

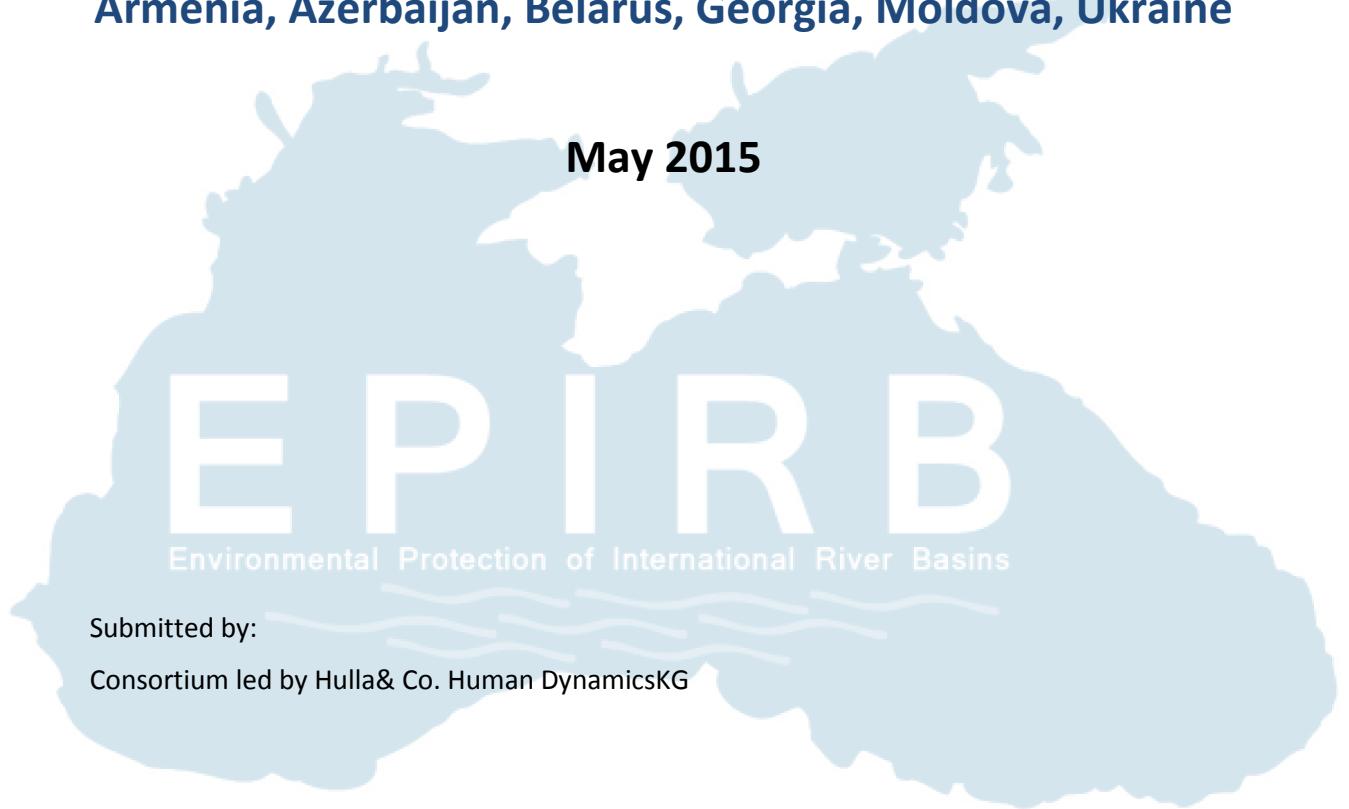


Environmental Protection of International River Basins Project

Contract No. ENPI/2011/279-666

JFS III
SURVEY DESIGN MANUAL:
BIOLOGICAL

Armenia, Azerbaijan, Belarus, Georgia, Moldova, Ukraine



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EPIRB
Environmental Protection of International River Basins

Submitted by:

Consortium led by Hulla& Co. Human DynamicsKG

ABBREVIATIONS

AA	Annual Average
AM	Armenia
AQEM	"The Development and Testing of an Integrated Assessment System for the Ecological Quality of Streams and Rivers throughout Europe using Benthic Macroinvertebrates" project
ASPT	Average Score Per Taxon
AZ	Azerbaijan
BBI	Belgian Biotic Index
BMWP	Biological Monitoring Working Party (Index)
BOD	Biochemical Oxygen Demand
BY	Belarus
CEN	European Committee for Standardization (European standard / European norm)
DO	Dissolved Oxygen
EC	European Community / European Commission
EEA	European Environmental Agency
EN	European Norm
EP	Ecological Potential
EPA	Environmental Protection Agency
EQR	Ecological Quality Ratios
ES	Ecological Status
EU	European Union
GE	Georgia
GIG	Geographic Intercalibration Group
H ₂ S	Hydrogen Sulfide
ISO	International Organization for Standardization (international standard)
ITC	Index of Trophic Completeness
MD	Moldova
MI	Macro-invertebrates
MZB	Macrozoobenthos
N	Nitrogen
N-NH ₄	Ammonium, as Nitrogen
N-NO ₂	Nitrite Nitrogen
N-NO ₃	Nitrate Nitrogen
O ₂	Oxygen (dissolved / oxygen saturation)
P	Phosphorus
QC/QA	Quality Control/Quality Assurance
RB	River Basin
RBA	Rapid Biological Assessment
RETI	Rhithron-feeding Type Index
RTI	Rhithron-Type-Index
STAR	Standardisation of River Classifications: Framework method for calibrating different biological survey results against ecological quality classifications to be developed for the Water Framework Directive (EU Project)
SWB	Surface Water Body
T°C	Temperature (degree Celsius)
TN	Total Nitrogen
TP	Total Phosphorus
UA	Ukraine
UK	United Kingdom
US	United States of America
WB	Water Body
WFD	Water Framework Directive

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JOINT FIELD SURVEY DESIGN

The Joint Field Survey design for 2015 has been conceived to put the focus on the sites definition to cover the gaps in the results achieved before 2015, the biological, hydromorphological and physico-chemical review of parameters and the use and the training on the updated protocols.

The objectives of the JFS are to get more data in order to increase the knowledge and the accuracy of the ecological status classification methodology with reference sites and all as a supportive technical tool include: assessment of ecological status, detection of change in surveillance, operational and investigative monitoring programmes, diagnosis of environmental stress, and assessment of both acute and chronic stressors (pollution control).

1. PARAMETERS

During the Joint Field Services in each of the 7 pilots, the teams will participate in the collection of the samples and in the measure of different parameters of different elements involved in the ecological status classification of waterbodies.

1.1 BIOLOGICAL ELEMENTS

As part of the ecological status assessment, macroinvertebrates have been selected to be the main group of the biological elements that is going to be sampled during JFS III.

In future stages of the project other biological elements could be included in the monitoring process.

The normative definitions of ecological status classes given in Annex V of WFD include five characters of river benthic invertebrate assemblages to be considered:

- (i) composition and
- (ii) abundance of taxa,
- (iii) the ratio of disturbance sensitive taxa to insensitive taxa,
- (iv) the level of diversity, and
- (v) occurrence of major taxonomic groups.

After the field works, in the laboratory the taxa will be identify till family or genus level to get the raw data for the next steps: calculating with ASTERICS different indexes.

1.2 PHYSICO-CHEMICAL ELEMENTS

1. GENERAL CONDITIONS (WATER)

To be determined on-site (water temperature, pH, dissolved oxygen, electrical conductivity) and in the laboratory (other parameters) at all JFS-III sites.

Please check the parameters listed below; I included the scheme stipulated for JFS-II.

Quality elements	Indicative parameters
<i>thermal conditions</i>	<i>Water temperature</i>
<i>oxygenation conditions</i>	<i>Dissolved Oxygen, (ppm & oxygen saturation)</i>
	<i>BOD5</i>
	<i>COD [according to the potassium dichromate method]</i>
<i>nutrient conditions</i>	<i>NO3</i>
	<i>NH4</i>
	<i>PO4 (orthophosphates)</i>
<i>salinity</i>	<i>Electrical Conductivity</i>
	<i>Cl</i>
	<i>SO4</i>
	<i>Total Dissolved Solids (total mineralization)</i>
	<i>Turbidity</i>
<i>acidification status</i>	<i>pH</i>
<i>Other elements</i>	<i>Total Suspended Solids</i>

2. SELECTED MICRPOLLUTANTS (WATER SAMPLES)

While referring to the JFS-II ‘nomenclatura’ below,

<i>specific relevant pollutants</i>	<i>As, Cd, Pb, Ni, Cu, Zn</i>
<i>Micro pollutants</i>	<i>Organic Chlorine Pesticides : Aldrin, Hexachlorobenzene, Endrin and pp DDT</i>

It is suggested the parameters analysis only at the following sites.

Please notice that Paul Buijs mentioned the names as mentioned in the Excel/Word files submitted by the countries (which were sometimes different in the JFS-I and the JFS-II reports).

Country	Pilot Basin	specific relevant pollutants (heavy metals)	Micro pollutants (selected organochlorine pesticides)
Azerbaijan	'Central Kura'	<ul style="list-style-type: none"> • Cadmium (Cd): <ul style="list-style-type: none"> ○ Gushchıçay River, Gushchu village; ○ Jogazchay River, Alpout village (Gazakh); ○ Qoşqarçay River, Khoshbulag village; ○ Tovuzchay River, Oysüzlü village. • Lead (Pb): <ul style="list-style-type: none"> ○ Qoşqarçay River, "Khoshbulag village"; ○ Qoşqarçay River, "Met-factory". 	none
Armenia	Akhuryan (incl. Metsamor)	none	none
Belarus	Upper Dnieper	none	none
Georgia	Chorokhi- Adjaristskali	As, Cd, Pb, Ni, Cu, Zn: <i>all</i> JFS-III sites	none
Moldova	Prut	<ul style="list-style-type: none"> Nickel (Ni) <ul style="list-style-type: none"> ○ r. Glodeanca-s. Duşmani; ○ r. Nîrnova-s. Ivanovca; ○ r. Sărata-s. Sărăteni aval; ○ r. Sărata-s. Vîlcele. 	<ul style="list-style-type: none"> 'DDT total' [= 4,4'-DDT (p,p'-DDT); 2,4'-DDT (o,p'-DDT); 4,4'-DDE (p,p'-DDE)] <ul style="list-style-type: none"> ○ r. Lăpuşna – Lăpuşna ○ r. Ilenuşa – Ilenuşa ○ r. Valea-Galmage - s. Zîrneşti Heptachlor: <ul style="list-style-type: none"> ○ r. Valea-Galmage - s. Zîrneşti
Ukraine	Prut	Copper (Cu) and Lead (Pb): <i>all</i> JFS-III sites	none
Ukraine	Upper Dnieper	Cadmium (Cd) and Lead (Pb): <i>all</i> JFS-III sites	aldrin, DDT, endrin, HCB: <i>all</i> JFS-III sites

3. SCREENING FOR PRIORITY SUBSTANCES (WATER AND SEDIMENT SAMPLES)

Please notice that the specification of both which Priority substances to be analyzed as well as the locations are still being reviewed.

Country	Pilot Basin	<i>Suggested locations</i>
Armenia	Akhuryan	Akhuryan – ‘somewhere downstream’.
		Metsamor – SW-20: Metsamor-Ranchpar
Azerbaijan	'Central Kura'	Gushchycay River – close before its confluence with the Kura River
		Qoşqarçay River – close before its confluence with the Kura River
Belarus	Upper Dnieper	Svisloch River – downstream discharge WWTP Minsk
		Berezina River – downstream Svetlogorsk
Georgia	Chorokhi- Adjaristskali	Chorokhi River – downstream confluence with Adjarkal
		(perhaps, Adjarkal River – close before its confluence with Choroki River)
		A site inside Batumi agglomeration, still to be determined
Moldova	Prut	<i>Note that between brackets the names of the routine monitoring sites of the State Hydrometeorological Service are mentioned</i>
		Prut River – near the border with Ukraine (r.Prut – s.Şirăuţi, 0,2 km în amonte (Lipcani));
		Prut River – downstream confluence of Jijia (r.Prut – s.Valea Mare, în aval de gura r.Jijia);
		Prut River – close before its confluence with the Danube River (r.Prut - s.Giurgiuleşti).
Ukraine	Prut	Prut River - downstream Chernivtsi (for example, the JFS-II site “Prut нижче м. Чернівці”);
		Prut River – close to the border with Moldova
Ukraine	Upper Dnieper	Desna River – close before its confluence with the Dnieper (I hope that you can specify the site more precisely for inclusion in the JFS-III programme);
		Dnieper River – downstream Kyiv

1.3 HYDROMORPHOLOGICAL ELEMENTS

The hydromorphological site protocol consists of 5 separate parts: Identification, channel parameters, riparian and floodplain features, catchment features and hydrological parameters. The first parameters are used to identify the site and the exact location within the catchment. Many of the parameters can be assessed from maps; the remaining should be assessed from other relevant sources. Individual map parameters should preferably be derived from maps having identical scales to ensure consistent parameter estimation. The surveyor, date of survey, and a photo or a sketch of the site is also included in the identification part of the protocol.

The protocol will be applied in all the locations proposed in JFS III.

2. PROTOCOLS AND INSTRUCTIONS

During the JFS III some new protocols and procedures will be used in the field works for the 7 pilot river basin, in the Annexes the Instructions for Macroinvertebrates and Hydromorphological are available.

2.1 MACROINVERTEBRATES PROTOCOL AND INSTRUCTION

For macroinvertebrates, a modified multi-habitat sampling method is proposed to be used. It is based on the techniques in accordance with AQEM/STAR methodology. The replicates will be sampled per monitoring location with sizes 30 x 30 cm (0,5 mm mesh size). The replicates will be selected after detailed habitat observation according to the percentage ratio of the main bottom substrata (pro-rata multi-habitat approach). Both kick-sampling and wash sampling will be made. Usually, it is a ratio 50:50 between kick-sampling and wash sampling for upper part of the river basin and 100 % kick-sampling for the downstream part (alluvial plain). The sampling area is 1 m². Samples should be fixed with the ethanol 80%, stored in the cooling box and delivered to the laboratory for sorting and identification.

In the instruction document there are clear definition of the fields included in the field protocol and the explanation of how to fulfil it.

3. SITES LOCATION

Chorokhi-Adjaritskali River basin

Number of Site	CODE:	River/Sampling site	Location	Type of waterbo die	GPSLongitude X (EAST)	GPSLatitude Y_(NORTH)
1	RF1	Boloko	Zeda Tkhilnari Village		41,639167	41,537111
2	RF2	Skurdidi	Skurdidi (exR3)		41.48.09.7	41.30.02.2
3	RF3	Akavreta	Namonastrevi Village			
4	RF4	Kinstrishi	below Kintrishi NP		41,961806	41,789194
5	RF5	Machakhela	Kokoleti (exR7)		41,801028	41,500556
6	RF6	Naghvarevistskali	Tskhemlisi Village		42.09.34.9	41.40.31.8
7	RF7	Adjaritskali	Adjaritskali		41,726398	41,542301
8	RF8	Adjaritskali	Tskhmoris Village		42.03.31.1	41.38.36.1
9	MS1	Adjaritskali	Shuakhevi town center	WB	42.11.06.2	41.37.27.1
10	MS2	Chorokhi*	Mouth	HMWB	41,596333	41,594833
11	MS3	Adjaritskali	Khulo	WB	42,318306	41,638694
12	MS4	Adjaritskali	Makhuntseti	HMWB	41,886444	41,583694
13	MS5	Chorokhi*	Kirnati	HMWB	41,711666	41,511708
14	MS6	Boloko	Mouth	WB	41,64559	41,579477
15	MS7	Korokitskali	Ortabatumi	WB	41,707222	41,650417
16	MS8	Chakvitskali	Mouth	WB	41,89526	41,690214
17	MS9	Bartskhana	Mouth	WB	41,673795	41,641753
18	MS10	Kinkhisha	Mouth	WB	41,796911	41,795891
19	2SW-08	Achkva	Kobuleti		41,799722	41,821722
20		Akhanundeli	Tsoniarisi			
21	SW6 JFSI	Skhalta	Tsablana Village		42,365002	41,576801
22		Makho	upstream Makho			
23		Dologani	upstream Dologani			
24		Urekhi or mouth of Mejiini				
25		Mouth of Sacchino				

Akhuryan River basin

Number of Site	CODE:	River/Sampling site	Location	Type of waterbo die	GPSLongitude X (EAST)	GPSLatitude Y_(NORTH)
1	SW1_JF SII	Yeghnajur	Garnarich Village before Lake Arpi	WB	43,60611111	41,08194444
2	SW15_J FSII	Artikdjur	above Artik	WB	43,96277778	40,61222222
3	SW10_J FSII	Akhuryan	above Gyumri City	WB	43,81177778	40,78572222
4	SW19_J FSII	Metsamor	Gai Village	WB	44,45944444	40,15944444
5	SW2_JFI I	Karmirjur	Shaghik		43,58416667	41,05916667
6	SW3_JFI I	Dzoraget	Zorakert		43,66305556	41,09722222
7	SW4_JFI I	Tavshut	Tavshut		43,52555556	41,089
8	SW5_JF SII	Lernajur	Lernagyugh		43,90916667	40,94361111
9	SW6_JF SII	Hartashen	Hartashen		43,92888889	41,00638889
10	SW7_JF SII	Akhuryan	Berdashen – Poqr Sepasar		43,66305556	41,06388889
11	SW8_JF SII	Akhuryan	Amasia		43,78944444	40,94944444
12	SW9_JF SII	Akhuryan	Krasar		43,81305556	41,02388889
13	SW11_J FSII	Akhuryan	Down to Gyumri		43,78527778	44,74194444
14	SW12_J FSII	Ashocq	Krasar		43,82083333	41,03027778
15	SW13_J FSII	Kety	Kety		43,84444444	40,86638889
16	SW14_J FSII	Jajur	Jajur		43,96277778	40,865
17	SW16_J FSII	Karkachun	Benyamin		43,83638889	40,69944444
18	SW17_J FSII	Jrarat	Karnut		43,95333333	40,76833333
19	SW18_J FSII	Selav Mastara	Qarakert		43,83055556	40,25638889
20	SW20_J FSII	Metsamor (Sevjur)	Ranchpar		44,36777778	40,03055556
21	MS	Armavir main canal middle		AWB		
22	MS	Armavir main canal end		AWB		
23	MS	Akhuryan main canal middle		AWB		
24	MS	Akhuryan main canal end		AWB		

Central Kura River basin

Number of Site	CODE:	River/Sampling site	Location	Water type	GPSLongitude X (EAST)	GPSLatitude Y_(NORTH)
1	MS	Jagazchay	Alpod	WB	45,20363889	41,07927778
2	MS	Akhinjachay	Qaralar	WB	45,48916667	40,72066667
3	MS	Ganjachay	Zurnabad	WB	46,24177778	40,49825
4	MS	Qoshkachay	Khoshbulaq	WB	46,03077778	40,43725
5	MS	Zayamchay	Yaniqli	WB	45,67241667	40,75713889
6	MS	Akhinjachay	Upper Tovuz	WB	45,57922222	40,94775
7	MS	Tovuzchay	Oysuzlu	WB	45,56983333	40,94405556
8	MS	Agstafachay	Below reservoir	WB	45,27172222	41,05197222
9	MS	Shamkirchay	Below highway bridge	WB	46,10930556	40,80988889
10	MS	Ganjachay	Topalhasanli	WB	46,29783333	40,56827778
11	MS	Agstafachay	Mollarjafirli	WB	45,42402778	41,15883333
12	MS	Tovuzchay	Alimardanli	WB	45,70686111	41,03908333
13	MS	Zayamchay	Khonogalo	WB	45,76227778	40,87680556
14	MS	Qoshkachay	Met-factory	WB	46,23694444	40,63752778
15	MS	Gedebedchay	Gedebed	WB	45,82505556	40,56758333
16	MS	Ganjachay	Below Goygol City	WB	40,16061111	46,64734333
17	MS	Ganjachay	Below Ganja City	HMWB	46,36588889	40,67569444
18	MS	Shamkir main canal	to be selected	AWB		
19	MS	Shamkir main canal	to be selected	AWB		
20	MS	Dallar canal	to be selected	AWB		
21	MS	Konullu canal	to be selected	AWB		
22	sw5 JFSI	<i>Qoshqarchay</i>	Yalqishlaq 65 m after road bridge			
23	WBR-05	Agstafachay	afther confluence Joghazchay			
24	WBR-08	Asrikchay	mouth of the river			

Dnieper (UA) River basin

Number of Site	CODE:	River/Sampling site	Location	Type of waterbodie	Latitude	Longitude
1	MS	Suhyi Vyr	Gribova Rudnya	WB	51,940361	31,086694
2	MS	Snov	Sedniv	WB	51,63	31,583972
3	MS	Zdvizh	Fenevich	WB	50,82025	30,102056
4	MS	Desna-Ulyanovka	Ulyanovka	WB	51,510306	31,438611
5	MS	Dnieper	Border BY/UA	WB		
6	MS	Desna-Puhovka	Puhovka	WB	50,607167	30,701139
7	MS	Teterev	Voropayevka	WB	50,979694	30,005944
8	MS	Pripyat	After confluence with Uzh	WB		
9	MS	Nemelnya	Border BY/UA		52,030222	31,003333
10	MS	Dnieper	Inflow to Kievskoe Reservoir	WB		
11	MS	Lubich	Lytki	WB	50,7245228	30,7502958
12	MS	Desna	Below Chernigov	WB	51,4669444	31,2202778
13	MS	Desna	Mouth	WB		
14	MS	Mesha	Rudnaya	WB	51,0635	30,842611
15	MS	Vzdvizha	Ivanovka	HMWB	51,388139	31,294972
16	MS	Belous	Koshivka	WB	51,551583	31,192167
17	MS	Irpen'	Dymydiv	HMWB	50,712111	30,338639
18	MS	Vorzna	Border BY/UA	WB	51,611333	30,595611
19	SW 30	Bilous	(1 km downstream of Chernigiv WWTP)	JFSII	51,4347222	31,2263889
20		Desna	arm of the Dnieper river	JFSII	50,5558333	30,5383333

Prut (MD) River basin

Number of Site	CODE:	River/Sampling site	Location	Type of waterbodie	Latitude	Longitude
1	MS	Prut	Criva	WB	48°15'35.39"	26°37'49.17"
2	MS	Prut	Pererita	WB	48°11'49.6"	26°54' 44.9"
3	MS	Prut	Giurgiulesti	WB	45°28'17,45"	28°11'51,26"
4	MS	Prut	Valea Mare Mare (below confluence with Jijia)	WB	47°06'29,52"	27°52/28,97
5	MS	Nirnova	Ivanovca	WB	46°53'15,3"	28°11'23,6"
6	MS	Camenca	Camenca	WB	47°49'30,6"	27°21'56,3"
7	MS	Vilia	Tetcani	HMWB	48°11'00,4"	026°57'45,2"
8	MS	Sarata	Sarateni	HMWB	46°35'42.3'	28°27'52.4'
9	MS	Lapusna	Lapusna	HMWB	46°53'27,3"	28°24'09,2"
10	MS	Larga-1	Chircani	HMWB	46°05'55,4"	28°11'3,1"
11	MS	Girla Mare	Petresti	HMWB	47°18'38.9"	27°42'35.6"
12	MS	Tigheci	Tigheci	HMWB	46°22'50,2"	28°22'27,8"
13	MS	Racovat	Below Gordinesti	HMWB	48°09'46,8"	027°10'09,7"
14	MS	Varsava	Valea Mare	HMWB	47°07'50,9"	027°51'49,3"
15		r. Ilenuța –	Ilenuța			
16		r. Valea-Galmage	- s. Zîrnești		46,0020833	28,1732778
17		Prut	Valea Mare Mare (before confluence with Jijia)	WB		
18	SW-9	Glodeanca	s.Dusmani	JFSII	47,7194722	27,48775
19	SW-7	Draghiste	s.Fetesti	JFSII	48,1543889	27,1120278
20	SW-1	Zeleonaia	Drepcauti	JFSII	48,2677778	26,7630278

Prut (UA) River basin

Number of Site	River/Sampling site	Location	Type of waterbodie	Latitude	Longitude
1	Yablunetskiy Prutets	Below Bukovel	WB	48,347778	24,492778
2	Prut	Turbaza Zaroslyak	WB	48,198333	24,58
3	Belyi Cheremosh	Yablunetska	WB	48,016611	24,912139
4	Ribnitsya	Yavory	WB	48,217028	24,942
5	Koroviya	Chagor	WB	48,22725	25,998139
6	Iltsya	Iltsi	WB	48,1535	24,755028
7	Ryngach	Hotyn-Novoselitsa	WB	48,278083	26,25975
8	Shybeny	Shybeny	WB	47,992083	24,6905
9	Prut	Luzhany	WB	48,342417	25,723722
10	Prut	Below Chernovtsy (Border UA/MD)	WB	48,2752778	26,0319444
11	Prutets Chemigivskiy	Mykulychyn	WB	48,405694	24,613833
12	Lopatynka	Shishkovtsy	WB	48,431056	27,149778
13	Prut	Below Kolomia	WB	48,4986111	25,0886111
14	Prut	Below Sniatyn	WB	48,3494444	24,5705556
15	Prut	Below Zabalotyv	WB	48,4575	25,3180556
16	Rakytna	Chernovtsy-Hotyn	WB	48,257972	26,233444
17	Beleluya	Toropivtsy	WB	48,556278	25,490917
18	Prut	UA0201/16	WB		
19	Zeleonaia	Border Ukraine and Moldova			
20	Sovycya	below Kitsman			

Dnieper (BY) River basin

Number of Site	CODE:	River/Sampling site	Location	Type of waterbodie	Latitude	Longitude
1	RC_MS	Volches	Ustye	WB	53,6188998	31,53870853
2	RC_MS	Ut	Tereshkovichi	WB	52,2749331	30,94602875
3	RC_MS	Greza	Vyazma	WB	53,3766833	29,94339062
4	RC_MS	Vabich	Barsuki	WB	53,9131167	29,82962622
5	RC_MS	Usha	Usha	WB	53,3968977	29,23250683
6	BY0108/04	Berezina	Vil. Uglaty	JFS II	52,897886	29,869439
7	BY0108/05	Berezina	Vil. Chirkovichi	JFS II	53,4202928	28,01644466
8	BY010803/01	Gayna	Vil. Sutoki	JFS II	53,9757397	28,82170667
9	BY010805/01	Plissa	t. Smolevichi	JFS II	52,680315	29,6613475
10	BY010805/02	Plissa	t. Borisov	JFS II	54,1883018	28,06452433
11	BY01101301/02	Zhadunka	Vil. Studenec	JFS II	53,5783492	30,85149483
12	BY011015	Uza	Vil. Bobovich (Gomel d-t)	JFS II	54,1878612	28,5610415
13	BY010814/01	Ola	Vil. Pleskovichi	JFS II	54,116189	27,59216417
14	BY011012	Lipa	Vil. Lipa	JFS II	52,8019343	29,69121033
15	BY01100803/01	Resta	Vil. Poteryaevka	JFS II	53,043917	29,59073017
16	BY0106/04	Drut	City Rogachev	JFS II	52,3900933	31,481601
17	BY010806/01	Rova	Vil. Semenkovichi	JFS II	52,3407738	30,924985
18	BY0106/03	Drut	City Belynichi	JFS II	54,1422107	28,05716133
19	BY0106/01	Vyacha	vil. Nelidovichi	JFS II	53,9783778	29,69854167
20	BY011014/02	Iput	City Gomel	JFS II	52,7327878	31,78370633
21		Svisloch	Osipovichi	MS		

4. ANNEXES

ANNEX 1: WATER QUALITY PHYSICO–CHEMICAL SAMPLING MANUAL

ANNEX 2: INSTRUCTION FOR HYDROMORPHOLOGICAL SITE PROTOCOL FOR THE JOINT FIELD SURVEYS

INSTRUCTION FOR HYDROMORPHOLOGICAL SITE PROTOCOL FOR THE JOINT FIELD SURVEYS

SAMPLING PROCEDURE FOR RIVER MACRO-INVERTEBRATES IN JOINT FIELD SURVEYS OF EPIRB PILOT RIVER BASINS

Short instruction on How to fill in the Site Protocol