and Conclusion

Records of *E. laidlawi* larvae from two streams in Bumthang District extends the distribution of *E. laidlawi* within Bhutan and in its current known range within the Himalaya to the eastern most part after Trongsa District, the earlier eastern most range. The record from Chhukha District extends its range to the southern most part in Bhutan. The record from Nikachhu River confirms the previous record from its tributary by Brockhaus & Hartmann (2009) and shows the importance of the area as *E. Laidlawi*'s habitat range. The record from Thimphu extends its range within the district southward after that of Brockhaus & Hartmann (2009). Records of *E. laidlawi* larvae from Toebirongchhu sub-watershed in Dorokna

Table 1. Streams and reaches with district names and number of *E. laidlawi* larvae recorded.

Stream and reach	District	No. of larvae
Stream 1(S1)	Bumthang	3
Stream 2(S2)	Bumthang	3
Nikachhu (N1)	Trongsa	2
N2	Trongsa	1
Dorokna (D1)	Punakha	1
D2	Punakha	1
D7	Punakha	1
D8	Punakha	1
Jichulum (J1)	Punakha	3
J2	Punakha	1
J3	Punakha	1
J4	Punakha	1
Drechhu(Dr1)	Thimphu	1
Lobnekha(Lo1)	Chhukha	1

Image 5. *Epiophlebia laidlawi* larvae from two streams in Bumthang District with larva number (1–6) and larval stages.

Table 2. Larval development stage with descriptions and sex of larva number 1–8.

Larvae No.	Stream	Reach	Body length (mm)	Head capsule width (mm)	Wing pad	Stage / Instar	Sex
1	Stream 1	S1	15.2	4.2	Touching 1st abdominal segment	F2	F
2	Stream 2	S2	15.1	4.2	Touching 1st abdominal segment	F2	F
3	Sream 1	S1	15.1	4.2	Touching 1st abdominal segment	F2	F
4	Stream 1	S1	8.8	2.9	Reaching enterior margin of abdomen	F4	M
5	Stream 2	S2	5.2	1.8	Not developed	F6	ND
6	Stream 2	S2	3	0.6	Not developed	F8	ND
7	Lobnekha	Lo1	25.4	6.2	Reaches posterior margins of 4th abdominal segment	F0	F
8	Drechhu	Dr1	11	3.5	Touching first abdominal segment	F3	F

ND - Not able to determine.

Image 6. *Epiophlebia laidlawi* larva in Lobnekha stream (a) dorsal view and (b) ventral view (ovipositor encircled).

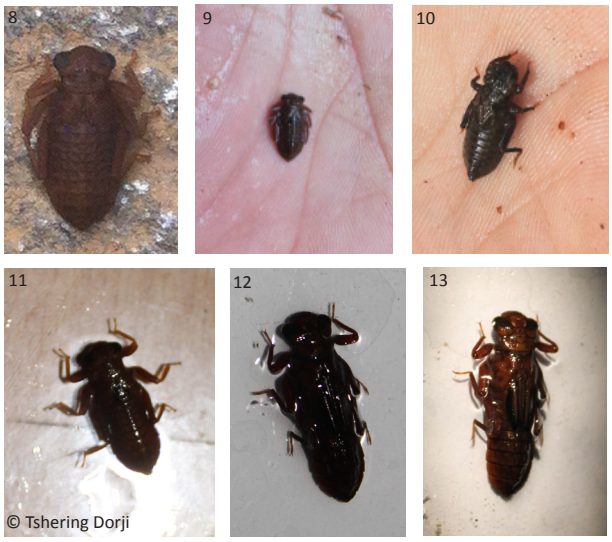

Image 7. *Epiophlebia laidlawi* larvae from (8) Drechhu (Wangsisina), (9) Dorokna (D2), (10) Jichulum (J2), (11) Nikachu (N1), (12) Nikachhu (N1), and (13) Nikachhu (N2).

and Jichulum streams add Punakha as the sixth district after Haa, Thimphu, Trongsa (Brockhaus & Hartmann, 2009), Chhukha and Bumthang (Image 8). It also adds Punatshangchhu basin as another river basin with *E. laidlawi* and fills the gap between Wangchhu basin and Drangmechhu basin.

The failure to record *E. laidlawi* larvae from streams located southward than the Lobnekha Stream in Chhukha could be either due to limited sampling effort as only one kick sample per stream was done or it could have been due to its restricted range within colder streams (Brockhaus & Hartmann 2009; Nesemann et al. 2011; Büsse et al. 2012; Carle 2012; Li et al. 2012). Streams and reaches within Toebirongchhu sub-watershed, along Phochhu and Mochhu, and Lamai Gonpa stream could be due to a relatively more disturbed habitat. Almost all streams and reaches from where the larvae were not recorded had high anthropogenic disturbance like abstraction of water for irrigation, dumping of waste material and reduced riparian vegetation. *E. laidlawi* is sensitive to human disturbance (Brockhaus & Hartmann 2009; Nesemann et al. 2011). This possibly explains the absence of the species from the above mentioned localities.

The larval development stage adds to the record of F0 from the whole *E. laidlawi* range and to the month of collection, thus adding to its poorly known biology (Brockhaus & Hartmann 2009; Nesemann et al. 2011). The F0 larvae were recorded from the March and October samples in previous studies (Brockhaus & Hartmann 2009), while the current study collected it in February 2014 from Lobnekha Stream, while the the one from Nikachhu reach N2 was collected in December 2012.

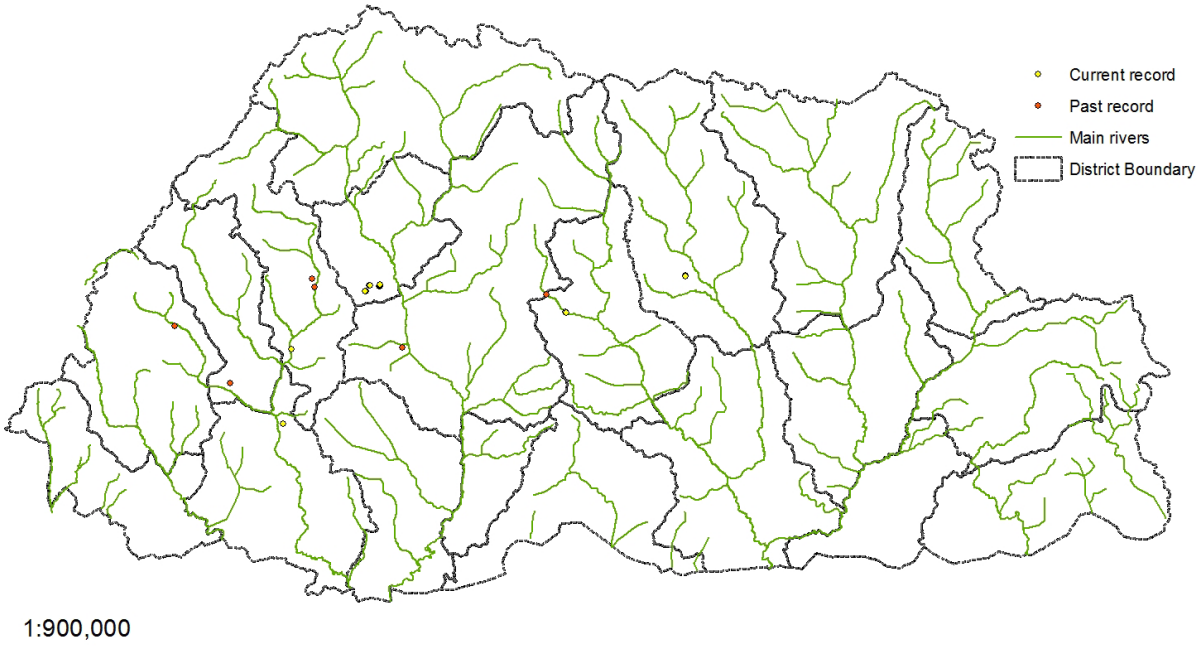

Image 8. Past and current record of *Epiophlebia laidlawi* larval presence in Bhutan.

This confirms the presence of F0 larva from October through, December and Februray till March. Besides, F3 and F8 are new instars described from Bhutan and adds to the list of larval stages described by Brockhaus & Hartmann (2009).

The current study recorded larvae from new localities within Bhutan with very limited sampling effort and seasons. This may highlight chances of its widespread distribution within Bhutan thereby making Bhutan a safe haven for its conservation. However, it is currently not being recognised as a species of conservation concern in Bhutan. Therefore, there is need for a systematic survey of *E. laidlawi's* distribution within Bhutan and also to sensitize concerned stakeholders on its conservation status.

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Appendix 1. Sampling reaches with district, stream name, sampling date, coordinates and altitude.

District	Stream name	Sampling reach	Sampling date	Latitude (N)	Longitude (E)	Altitude (m)
Bumthang	Stream 1	S1	11.V.2012	27.54417	90.72136	2922
	Stream 2	S2	11.V.2012	27.54133	90.72303	2916
	Lamai Gonpa	S3	11.V.2012	27.54531	90.73308	2786
		S4	11.V.2012	27.54750	90.74789	2628
Trongsa	Nikachhu	N1	01.XII.2012	27.44833	90.37397	2251
		N2	01.XII.2012	27.44875	90.37397	2177
		N3	02.XII.2012	27.43350	90.46317	1466
	Mangduechhu	M1	04.XII.2012	27.48281	90.49111	1818
		M2	02.XII.2012	27.43392	90.47750	1390
		M3	03.XII.2012	27.36689	90.53447	1014
		M4	03.XII.2012	27.36197	90.54800	990
		M5	03.XII.2012	27.35525	90.56114	977
Punakha	Dorokna*	D1	03.X.2013/10.III.2014	27.50478	89.79044	2067
		D2	03.X.2013/10.III.2014	27.50592	89.79008	2047
		D3	03.X.2013/10.III.2014	27.50678	89.79047	2042
		D4	03.X.2013/10.III.2014	27.50747	89.79075	2017
		D5	03.X.2013/10.III.2014	27.51486	89.79886	2000
		D6	03.X.2013/10.III.2014	27.50933	89.79333	1988
		D7	03.X.2013/10.III.2014	27.51928	89.80006	1956
		D8	/03.X.2013/10.III.2014	27.52103	89.80344	1889
	Jichulum*	J1	03.X.2013/10.III.2014	27.51731	89.83106	1653
		J2	03.X.2013/10.III.2014	27.51811	89.83186	1647
		J3	03.X.2013/10.III.2014	27.52081	89.83250	1602
		J4	03.X.2013/10.III.2014	27.52225	89.83264	1568
		J5	03.X.2013/10.III.2014	27.52428	89.83425	1527
		J6	03.X.2013/10.III.2014	27.52658	89.83503	1521
		J7	03.X.2013/10.III.2014	27.52981	89.83581	1485
		J8	03.X.2013/10.III.2014	27.53067	89.83536	1455
	Toebirongchhu*	T1	30.X.2013/10.III.2014	27.52897	89.86147	1283
		T2	30.X.2013/10.III.2014	27.52733	89.86181	1272
		T3	30.X.2013/10.III.2014	27.52697	89.86514	1267
		T4	30.X.2013/10.III.2014	27.52597	89.86642	1257
		T5	30.X.2013/10.III.2014	27.52519	89.86814	1262
		T6	30.X.2013/10.III.2014	27.52483	89.86917	1257
		T7	30.X.2013/10.III.2014	27.52894	89.87597	1192
		T8	30.X.2013/10.III.2014	27.52919	89.87783	1155
	Metshina	M1	20.X.2013/10.III.2014	27.50808	89.85478	1556
		M2	20.X.2013/10.III.2014	27.50967	89.85733	1522
		M3	20.X.2013/10.III.2014	27.51275	89.86186	1449
		M4	20.X.2013/10.III.2014	27.51392	89.86389	1416
		M5	20.X.2013/10.III.2014	27.52506	89.86900	1385
		M6	20.X.2013/10.III.2014	27.52697	89.87664	1329
		M7	20.X.2013/10.III.2014	27.51733	89.88353	1289
		M8	20.X.2013/10.III.2014	27.51797	89.88781	1260
	Changyul	C1	27.II.2014	27.59200	89.85831	1244
		C2	27.II.2014	27.59131	89.85700	1255
	Lhakhurongchhu	L1	27.II.2014	27.62481	89.82381	1311
		L2	27.II.2014	27.62567	89.82506	1315
	Tozhirongchhu	To1	27.II.2014	27.63442	89.80131	1330
		To2	27.II.2014	27.63247	89.80011	1365
		To3	27.II.2014	27.63011	89.79669	1395
	Dungkar rongchhu	Du1	27.II.2014	27.65683	89.77256	1395
		Du2	27.II.2014	27.65706	89.77114	1406
		Du3	27.II.2014	27.65719	89.77114	1433
	Shenarongchhu	S1	07.III.2014	27.59497	89.87397	1271
		S2	07.III.2014	27.59556	89.87656	1272
		S3	07.III.2014	27.59569	89.87831	1292
	Loreynarongchhu	L1	07.III.2014	27.64628	89.86731	1310
		L2	07.III.2014	27.64656	89.86697	1320
	Zeymarongchhu	Z1	07.III.2014	27.64692	89.87019	1311
		Z2	07.III.2014	27.64731	89.87169	1302
	Punadamchhu	P1	07.III.2014	27.65197	89.86783	1340
	Kebirongchhu	Ke1	07.III.2014	27.65486	89.87269	1321
		Ke2	07.III.2014	27.65439	89.87297	1316

Thimphu	Drechhu (Wangsisina)	Dr1	02.II.2014	27.35450	89.57431	2201
	Dagalarongchhu (Geneykha)	Da1	02.II.2014	27.65328	89.56508	2318
Chhukha	Lobnekha	Lo1	02.II.2014	27.16039	89.55092	2179
	Stream 1	CS1	02.II.2014	27.05928	89.55633	1670
	Takti	Ta1	02.II.2014	27.02050	89.56375	1924
	Tshatshilakha	Ts1	02.II.2014	26.99864	89.57567	2043
	Stream 2	CS2	06.II.2014	26.97133	89.55922	2025
	Gedu	Ge1	02.II.2014	26.95175	89.52717	1915
	Stream 3	CS3	02.II.2014	26.94944	89.52481	1845