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Reference individuals, blood collection, treatment of samples and descriptive data from the questionnaire in the Nordic Reference Interval Project 2000

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The rules for recruitment of reference individuals, inclusion and preparation of individuals, blood collection, treatment of samples (and control materials) and analysis at the 102 medical laboratories attending the Nordic Reference Interval Project (NORIP) are given as well as the rules for central exclusion of reference individuals. The individuals (18–91-year-olds) should be evenly distributed on age and gender groups. The 3002 reference individuals who contributed at least one reference value to the finally suggested reference intervals were characterized using the information in the questionnaire. Gender, age and country are the main entries in the tables. Other variables in the cross-tables or figure are height, weight, body mass index, ethnic origin, heredity for diabetes, chronic disease, oestrogens or oral contraceptives, other medication, hard physical activity, previous blood donations, smoking habits, use of alcohol, hours since last meal and time of blood collection (hour, day of week, month, year). The Danes had the highest alcohol consumption and the Icelanders had the highest body mass index. The information in this article may interest potential users of the Nordic Reference Interval Project bio-bank and database (NOBIDA) in which serum, Li-heparin plasma and EDTA buffy coat from the mentioned individuals are stored below −80°C.

Key words: bio-bank; blood; DNA; exclusion; inclusion; NOBIDA; plasma; preanalytical; recruitment; serum

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INTRODUCTION

In the Nordic Reference Interval Project (NORIP [1], see also http://www.furst.no/norip/) the main goal was to obtain reference values for clinical biochemical properties frequently measured in serum or plasma. The recruitment of reference individuals, inclusion of individuals, blood collection and analyses were carried out at each of 102 medical laboratories in the Nordic countries in accordance with a common protocol. Exclusions of individuals and data as well as calculations of reference intervals were performed centrally. In this paper we describe the common protocol in the "Material and Methods" section. In the "Results" section we try to characterize the reference individuals who finally contributed with at least one reference value to the project.

MATERIALS AND METHODS

In general, a common protocol was followed. The most important deviations between the countries were: In Sweden and Iceland the head of the local laboratory can identify the person from the project number. In the other countries the laboratories cannot identify the persons. In Sweden the questions about alcohol used the classes "not regularly, 1–3 daily measures and >3 daily measures instead of the corresponding weekly amounts used in the other countries.

Samples to prepare DNA were not taken in Iceland. The DNA question was addressed differently in the remaining countries. Below, we describe the common protocol for selection of reference individuals, preparation of individuals, collection of blood and handling and storage of specimens. The letter of information and the questionnaire are given as citations, but of course these documents were translated to the language of each country before use. In the standard operational procedure (SOP) for treatment of the samples in each country, the national words for "shall" or "must" or "have to" were often used. Below, we in general use words like "should" or "was to" to indicate what the SOP told the laboratories to do. We have no indications of deviations from the SOP.

Reference individuals

Recruitment. The rules were: "Reference individuals can be recruited from the laboratory personnel and their relatives or acquaintances, from new blood donors or those that seldom donate blood. Equal sex and age distribution should be sought according to the protocol".

The protocol (SOP) states that the 25–50 individuals recruited by each laboratory should be distributed evenly between 4 age groups: 18–30, 31–50, 51–70 and more than 70 years. The sex distribution in each age group should be even. The type (or way) of recruitment was not registered.

With small variations from country to country, the primarily selected reference individuals received the following information and the questionnaire in their own language at the appropriate time before the sampling. It should be made easy to refrain from coming if the required criteria were not met. The papers were completed with the assistance of a medical technologist or nurse at the time of blood sampling. Below "cpr.nr." denotes central person registration number in the national register of the authorities in each country.

Informed consent

"Dear Reference Individual

Results from laboratory investigations are used by doctors to diagnose disease. The results from several common laboratory investigations do not distinguish easily between healthy and diseased individuals, i.e. we do not know with certainty what is normal. This is why the Nordic hospital laboratories have started a joint project in order to establish reference ranges for common blood analyses in the Nordic countries.

If you are 18 years of age or older and are feeling healthy, you are kindly asked to participate in this project as a reference individual by:

- Donating a blood sample (about 50 mL which is about one-tenth of the usual amount donated by blood donors in blood banks).
- Giving permission to do the analyses performed in the hospital laboratory on your

anonymous sample including genotypes (DNA-types) of relevance for the measured concentrations.
- Giving permission to store the anonymous samples for later repetition of similar analyses by the laboratories.
- Answering the enclosed questionnaire.

The results will be made anonymous for everyone except you in the following way:

At the time of blood sampling you will be given a sample number that will be used for labelling the samples.

Information from the questionnaire as well as the analytical results will be tied to that sample number. Your name and "cpr.nr." will not be linked to the sample number (and we do not wish to have the "cpr.nr."). In that way you will be the only person that can link yourself to the sample number.

As we do not have your name or "cpr.nr." linked to analytical data, we cannot send you the laboratory results, even if we desired to do so.

The analytical results from all samples in the project will be accessible on the Internet identified by the sample number. Each participant can also contact the blood-sampling laboratory and ask for the results should he/she wish to do so. The sample number must then be used as means of identification. If you do not want anyone to know which sample you have donated, you should destroy your sample number.

Further information will be given by the doctor in charge: _____
Tel.: _____

The blood sampling is done by the usual sampling procedures from a vein in the arm. This is considered completely harmless. In a few instances you may get a blue mark around the point of puncture and slight discomfort that should disappear within a short time. It will be your own choice to participate and you may withdraw your participation until you leave the laboratory after blood sampling.

By signing this document you have accepted participation in the project.

_____

Signature of the participant

This form should be signed in duplicate, one copy for the participant and one for the laboratory.

To the medical technologist:

The sample number should not be put on the form that is retained at the laboratory."

Questionnaire

"*Questionnaire to participant*

Sample number _____ (to be filled out by the nurse)
You **cannot** participate in this investigation if:

- You are feeling unwell today.
- You have been an inpatient in a hospital or if you have been dangerously ill (according to your own judgement) during the past month.
- You have had more than 2 measures of alcohol in the last 24 hours (a measure is equal to 12 g alcohol, the equivalent of the amount of alcohol found in one pilsner beer or one glass of wine).
- You have given blood as a donor in the past 5 months.
- You are pregnant or are breastfeeding.
- You have taken prescribed drugs other than the P-pill during the past 2 weeks.
- If you are a smoker you must refrain from smoking one hour prior to the blood sampling.

Questions to be answered by the participant:
Age: _____ years Sex: ☐ female ☐ male
Height: _____ cm Weight _____ kg

For women: Date of 1st day of last menstrual period:

Day of month:_____ month:_____ year:_____
(Need not be answered if irrelevant)

Ethnic origin _____ (If you think your genetic origin is half Nordic or more, please write "Norden", otherwise write the country/ies which the major part of your genetic heritage originates from).
How many years have you lived in your present country of residence? _____

Do you normally smoke (mark with an x):
☐ 0 cigarettes/cigars/pipes per day?
☐ 1 – 5 cigarettes/cigars/pipes per day?
☐ More than 5 cigarettes/cigars/pipes per day?

Do you normally drink (mark with an x)
☐ 0 measures of alcohol per week?
☐ 1 – 21 measures of alcohol per week?
☐ More than 21 measures of alcohol per week?

Yes No
☐ ☐ Do any of your siblings or parents have diabetes or did they have when alive?
☐ ☐ Have you been diagnosed with a disease that requires continuous monitoring or treatment by a doctor or in a hospital? If yes, kindly specify the disease:
_____(disease)

Yes No
☐ ☐ Have you taken the P-pill or oestrogen preparations (female sex hormones) during the past month?
☐ ☐ During the past week, have you taken any medications (including iron tablets) other than the ones mentioned above? If yes to one or both of the above two questions, write the name(s) of the medication(s):
_____

Yes No
☐ ☐ Have you participated in strenuous sports during the past week (e.g. run more than 10 km in one go, trained in fighting sports, like karate, boxing or equivalent or been active in body building)?

To be filled out with the medical technologist (or nurse):

Number of hours from the last meal before blood sampling: _____ hours

To the medical technologist (or nurse):

Time of sampling:
Weekday 1 – 7 (Monday = 1 Sunday = 7): _____Day of month (1 – 31):_____
Month 1 – 12 (January = 1 December = 12): _____Time of the day (00.00 until 24.00):_____
Identification of the laboratory (Labquality number):_____

The sample number, which is used for identifying the samples taken from the reference individual who has filled this form, shall be written or labelled on the form and samples. The name of the individual or the "cpr. nr." **must not** be written on the form or any of the samples."

Inclusion and exclusion criteria

The rules for local inclusion are described above in the letter of consent and the questionnaire. A priori, the uniform criteria for central exclusion of all results from a person were: plasma- or serum-glucose $\geq$ 11.0 mmol/L or other analytical results that clearly indicate a disease. The rules for such exclusion of persons not related to glucose were decided later. These exclusions were made as follows: NORIP-results (R) from all persons were first divided by the mean (C) of the results of the common calibrator CAL (see later) measured in the same analytical series giving the ratio (R/C). Then the distribution (all results) of the natural logarithm to (R/C) was considered. The discrimination limits used in these distributions: median $\pm$ 3s, median $\pm$ 4s and median $\pm$ 5s were then calculated as follows: the median and s was calculated for the results in thawed serum. For the following properties they were calculated separately for each gender: Urate, aspartate transaminase (AST), alanine aminotransferase (ALT), creatine kinase (CK), gamma-glutamyltransferase (GT), HDL-cholesterol, creatinine, urea and albumin. For other properties, the median and s were calculated for the group of all persons. The used standard deviation s was calculated for all biochemical properties as $s = (\ln H - \ln L)/4$ where H and L were 0.975 and 0.025 fractiles in the (R/C) distribution. The ln (R/C) values were compared with these limits also when the analysed sample was fresh serum or fresh or thawed plasma.

The final exclusion rules for individuals were:

1. Glucose concentration $\geq$ 7.0 mmol/L in thawed serum from individuals, who had fasted 12 h or more. If results from fresh or thawed plasma were available for the person, these, too, should be $\geq$ 7.0 mmol/L.
2. Glucose concentration $\geq$ 11.1 mmol/L in thawed serum. If results from fresh or thawed plasma were available for the person, these, too, should be $\geq$ 11.1 mmol/L.

3. 5s3s*: Results (ln (R/C)) on thawed serum exceeding median ± 5s for one biochemical property and median ± 3 s for another biochemical property. If other materials have been analysed for the person, the rule should be fulfilled for the two considered properties in at least one of the additional materials.
4. 4s4s*: Results (ln (R/C)) on thawed serum exceeding median ± 4s for one biochemical property and median ± 4s for another biochemical property. If other materials have been analysed for the person, the rule should be fulfilled for the two considered properties in at least one of the additional materials.

*The 5s3s and 4s4s rules were not applied on low values of ALT, AST, CK, GT, ALP, LD (lactate dehydrogenase), creatininium, carbamide (urea), triglyceride and/or bilirubin

Payment

Individuals should not be paid for participation in the project but could be compensated for time lost from work or for transportation expenses.

Assurance

The reference persons should have the same rights as patients.

Ethics

The important issues are covered by the above information and questionnaire. The laboratories paid to the central organization to participate. The project was centrally supported by the NFKK (Scandinavian Society of Clinical Chemistry). Nunc delivered the cryotubes, free of charge. Local reagents for the analysis were in some cases sponsored by the supplier.

Ethics committees approved the project, with the following identifications:

Denmark: KF01-072/99.

Norway: Nordic reference intervals in clinical chemistry, approved 15.04.99 by the regional ethics committee for medical research, Health Region I ref. 125/99-99063.

Sweden: Nordic reference intervals in clinical chemistry, Multicentre Study with the following numbers in the local Swedish committees:

Lund: LU 73-00
Göteborg: Gbg M 020-00
Stockholm: KI 00 – 070
Linköping: Li 00-039
Umeå: Um 00-042
Uppsala: Ups 00-098
Örebro: Ör 144/00

Finland: Ministry of Health and Social Affairs, Medical Research Ethics Section : DNO 35/04/00.

Iceland: Nordic Reference Values.

Preparation of the reference individuals before collection of blood

More than half of the persons should have their samples taken in fasting state between 07.00 and 10.00 h.

On the day of investigation and before blood collection, the persons should have moved freely out of bed. Immediately before blood collection, they should sit for at least 15 min (a short walk from a waiting room could be discounted).

Samples

Sampling conditions and tubes. A technician or nurse should draw less than a total of 50 mL blood from an antecubital vein using the standard technique with the least possible use of a tourniquet.

The blood should be collected in plain tubes (without gel) for serum, in Li-heparin tubes (without gel) for plasma and in EDTA tubes for whole blood and buffy coat. The heparin and EDTA tubes should be turned 10 times when filled with blood. The laboratory could use their routine tubes if they fulfilled the above criteria.

It was allowed locally to arrange that samples collected in one laboratory could be analysed for some or all components in another laboratory (both the sampling laboratory and the analysing laboratory could be identified from the report for every sample and analysis).

Treatment of the primary tubes. The tubes should be kept in the dark when possible. The plain tubes with coagulating blood should be kept at room temperature until centrifugation after 30 min and before 1½ h from sampling.

The Li-heparin tubes should be kept at room temperature and centrifugation started within 15 min after sampling. The EDTA blood could be used for routine haematology as soon as possible and kept at room temperature and/or 4–8°C until production and freezing of the buffy coat within 24 h.

Centrifugation. The tubes were centrifuged for 10 min at more than $1500 \times g$ (e.g. more than 3000 rpm with a radius of 16 cm).

Aliquoting to secondary tubes. Serum should be aliquoted to secondary tubes within 2 h after sampling. Plasma should be administered to secondary tubes within 30 min after sampling. After centrifugation of the EDTA tubes the layer between plasma and red cells (about 1mL) should be pipetted to a secondary tube. The secondary tubes were 1mL Nunc cryotubes with screw stoppers and silicone seals, "star foot" (conical), Cat. No. 366656.

Freezing. The secondary tubes for frozen specimens each with 1 mL serum or plasma should be frozen at < -20°C within 4 h after sampling. Buffy coat should be frozen within 24 h after sampling. The tubes should be transferred to temperatures < -70°C within 1 month if still at the laboratory. Seven serum tubes, 1–2 plasma tubes and the buffy coat were delivered to the Nordic Reference Interval Project bio-bank and database (NOBIDA) bio-bank [2]. Additional frozen tubes could be used for local analysis.

Control or reference materials

The participating laboratories received the following liquid reference material in frozen form either from Labquality or the national quality assurance organization. The reference material should be kept frozen until analysed (< -20°C for one month or < -70°C).

- CAL: Pool of unmodified serum from healthy donors, with the concentration of 18 components determined by reference methods. [3].
- HIGH: Donor serum pool, concentrated by removing water by freezing [3].
- LOW: Donor serum pool, diluted from HIGH by adding one part of NaCl/$CaCl_2$ solution [3].
- P: Serum pool from donors on the P-pill [3].
- X: Donor serum pool prepared in the same way as CAL [3].

Thawing of person samples and control materials before analysis

On the day of the analysis, the frozen materials should be kept for at least 1 h at room temperature, in the dark. They should subsequently be turned 10 times. In the case of flocculation, the tube could be centrifuged.

All samples and control materials should be analysed within 4 h after thawing.

Fresh samples

The secondary tubes for plasma and serum to be analysed without freezing were chosen freely by the local laboratories. These specimens should be kept for less than 8 h at room temperature and for less than 48 h at about 4°C before analysis.

Analysis

All laboratories should analyse thawed serum from all reference individuals and thawed plasma from at least 10% of the individuals in one analytical series for each property to be analysed. In these series, the full amount of control serum should be used (CAL, X, P, High and Low); CAL was to be measured 10 times, placed evenly between samples in the analytical run. The other control material was to be measured 3 times, once at the beginning of the run, once in the centre and once at the end of each run.

In addition, laboratories could analyse fresh samples in other series including 10 replications of thawed control serum X evenly distributed in the run (or the full amount of control sera as described above).

The analytical series should be accepted by the normal control procedures of the laboratories.

Analytical methods

The laboratories should use their routine methods. However, only Vitros and "IFCC 37°C" methods should be used for enzymes. Total iron-binding capacity (TIBC) could be

measured using either immunochemical methods for transferrin or iron-binding methods.

Samples to bio-bank

As already mentioned, 7×1 mL serum, buffy coat from 5 mL EDTA-blood and 2×1 mL Li-heparin plasma from each reference person should be delivered to the NOBIDA bio-bank [2] to be stored below $-80^{\circ}C$ in the Nunc secondary tubes. The rules for the bio-bank including ethics are described elsewhere [2].

Data treatment

The tables and figures presented below originate from data in a Microsoft Excel spreadsheet file containing all centrally registered reference individuals with their questionnaire information and CAL-corrected reference values [1] from thawed serum as well as reference values from the haematological analysis (one row for each person). Pål Rustad created the raw version of this file from the central database. The analytical results in the file were all accepted as reference values (i.e. have passed the exclusion process). In the raw file, the rows for the centrally excluded whole persons were deleted as well as rows for persons who have not contributed any reference value (from any material) to the reference intervals. The statistical treatment was done in SPSS version 11.5 for Windows after importing the Excel file into this system. Help variables for quantifying or grouping of the information were created when needed. The probability "p" of the observed chi-square or chi-squares deviating more given independent classification by rows and columns in the cross-tables is given with three decimals in Tables I–IV and VI. The probability "p" for the observed F-ratio or those deviating more in a one-way analysis of variance (ANOVA) given common means for the quantitative properties in the countries is given with three decimals in Table V. The premises for these tests were not considered. The hypothesis of independence between the row- and column-classification criteria is rejected if the p-value for chi-square is <0.05. The hypothesis of a common mean for the Nordic countries is rejected when the p-value in an ANOVA is <0.05.

Other medication than oestrogens/oral contraceptives

Each person who stated that they had used "other medication" was allocated to one of 7 groups of indication or application and to one of the two groups (prescribed or not prescribed drugs) (Tables VII–IX). Those who stated that they had used other drugs without naming the drugs in an interpretable way or not at all could, however, not be allocated. If a person used several drugs, the drug used for allocation to one of the 7 groups was chosen according to the following order of priority of the drug group: 1, 2, 3, 4, 5, 6. If the same drug could be allocated to more than one group, the order of priority for allocation of the person was: 5, 1, 2, 3, 4, 6. Group 7 could contain prescribed drugs. If acetylsalicylic acid was used for prevention of thromboembolic disease or in daily amounts of about 150 mg or less, the person was allocated to group 7 (higher doses to group 4). Prescribed drugs had higher priority for allocation of the person than non-prescribed drugs. If the active substance in a drug necessitated a prescription in some preparations (high doses or high number of tablets in a package) but not in other preparations, then all preparations were treated as non-prescribed drugs. If a drug did not require a prescription in one of the countries Denmark, Finland, Norway or Sweden, then it was considered as a non-prescribed drug in all countries. According to these rules, many NSAIDs (non-steroid anti-inflammatory drugs) and paracetamol were considered as non-prescribed drugs. The rules could not classify all cases but were then used as guidelines. For the allocations, the information at the following web addresses for the national drug catalogues were used http://www.lk-online.dk/, http://www.tohtori.fi/laakeopas/, http://www.fass.se/LIF/home/index.jsp, http://www.felleskatalogen.no/.

RESULTS

Recruitment and inclusion

Other means of recruitment than those suggested were also used. At least one laboratory advertised in the local newspaper to recruit healthy elderly people. Other laboratories contacted clubs with supposed healthy elderly people. Contrary to the exclusion criteria, 86 individuals taking prescribed medicine were included; of these 35 were

TABLE I. Distribution of the 3002 reference individuals over countries and time of blood collection.

Time of blood collection	Category	Denmark		Finland		Iceland		Norway		Sweden		Total		P*
		n	%	n	%	n	%	n	%	n	%	n	%	
Year	1999	0	*0*	0	*0*	0	*0*	78	*9*	0	*0*	78	*3*	0.000
	2000	510	*88*	881	*100*	22	*26*	721	*87*	101	*16*	2235	*74*	
	2001	71	*12*	1	*0*	64	*74*	25	*3*	528	*84*	689	*23*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Month	Jan.	39	*7*	144	*16*	3	*3*	179	*22*	202	*32*	567	*19*	0.000
	Feb.	36	*6*	146	*17*	2	*2*	229	*28*	205	*33*	618	*21*	
	Mar.	90	*15*	52	*6*	16	*19*	93	*11*	29	*5*	280	*9*	
	Apr.	65	*11*	168	*19*	3	*3*	52	*6*	28	*4*	316	*11*	
	May.	94	*16*	266	*30*	25	*29*	89	*11*	56	*9*	530	*18*	
	Jun.	54	*9*	13	*1*	17	*20*	4	*0*	7	*1*	95	*3*	
	Jul.	17	*3*	0	*0*	2	*2*	0	*0*	3	*0*	22	*1*	
	Aug.	12	*2*	0	*0*	1	*1*	25	*3*	0	*0*	38	*1*	
	Sep.	96	*17*	23	*3*	0	*0*	5	*1*	0	*0*	124	*4*	
	Oct.	40	*7*	9	*1*	2	*2*	7	*1*	0	*0*	58	*2*	
	Nov.	38	*7*	61	*7*	9	*10*	73	*9*	0	*0*	181	*6*	
	Dec.	0	*0*	0	*0*	6	*7*	68	*8*	99	*16*	173	*6*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Day of week	Mon.	76	*13*	74	*8*	5	*6*	104	*13*	40	*6*	299	*10*	0.000
	Tues.	137	*24*	280	*32*	23	*27*	179	*22*	140	*22*	759	*25*	
	Wed.	149	*26*	294	*33*	10	*12*	202	*25*	161	*26*	816	*27*	
	Thurs.	104	*18*	217	*25*	25	*29*	207	*25*	160	*25*	713	*24*	
	Fri.	85	*15*	15	*2*	18	*21*	125	*15*	111	*18*	354	*12*	
	Sat.	27	*5*	0	*0*	1	*1*	4	*0*	14	*2*	46	*2*	
	Sun.	3	*1*	2	*0*	4	*5*	3	*0*	3	*0*	15	*0*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Time of day (h)	6–9	404	*70*	845	*96*	65	*76*	567	*69*	515	*82*	2396	*80*	0.000
	10–12	109	*19*	37	*4*	14	*16*	168	*20*	73	*12*	401	*13*	
	13–24	43	*7*	0	*0*	7	*8*	89	*11*	41	*7*	180	*6*	
	N/A	25	*4*	0	*0*	0	*0*	0	*0*	0	*0*	25	*1*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Hours since last meal (h)	0–3	203	*35*	1	*0*	4	*5*	230	*28*	117	*19*	555	*18*	0.000
	4–7	30	*5*	1	*0*	8	*9*	55	*7*	18	*3*	112	*4*	
	8–9	21	*4*	40	*5*	7	*8*	52	*6*	33	*5*	153	*5*	
	10–11	101	*17*	365	*41*	33	*38*	194	*24*	130	*21*	823	*27*	
	12–21	226	*39*	473	*54*	34	*40*	290	*35*	330	*52*	1353	*45*	
	N/A	0	*0*	2	*0*	0	*0*	3	*0*	1	*0*	6	*0*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	

*Probability for chi-square.

using "major drugs". Altogether, 3036 persons were included in the project.

Exclusion of individuals in the central database

Central exclusions on the basis of the questionnaire were not made.

Finally, all results from 28 individuals were centrally excluded from the original sample group based on their analytical results according to the rules described in the "Material and Methods" section. Results from 14 of these individuals exceeded the glucose limits. Other characteristics of the 28 persons were: 5 had chronic disease (3 had known diabetes, 1 had prostate disease and 1 had hypertension), 11 had chronic disease and/or other medicine than oral contraceptives or oestrogens. At least two individuals took prescribed medicine other than oral contraceptives or oestrogens (these two should not have been included).

For unknown reasons no analytical data were obtained from 4 persons included in the study. For 2 persons included in the project we have analytical data but no personal data from the questionnaire. The following tables and figure contain data from the $3036-28-4-2=3002$ persons who contributed personal data and at least one reference value to the final reference intervals.

TABLE II. Distribution of the 3002 reference individuals over gender and other characterizing factors.

		Females		Males		Total		
Factor*	Category	n	%	n	%	n	%	P**
Age group (years)	18–30	410	*26*	383	*27*	793	*26*	0.360
	31–50	514	*32*	414	*29*	928	*31*	
	51–70	434	*27*	398	*28*	832	*28*	
	>70	232	*15*	217	*15*	449	*15*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Country	Denmark	313	*20*	268	*19*	581	*19*	0.850
	Finland	454	*29*	428	*30*	882	*29*	
	Iceland	48	*3*	38	*3*	86	*3*	
	Norway	438	*28*	386	*27*	824	*27*	
	Sweden	337	*21*	292	*21*	629	*21*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Nordic origin	Yes	1547	*97*	1342	*95*	2889	*96*	0.003
	No	40	*3*	68	*5*	108	*4*	
	N/A	3	*0*	2	*0*	5	*0*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Heredity for diabetes	Yes	197	*12*	151	*11*	348	*12*	0.256
	No	1341	*84*	1221	*86*	2562	*85*	
	N/A	52	*3*	40	*3*	92	*3*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Chronic disease	Yes	78	*5*	79	*6*	157	*5*	0.289
	No	1510	*95*	1333	*94*	2843	*95*	
	N/A	2	*0*	0	*0*	2	*0*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Oestrogens or oral contraceptives	Yes	525	*33*	0	*0*	525	*17*	0.000
	No	1051	*66*	15	*1*	1066	*36*	
	N/A	14	*1*	1397	*99*	1411	*47*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Other medication	Yes	334	*21*	190	*13*	524	*17*	0.000
	No	1199	*75*	1169	*83*	2368	*79*	
	N/A	57	*4*	53	*4*	110	*4*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Hard physical activity	Yes	142	*9*	283	*20*	425	*14*	0.000
	No	1445	*91*	1125	*80*	2570	*86*	
	N/A	3	*0*	4	*0*	7	*0*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Blood donations	Yes	233	*15*	253	*18*	486	*16*	0.049
	No	1222	*77*	1049	*74*	2271	*76*	
	N/A	135	*8*	110	*8*	245	*8*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Tobacco (units/day)	0	1319	*83*	1156	*82*	2475	*82*	0.011
	1–5	92	*6*	86	*6*	178	*6*	
	>5	138	*9*	153	*11*	291	*10*	
	N/A	41	*3*	17	*1*	58	*2*	
	Total	1590	*100*	1412	*100*	3002	*100*	
Alcohol (units/week)	0	803	*51*	552	*39*	1355	*45*	0.000
	1–21	773	*49*	838	*59*	1611	*54*	
	>21	0	*0*	13	*1*	13	*0*	
	N/A	14	*1*	9	*1*	23	*1*	
	Total	1590	*100*	1412	*100*	3002	*100*	

For further explanations of the "factors" and categories, see the questionnaire.
**Probability for chi-square.

Tables characterizing blood collection and used reference individuals

Abbreviations used in the tables are: F = Females, M = Males, N/A = not answered, n = number of persons, p = probability. Country, gender and age are the main entries to Tables I–V for classifying persons according to the other information in the questionnaire. In Tables VI–IX chronic disease and use of medication other

TABLE III. Distribution of the 3002 reference individuals over age groups and other characterizing factors.

		18–30 y		31–50 y		51–70 y		>70 y		Total		
Factor	Category	n	%	n	%	n	%	n	%	n	%	P*
Country	Denmark	160	*20*	188	*20*	149	*18*	84	*19*	581	*19*	0.020
	Finland	221	*28*	263	*28*	258	*31*	140	*31*	882	*29*	
	Iceland	27	*3*	36	*4*	20	*2*	3	*1*	86	*3*	
	Norway	224	*28*	262	*28*	230	*28*	108	*24*	824	*27*	
	Sweden	161	*20*	179	*19*	175	*21*	114	*25*	629	*21*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Gender	F	410	52	514	55	434	52	232	52	1590	53	0.360
	M	383	48	414	45	398	48	217	48	1412	47	
	Total	793	100	928	100	832	100	449	100	3002	100	
Nordic origin	Yes	756	*95*	892	*96*	802	*96*	439	*98*	2889	*96*	0.138
	No	37	*5*	35	*4*	27	*3*	9	*2*	108	*4*	
	N/A	0	*0*	1	*0*	3	*0*	1	*0*	5	*0*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Heredity for diabetes	Yes	51	*6*	111	*12*	122	*15*	64	*14*	348	*12*	0.000
	No	712	*90*	779	*84*	689	*83*	382	*85*	2562	*85*	
	N/A	30	*4*	38	*4*	21	*3*	3	*1*	92	*3*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Chronic disease	Yes	19	*2*	27	*3*	40	*5*	71	*16*	157	*5*	0.000
	No	774	*98*	899	*97*	792	*95*	378	*84*	2843	*95*	
	N/A	0	*0*	2	*0*	0	*0*	0	*0*	2	*0*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Oestrogens or oral contraceptives	Yes	217	*27*	108	*12*	166	*20*	34	*8*	525	*17*	0.000
	No	194	*24*	405	*44*	271	*33*	196	*44*	1066	*36*	
	N/A	382	*48*	415	*45*	395	*47*	219	*49*	1411	*47*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Other medication	Yes	124	*16*	141	*15*	145	*17*	114	*25*	524	*17*	0.000
	No	640	*81*	742	*80*	658	*79*	328	*73*	2368	*79*	
	N/A	29	*4*	45	*5*	29	*3*	7	*2*	110	*4*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Hard physical activity	Yes	219	*28*	125	*13*	65	*8*	16	*4*	425	*14*	0.000
	No	573	*72*	798	*86*	766	*92*	433	*96*	2570	*86*	
	N/A	1	*0*	5	*1*	1	*0*	0	*0*	7	*0*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Blood donations	Yes	121	*15*	156	*17*	156	*19*	53	*12*	486	*16*	0.019
	No	604	*76*	688	*74*	612	*74*	367	*82*	2271	*76*	
	N/A	68	*9*	84	*9*	64	*8*	29	*6*	245	*8*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Tobacco (units/day)	0	632	*80*	773	*83*	676	*81*	394	*88*	2475	*82*	0.006
	1–5	55	*7*	56	*6*	44	*5*	23	*5*	178	*6*	
	>5	95	*12*	81	*9*	89	*11*	26	*6*	291	*10*	
	N/A	11	*1*	18	*2*	23	*3*	6	*1*	58	*2*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	
Alcohol (units/week)	0	361	*46*	354	*38*	369	*44*	271	*60*	1355	*45*	0.000
	1–21	424	*53*	565	*61*	451	*54*	171	*38*	1611	*54*	
	>21	2	*0*	2	*0*	6	*1*	3	*1*	13	*0*	
	N/A	6	*1*	7	*1*	6	*1*	4	*1*	23	*1*	
	Total	793	*100*	928	*100*	832	*100*	449	*100*	3002	*100*	

*Probability for chi-square.

than oestrogens and oral contraceptives are presented in more detail.

Age and body mass index

In Figure 1, a slightly increased body mass index (BMI) with age was found for all persons (linear regression: $BMI = 0.036 \times age + 22.4$, where the unit for BMI is in kg/m^2 and age is in years, SD slope = 0.0031, SD Y intercept = 0.16, n = 2991). Notice the presence of relatively few individuals of more than 80 years of age. For 11 persons, all below 72 years of age, the BMI could not be calculated.

TABLE IV. Distribution of the 3002 reference individuals over countries and other characterizing factors.

		Denmark		Finland		Iceland		Norway		Sweden		Total		
Factor	Category	n	%	N	%	n	%	n	%	n	%	n	%	P*
Age group (years)	18–30	160	*28*	221	*25*	27	*31*	224	*27*	161	*26*	793	*26*	0.020
	31–50	188	*32*	263	*30*	36	*42*	262	*32*	179	*28*	928	*31*	
	51–70	149	*26*	258	*29*	20	*23*	230	*28*	175	*28*	832	*28*	
	>70	84	*14*	140	*16*	3	*3*	108	*13*	114	*18*	449	*15*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Gender	F	313	*54*	454	*51*	48	*56*	438	*53*	337	*54*	1590	*53*	0.850
	M	268	*46*	428	*49*	38	*44*	386	*47*	292	*46*	1412	*47*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Nordic origin	Yes	565	*97*	871	*99*	86	*100*	789	*96*	578	*92*	2889	*96*	0.000
	No	16	*3*	9	*1*	0	*0*	32	*4*	51	*8*	108	*4*	
	N/A	0	*0*	2	*0*	0	*0*	3	*0*	0	*0*	5	*0*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Heredity for diabetes	Yes	46	*8*	128	*15*	0	*0*	82	*10*	92	*15*	348	*12*	0.000**
	No	535	*92*	754	*85*	0	*0*	738	*90*	535	*85*	2562	*85*	
	N/A	0	*0*	0	*0*	86	*100*	4	*0*	2	*0*	91	*3*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Chronic disease	Yes	28	*5*	65	*7*	4	*5*	34	*4*	26	*4*	157	*5*	0.080
	No	553	*95*	816	*93*	82	*95*	790	*96*	602	*96*	2843	*95*	
	N/A	0	*0*	1	*0*	0	*0*	0	*0*	1	*0*	2	*0*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Oestrogens or oral contraceptives	Yes	99	*17*	164	*19*	12	*14*	133	*16*	117	*19*	525	*17*	0.362***
	No	226	*39*	287	*33*	35	*41*	300	*36*	218	*35*	1066	*36*	
	N/A	256	*44*	431	*49*	39	*45*	391	*47*	294	*47*	1411	*47*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Other medication	Yes	68	*12*	306	*35*	6	*7*	80	*10*	64	*10*	524	*17*	0.000
	No	512	*88*	573	*65*	0	*0*	719	*87*	564	*90*	2368	*79*	
	N/A	1	*0*	3	*0*	80	*93*	25	*3*	1	*0*	110	*4*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Hard physical activity	Yes	63	*11*	169	*19*	9	*10*	98	*12*	86	*14*	425	*14*	0.0002
	No	516	*89*	712	*81*	77	*90*	723	*88*	543	*86*	2571	*86*	
	N/A	2	*0*	1	*0*	0	*0*	3	*0*	0	*0*	6	*0*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Blood donations	Yes	78	*13*	238	*27*	18	*21*	101	*12*	51	*8*	486	*16*	0.000
	No	430	*74*	595	*67*	68	*79*	601	*73*	578	*92*	2272	*76*	
	N/A	73	*13*	49	*6*	0	*0*	122	*15*	0	*0*	244	*8*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Tobacco (units/day)	0	431	*74*	748	*85*	76	*88*	650	*79*	571	*91*	2476	*82*	0.000
	1–5	44	*8*	54	*6*	4	*5*	55	*7*	21	*3*	178	*6*	
	>5	91	*16*	71	*8*	6	*7*	86	*10*	37	*6*	291	*10*	
	N/A	15	*3*	9	*1*	0	*0*	33	*4*	0	*0*	57	*2*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	
Alcohol (units/week)	0	128	*22*	325	*37*	17	*20*	273	*33*	613	*97*	1356	*45*	0.000
	1–21	435	*75*	547	*62*	68	*79*	545	*66*	16	*3*	1611	*54*	
	>21	7	*1*	4	*0*	0	*0*	2	*0*	0	*0*	13	*0*	
	N/A	11	*2*	6	*1*	1	*1*	4	*0*	0	*0*	22	*1*	
	Total	581	*100*	882	*100*	86	*100*	824	*100*	629	*100*	3002	*100*	

*Probability for chi-square.
**P = 0.000 also when Iceland and all N/A are deleted.
***P = 0.264 when males and N/A are excluded.

DISCUSSION

For many reasons (number of reference individuals, economic, practical, acceptance of the results and for wide geographical distribution of the reference individuals) all laboratories in the Nordic countries were invited to participate. The rules for local recruitment of reference individuals therefore had to be simple. Random recruitment in age- and gender-stratified groups

TABLE V. Height, weight and body mass index.

Property	Statistics	Denmark	Finland	Iceland	Norway	Sweden	Total	P*
Height (cm)	N	579	882	86	821	628	2996	
	Mean	172.8	170.5	172.3	172.7	171.9	171.9	0.000
	Median	172	170	171.5	172	171	172	
	Std. deviation	9.0	9.2	9.1	9.0	9.2	9.2	
	Minimum	150	143	147	152	151	143	
	Maximum	200	198	197	198	204	204	
	0.025 fractile	158	155	155	158	156	156	
	0.25 fractile	166	163	167	166	165	165	
	0.75 fractile	179	178	177	180	179	178	
	0.975 fractile	192	188	196	191	190	190	
Weight (kg)	N	581	880	86	821	628	2996	
	Mean	71.3	71.3	74.5	71.4	71.6	71.5	0.226
	Median	70.0	70.0	73.0	70.0	70.0	70	
	Std. deviation	12.4	12.4	13.6	12.1	11.9	12.3	
	Minimum	42	43	53	40	44	40	
	Maximum	109	118	125	115	115	125	
	0.025 fractile	50	51	55	50	51	51	
	0.25 fractile	62	62	64	62	63	62	
	0.75 fractile	80	80	84	80	80	80	
	0.95 fractile	97	98	112	96	99	98	
Body mass index (mass/height^2) (kg/m^2)	N	579	880	86	818	628	2991	
	Mean	23.8	24.4	25.1	23.8	24.2	24.1	0.000
	Median	23.6	24.2	24.8	23.7	23.8	23.9	
	Std. deviation	3.2	3.3	4.0	2.9	3.0	3.14	
	Minimum	16.4	17.0	18.8	16.5	15.6	15.6	
	Maximum	36.5	40.3	43.3	35.9	38.1	43.3	
	0.025 fractile	18.5	18.8	19.1	18.7	19.0	18.8	
	0.25 fractile	21.3	22.2	21.8	21.9	22.0	21.9	
	0.75 fractile	25.7	26.5	27.2	25.6	25.8	25.9	
	0.975 fractile	31.0	31.4	32.8	30.3	31.3	31.1	

*Probability in one-way analysis of variance.

of the whole population surrounding the laboratories would have been more appropriate [4]. This has recently been tried in Vejle county, Denmark [5]. In the Vejle project, 2100 persons were invited and 755 accepted the invitation to be further investigated for exclusion or inclusion, so even when properly tried, it is difficult to ensure representative reference individuals. Another Scandinavian project [6] used a pre-established age- and gender-stratified random population of reference individuals in Kristianstad county, Sweden, from which 350 persons were invited with a participation rate of 70%. Anyhow, although anybody who met the inclusion criteria in principle could participate in NORIP, it can be assumed that most of the included reference individuals had some personal connection to the local laboratory. This perhaps indicates that the "middle class" in the background population is over-represented. It is

TABLE VI. Chronic disease and other medications than oestrogens/oral contraceptives.

		Presence of chronic disease?								
		Yes		No		N/A		Total		
		N	%	N	%	N	%	N	%	P*
Other medication resent week	Yes	69	*44*	455	*16*	0	*0*	524	*17*	0.000
	No	85	*54*	2282	*80*	1	*50*	2368	*79*	
	N/A	3	*2*	106	*4*	1	*50*	110	*4*	
Total		157	*100*	2843	*100*	2	*100*	3002	*100*	

*Probability for chi-square.

TABLE VII. Distributions of drugs users by country and need of prescription.

	Denmark	Finland	Iceland	Norway	Sweden	Total
Drugs without prescription	55	254	3	64	47	423
Prescribed drugs	13	38	3	15	17	86
Total	68	292	6	79	64	509

Persons taking prescribed drugs other than oestrogens or oral contraceptives should not have been included. Prescribed drugs had higher priority for allocation of the person than non-prescribed drugs. If the active substance in a drug necessitated a prescription for some preparations (high doses or high number of tablet in a package) but not for other preparations, then all preparations were treated as non-prescribed drugs. If a drug did not demand a prescription in one of the countries Denmark, Finland, Norway or Sweden, then it was considered as a non-prescribed drug in all countries. According to these rules, many NSAIDs (non-steroid anti-inflammatory drugs) and paracetamol were considered as non-prescribed drugs.

TABLE VIII. Distribution of drugs users by country and group of indication or application.

	Denmark	Finland	Iceland	Norway	Sweden	Total
1. Major prescribed drugs (for heart, hypertension and metabolic disorders, etc.)	4	23	3	2	3	35
2. Anti-allergic drugs	1	9	0	6	5	21
3. Inhibitors of acid in stomach, etc.	4	0	1	1	2	8
4. Analgesics	44	49	1	49	39	182
5. Drugs for local application (eyes, skin, inhalation, etc.)	3	5	0	7	5	20
6. Food supplements inclusive "alternative medication"	6	192	0	5	3	206
7. Other	6	14	1	9	7	37
Total number of drug users	68	292	6	79	64	509

If a person used several drugs, the drug used for allocation of the person to one of the 7 groups was chosen according to the following order of priority of the drug group: 1, 2, 3, 4, 5, 6. If the same drug could allocate the person to more than one group, the order of priority for allocation of the person was: 5, 1, 2, 3, 4, 6. Group 7 could contain persons with prescribed drugs. Acetylsalicylic acid used to prevent thromboembolic disease allocated the person to group 7 (higher doses to group 4).

TABLE IX. Distributions of drug users by prescription and group of indication or application.

	Drugs without prescription	Prescribed drugs	Total
1. Major prescribed drugs (for heart, hypertension and metabolic disorders, etc.)	0	35	35
2. Anti-allergic drugs	15	6	21
3. Inhibitors of acid in stomach, etc.	3	5	8
4. Analgesics	175	7	182
5. Drugs for local application (eyes, skin, inhalation, etc.)	4	16	20
6. Food supplements inclusive "alternative medication"	205	1	206
7. Other	21	16	37
Total number of drug users	423	86	509

Prescribed drugs had higher priority for allocation of the person than non-prescribed drugs. If a person used several drugs, the drug used for allocation of the person to one of the 7 groups was chosen according to the following order of priority of the drug group: 1, 2, 3, 4, 5, 6. If the same drug could allocate the person to more than one group, the order of priority for allocations of the person was: 5, 1, 2, 3,4, 6. Acetylsalicylic acid used to prevent thromboembolic disease allocated the person to group 7 (higher doses to group 4).

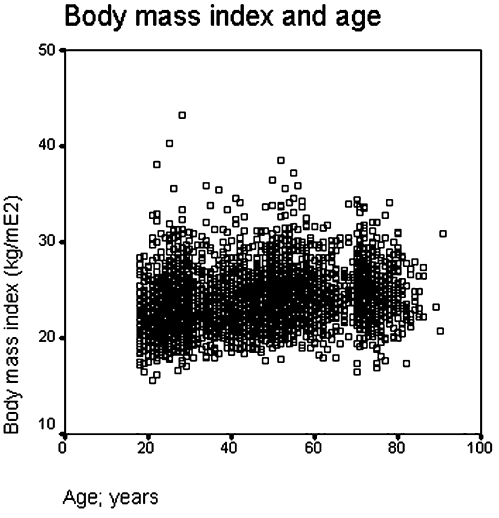

FIG. 1. Scatter plot for body mass index in kg/m^2 (ordinate) and age in years (abscissa) for all persons with both values (n = 2991). Linear regression: Y = 0.036X + 22.4.

fair to assume that this mechanism worked similarly in all countries and that the comparison of the included persons between countries will reflect differences and similarities between the countries and not between the different social classes included. Generally, it can be assumed that inclusion of individuals worked after the "first to the mill" principle in each age and gender class. Healthy elderly persons were difficult to recruit, but the laboratories tried and a reasonable number was reached.

The information in the questionnaire was not used for central exclusion of all results from a person. Such central "whole person-exclusions" were only done according to the already mentioned rules for abnormal analytical results. Individuals with diseases causing abnormal analytical results should be excluded. However, a single abnormal result could represent analytical error. Even if an abnormal result is true, it might not be connected to disease. The rules for exclusion were intended to take this into account. It should be remembered that single very abnormal results would be excluded as outliers in the corresponding distribution of reference values if relevant [7].

The information from the questionnaire (oestrogens, physical activity, non-fasting, etc.) in addition to age, gender and country were used to classify groups of reference individuals for certain purposes [7].

It would probably have been correct to exclude (centrally) those individuals who did not (according to the investigators) meet the inclusion criteria. We could also have excluded results for certain biochemical properties in certain individuals based on generally accepted effects of diseases or medication stated in the questionnaire upon these properties as described elsewhere [6]. However, no results or persons were centrally excluded because of drugs. Several reference intervals were calculated with and without results from persons with chronic disease or medication and the differences were marginal. The information is still available for further investigation.

The protocol for preparation of the subject and blood collection was inspired by the classic recommendations [8, 9].

It should be noticed that the reference intervals were meant for ambulatory patients with sampling taken in the sitting position and most of the values should and did come from fasting individuals in the morning.

The time schedule for treatment and analysis of the samples were made to emulate good and practical blood collection and handling of samples in the routine laboratory. The plasma was to be separated quickly (centrifugation within 15 min) from the cells to give reliable values for glucose (this is also a problem in the routine when central laboratory analysis is used). Obviously, routine samples collected in other places and brought or mailed to the laboratory are treated differently than the samples in NORIP.

The obligatory default procedure with frozen serum samples was chosen as the most practical and economic way to get many comparable results with analytical traceability to high metrological levels. The design with local analysis demanded a common calibrator and controls to be analysed in the same series as the samples. The calibrator (and controls) could only be reliably and commutably handled in the liquid frozen state. The full battery of calibrator and controls was expensive and should be used shortly after thawing. This battery could therefore not be used day after day for small amounts of fresh samples. Organizing everything (sampling and analysis for all persons) to be done on one day was not considered possible for all laboratories. Therefore

the samples could be gathered in the freezer together with the calibrator and controls and thawed and analysed simultaneously on the day of analysis. This default procedure would give reliable and traceable values for frozen samples. Using the calibrator X, the design allowed for further analysis on fresh samples, and in the project it could be shown that the values for frozen sera could be used as reference values for fresh sera [7].

The data from the questionnaires showed the expected similar gender and age distribution in all countries. This means that results for other properties for all persons could reveal differences between countries without correction for age and gender.

Danes buy much more alcohol than other Nordic peoples. Thus in the year 2000 the sales of alcohol equivalent to litres of pure alcohol per person aged 15 years or over were: Denmark: 11.5; Finland: 8.6; Iceland: 6.1; Norway: 5.6; and Sweden: 6.2 [10]. The Danes also admit this. In Table III, Iceland and Denmark have the lowest proportion of non-drinkers. Because of their low number, the reference individuals from Iceland may not be representative. The values for Denmark, Finland and Norway fit well with the corresponding mentioned sales. The Swedish proportion of non-drinkers in Table III is, however, probably too good to be true or at least not representative for the whole population [10]. It is most probably due to the way the question was presented in Sweden (as mentioned, the questions about alcohol in Sweden were changed to "not regularly", 1 – 3 daily measures and >3 daily measures, instead of the corresponding weekly amounts). The Danish way of life may have influenced, for example, alanine transaminase and gamma-glutamyltransferase values [11]. In relative terms, the Swedish group contained the highest number of non-smokers (consistent with [10]). Sweden also had relatively the most persons of non-Nordic ethnic origin. The Icelanders had the highest BMI. The Finnish had the lowest height and were the most physically active and made the most frequent use of medication. However, it is obvious that vitamins and other food supplements have been registered in the questionnaire in Finland far more often than in other countries. Whether this is due to different use or different questioning is not known.

The detailed characterization given here of the persons and samples may interest potential users of the NOBIDA bio-bank [2] in which the samples are stored below -80°C.

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