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Reference Population and Marathon Runner Sera Assessed by Highly Sensitive Cardiac Troponin T and Commercial Cardiac Troponin T and I Assays

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BACKGROUND: Endurance exercise can increase cardiac troponin (cTn) concentrations as high as those seen in cases of minor myocardial infarction. The inability of most cTn assays to reliably quantify cTn at very low concentrations complicates a thorough data analysis, and the clinical implications of such increases remain unclear. The application of recently developed highly sensitive cTn immunoassays may help resolve these problems.

METHODS: We evaluated the precommercial highly sensitive cardiac troponin T (hs-cTnT) assay from Roche Diagnostics and the Architect cardiac troponin I (cTnI-Architect) assay from Abbott Diagnostics by testing samples from a reference population of 546 individuals and a cohort of 85 marathon runners. We also measured the samples with the current commercial cTnT assay for comparison.

RESULTS: Although the hs-cTnT and cTnI-Architect assays were capable of measuring cTn concentrations at low concentrations (<0.01 μg/L), only the hs-cTnT assay demonstrated a CV of <10% at the 99th percentile of the reference population and a near-gaussian distribution of the measurements. After a marathon, 86% of the runners had cTnT concentrations greater than the 99th percentile with the hs-cTnT assay, whereas only 45% of the runners showed increased concentrations with the current cTnT assay. cTn concentrations remained significantly increased the day after the marathon. A multiple regression analysis demonstrated marathon experience and age to be significant predictors of postmarathon cTn concentrations ($P < 0.05$).

CONCLUSIONS: The hs-cTnT assay was the only assay tested with a performance capability sufficient to detect cTn concentrations in healthy individuals. The number of runners with increased cTn concentrations after a marathon depends highly on an assay's limit of detection (LOD). The assay with the lowest LOD, the hs-cTnT assay, showed that almost all runners had increased cTn concentrations. The clinical implications of these findings require further investigation.

Regular exercise is part of a healthy lifestyle and aids in the prevention of cardiovascular disease *(1)*. In endurance exercise such as marathon running, however, physical collapse is frequently observed during and after races, and such collapses are often associated with coronary artery disease or left ventricular hypertrophy *(1–3)*. The risk for such a cardiac event has been suggested to be comparable with that encountered in other daily activities and thus seems relatively low *(2–4)*. Nevertheless, the concentrations of highly specific cardiac markers such as the cardiac troponins (cTn)[3] are known to increase after prolonged exercise to concentrations similar to those seen after a minor myocardial infarction *(5–9)*, as we have recently reviewed for cardiac troponin T (cTnT) *(10)*. Because the consequences of cTn release are still unclear, the phenomenon of exercise-induced cTn release is an active topic of discussion and requires further study.

The recent development of more sensitive cTnT and cTnI immunoassays and their evaluation in different clinical settings *(11–13)* have prompted a redefinition of the diagnosis of acute myocardial infarction (AMI) as follows: an increase and/or decrease in the

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[3] Nonstandard abbreviations: cTn, cardiac troponin; cTnT, cardiac troponin T; cTnI, cardiac troponin I; AMI, acute myocardial infarction; LOD, limit of detection; hs-cTnT, highly sensitive cTnT (assay) from Roche Diagnostics; cTnI-Architect, Architect cardiac troponin I (assay) from Abbott Diagnostics.

concentrations of cardiac markers, preferably cTnT or cTnI, should be documented by at least one observation above the 99th percentile value of the reference population, and such results should be accompanied by clinical, electrocardiographic, or imaging findings *(14, 15)*. Until recently, however, most cTn assays lacked an analytical performance sufficient to detect cTn concentrations in a reference population or to distinguish reference values from the analytical noise. The inadequacy of cTn assays can be attributed either to the limit of detection (LOD) of the cTn assay being higher than reference values or to assay imprecision (i.e., CV) being >10% at the 99th-percentile value of the reference population *(16–18)*.

Because of the wide variation in cTn assays, comparisons of previous studies of exercise-induced cTn release make sense only for studies that have used the same cTn immunoassay. Such comparisons are especially difficult for cTnI studies because, in contrast to the patented cTnT assay, the approximately 10–20 cTnI immunoassays that have been developed use different antibodies directed against different epitopes *(14, 16, 17, 19)*. In addition, the various assays use different calibrator and control materials. We previously demonstrated that the cTnT concentration increases after prolonged exercise by 59% on average *(10)*. In brief, exercise-induced cTn release is characterized by a peak after the event is finished and a return to baseline concentrations within 1 day *(10)*. In the presence of clinical symptoms, exercise-induced cTn release would be indicative of AMI and would require further investigation. The use of recently developed highly sensitive cTn assays may provide new insights into the exercise-induced release of cTn.

We studied the analytical performance of 2 recently introduced cTn assays, the precommercial highly sensitive cTnT (hs-cTnT) assay from Roche Diagnostics and the cTnI Architect (cTnI-Architect) assay from Abbott Diagnostics. cTn concentrations were investigated both in a reference population and in a cohort of marathon runners. We included the current commercially available cTnT assay (fourth generation) in the study for comparison.

Materials and Methods

The reference population consisted of 546 apparently healthy persons from a health-check program at our hospital, and all of the individuals provided informed consent. To rule out individuals with cardiac syndromes, we included individuals in the study only when the following cardiac biomarker concentrations were all available: creatine isoenzyme MB, N-terminal pro–B-type natriuretic peptide, cTnT measured with the hs-cTnT assay, and cTnI measured with the cTnI-Architect assay (details of the cTn measurements are described below). Consequently, we excluded 45 individuals from the reference population. We also excluded 22 individuals because the concentration of one of these 4 biomarkers exceeded the mean + 3 SDs: creatine kinase isoenzyme MB mass (male cutoff, >10 μg/L; female cutoff, >7.9 μg/L; maximum, 13.37 μg/L) in 10 individuals, N-terminal pro–B-type natriuretic peptide concentration (cutoff, >41 pmol/L; maximum, 166 pmol/L) in 8 individuals, and cTn concentration (cTnT maximum, 0.134 μg/L; cTnI maximum, 0.217 μg/L) in 4 individuals.

Of the 836 runners who participated in the 2007 Maas Marathon (42.2 km), 85 runners were enrolled in the present study. This study was approved by the ethics committee (University Hospital Maastricht, The Netherlands), and all participants signed informed-consent forms. The maximum temperature on the day of the marathon was 23.4 °C with a south wind <14 m/s. We collected serum samples 0–2 h before the race, <1 h after the race, and on the day after the race in a subgroup of 23 runners whom we selected for logistical reasons. The samples were clotted and centrifuged, and the serum samples were stored at −80 °C until analysis. cTnI was measured with the Architect i2000SR (Abbott Diagnostics), with the LOD of 0.009 μg/L, a CV of ≤10% at 0.032 μg/L, and the 99th-percentile cutoff of 0.012 μg/L provided by the manufacturer. We measured cTnT on the Elecsys 2010 instrument (Roche Diagnostics) with the current commercially available cTnT immunoassay (fourth generation), with an LOD of 0.01 μg/L, a CV ≤10% at 0.03 μg/L, and a 99th-percentile cutoff at 0.01 μg/L. cTnT was also measured with the precommercial hs-cTnT assay. Complete validation of the hs-cTnT assay (same lot number) was performed in the research and development department of Roche Diagnostics. Intraassay CVs were 5.7% and 0.5% at 0.022 μg/L and 2.98 μg/L, respectively; interassay CVs were 3.0% and 1.4% at 0.021 μg/L and 3.03 μg/L, respectively. The linearity of the hs-cTnT assay was evaluated by serial dilution, from 1 part serum plus 9 parts diluent to 9 parts serum plus 1 part diluent (initial cTnT concentration in serum, 9.5 μg/L; Diluent Universal, Roche Diagnostics). The measured cTnT concentrations deviated from the expected concentrations by factors of 0.99 to 1.05. A comparison of the current cTnT assay and the precommercial hs-cTnT assay (cTnT up to 8 μg/L, n = 160) yielded the following regression equation: $y = 0.996x + 0.003$ μg/L, where x represents results obtained for the current commercial cTnT assay and y represents results for the hs-cTnT assay [$r = 0.9926$; 95th percentile of the residual distribution from the median = 0.331 (Passing–Bablok regression analysis)]. We also measured

creatine kinase and albumin concentrations with the Synchron LX 20 instrument (Beckman Coulter).

Data were analyzed with the Statistical Package for Social Sciences, Version 13.0 for Windows (SPSS). A nonparametric approach was used to calculate the upper reference limits (97.5th and 99th percentiles), and the Kolmogorov–Smirnov test was used to evaluate whether biomarker data deviated from a gaussian distribution. For variables with a gaussian distribution, we used the paired-samples *t*-test to evaluate differences between pre- and postexercise samples and used the independent-samples *t*-test to test sex differences. Multiple regression analysis was used to evaluate possible associations of sex, age, body mass index, and experience (i.e., number of prior completed marathons) with postmarathon cTn concentration. We log-transformed the data with nongaussian distributions and analyzed the data as described above if the transformed data approximated a gaussian distribution. For variables with a nongaussian distribution, we analyzed the original data with the nonparametric Wilcoxon signed rank test and the Mann–Whitney *U*-test. For statistical calculations, cTnT concentrations less than the LOD were set equal to the LOD. Unless otherwise stated, the threshold for statistical significance was set at a *P* level of 0.05.

Results

We used the NCCLS (now CLSI) EP5 guideline to establish precision profiles for the hs-cTnT and cTnI-Architect assays. These results are shown in the Data Supplement that accompanies the online version of this article at http://www.clinchem.org/content/vol55/issue1. The assay profiles produced 10%-CV cutoff concentrations at 0.009 μg/L and 0.032 μg/L, respectively. We used 10 measurements of cTn-negative serum (mean + 3 SDs) to establish LODs for the hs-cTnT and cTnI-Architect assays. The LOD was <0.001 μg/L for the hs-cTnT assay and 0.009 μg/L for the cTnI-Architect assay.

Fig. 1 shows at the left the cTn concentrations obtained for the reference population. With the cTnI-Architect assay (c), almost all measurements (97%) were below the LOD. In contrast, the hs-cTnT assay (b) yielded measurable concentrations for most of the samples in the reference population, indicated by the symmetrical box plot in Fig. 1. For the cTnI-Architect assay, the 99th-percentile value (0.013 μg/L) was less than the 10%-CV value. In contrast, the hs-cTnT assay had a CV of <10% at the 99th-percentile value (0.016 μg/L). The current commercially available cTnT assay (a; fourth generation) produced values that were all below the LOD (<0.01 μg/L). Finally, analysis of the

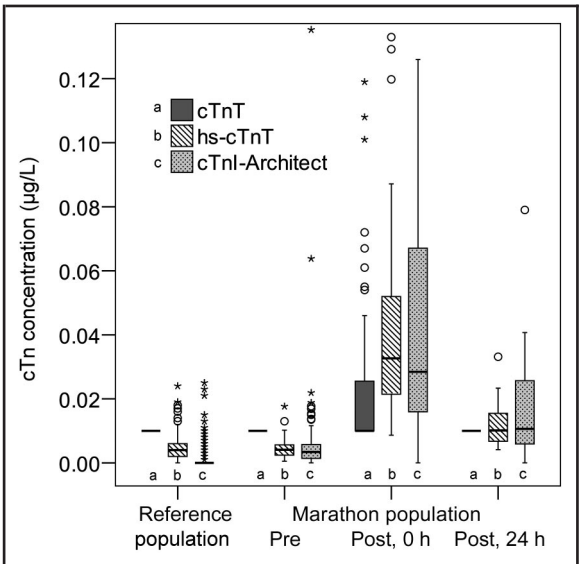

Fig. 1. cTn concentrations in the reference and marathon study populations as measured with the current commercially available cTnT assay, the hs-cTnT assay, and the cTnI-Architect assay.

Boxes represent the interquartile range (IQR), whiskers represent 1.5× the IQR values, and horizontal lines represent medians. Extreme values (○) represent values between 1.5× IQR and 3× IQR; outliers (∗) represent values >3× IQR.

hs-cTnT data revealed cTn reference values that were higher for males than for females ($P < 0.001$; Table 1).

Table 2 summarizes the baseline characteristics of the marathon population (85 runners). These data are comparable with those of the total marathon population of 836 runners (88% men; mean age, 45 years; mean running time, 3.76 h). The participants in our study seem to be highly experienced runners. Forty-four percent had previously completed 1–10 marathons, and 36% had completed >10 marathons.

Fig. 1 also shows that only the hs-cTnT and cTnI-Architect assays were able to measure prerace cTn concentrations. All prerace concentrations obtained with the current commercially available cTnT assay were below the assay's LOD. Table 3 shows that prerace concentrations obtained with the hs-cTnT assay were within the reference interval ($P = 0.282$). Prerace concentrations were significantly higher than the reference values ($P < 0.001$) when the cTnI-Architect assay was used; however, preexercise concentrations obtained with the cTnI-Architect assay should be considered with care, because 82% were below the LOD of the assay (<0.009 μg/L).

Immediately after the marathon, all runners in the study showed an approximately 10-fold increase in

Table 1. cTn reference values as measured with the hs-cTnT and cTnI-Architect assays.[a]

Study participants	hs-cTnT assay, μg/L	cTnI-Architect assay, μg/L[b]
Total (n = 479)		
Mean	0.004	0.001
Median	0.004	<0.001
97.5th Percentile	0.011	0.008
99th Percentile	0.016	0.013
Females (n = 215)		
Mean	0.003	0.001
Median	0.003	<0.001
97.5th Percentile	0.007	0.007
99th Percentile	0.008	0.012
Males (n = 264)		
Mean	0.005	0.001
Median	0.005	<0.001
97.5th Percentile	0.014	0.008
99th Percentile	0.018	0.013

[a] Data for both assays were not normally distributed ($P < 0.001$), including after log transformation. Differences between males and females in mean cTn concentration: hs-cTnT assay, $P < 0.001$; cTnI-Architect assay, $P = 0.788$.
[b] Ninety-seven percent of the measurements were below the LOD (0.009 μg/L).

cTnT and cTnI concentrations in the hs-cTnT and cTnI-Architect assays (Table 3). Albumin concentrations increased only slightly after the marathon; hence, we did not correct cTn concentrations for the effect of dehydration. The hs-cTnT assay showed that the cTnT concentration had increased to above the 99th percentile (0.016 μg/L) in almost all of the runners (86%). In contrast, only about half of the runners (45%) were above the 99th-percentile value (0.01 μg/L) when the current commercially available cTnT assay was used. Results obtained with the hs-cTnT assay were highly correlated with values obtained with the current cTnT assay (Spearman rank correlation coefficient = 0.955, for values above the 10%-CV cutoff concentration; $P < 0.001$). The cTnI-Architect assay yielded cTnI concentrations that were increased to greater than the 99th percentile in 81% of the runners. This percentage appeared comparable to that of the hs-cTnT assay; however, the 10%-CV cutoff (0.032 μg/L) was exceeded in only 47% of the runners when the cTnI-Architect assay was used. For the hs-cTnT assay, nearly all of the post-exercise concentrations (98%) were greater than the 10%-CV concentration (0.009 μg/L).

Multiple regression analysis (see Table S1 in the online Data Supplement) revealed running experience (the number of previously completed marathons) and age to be significant predictors of postmarathon cTn concentration as detected with the hs-cTnT assay (experience, $P = 0.005$; age, $P = 0.017$) and the cTnI-Architect assay (experience, $P = 0.001$; age, $P = 0.008$). Indeed, a comparison of the 2 outer quartiles of these cTn results showed that the runners with the lowest cTn concentrations had significantly more experience in marathon running than those with the highest cTn concentrations (hs-cTnT assay, $P = 0.005$; cTnI-Architect assay, $P = 0.036$). With respect to age, we found no significant difference between the 2 outer cTn quartiles for either the hs-cTnT assay ($P = 0.254$) or the cTnI-Architect assay ($P = 0.689$). We also noted no significant interaction between experience and age in the regression model (hs-cTnT assay, $P = 0.542$; cTnI-Architect assay, $P = 0.696$). Furthermore, sex and body

Table 2. Baseline characteristics of the reference and marathon study populations.

	Age, years[a]	Weight, kg[a]	Height, m[a]	Marathons completed, n[b]	Running time, h[a]
Reference population					
Total (n = 479)	51 (26–71)				
Female (n = 215)	49 (26–68)				
Male (n = 264)	53 (32–70)				
Marathon population					
Total (n = 85)	47 (45–49)	70 (69–72)	1.77 (1.75–1.79)	7 (125)	3.80 (3.69–3.90)
Female (n = 15)	46 (40–52)	57 (55–59)	1.66 (1.62–1.69)	5 (29)[c]	4.18 (3.88–4.48)
Male (n = 70)	47 (45–49)	73 (72–75)	1.79 (1.78–1.81)	8 (125)	3.71 (3.60–3.82)

[a] The data are consistent with a gaussian distribution and are presented as the mean (95% CI).
[b] The data did not fit a gaussian distribution and are presented as the median (97.5th percentile).
[c] The 92nd percentile is shown because of the small sample size (n = 15).

Table 3. Measurement statistics for the marathon study population for serum samples taken before, immediately after (0 h), and the day after (24 h) the race.

	Fourth-generation cTnT assay, μg/L	hs-cTnT assay, μg/L	cTnI-Architect assay, μg/L	CK, U/L	Albumin, g/L
Prerace samples (n = 85)					
Mean	<LOD	0.004	0.007	141	43.8
Median	<LOD	0.004	0.003	117	43.8
97.5th percentile	<LOD	0.010	0.022	293	48.0
Normality test, *P*	<0.001	0.200[a,b]	<0.001	0.200[a,b]	0.200[b]
Male/female difference, *P*	1.000	0.008[a]	0.021	0.002[a]	0.136
Postrace samples, 0 h (n = 85)					
Mean	0.026	0.042	0.057	508	47.8
Median	<LOD	0.033	0.029	378	48
97.5th percentile	0.119	0.133	0.231	2249	52.7
Normality test, *P*	<0.001	0.200[a,b]	0.200[a,b]	0.021[a]	0.200[b]
Prerace/postrace difference, *P*	<0.001	<0.001[a]	<0.001[a]	<0.001[a]	<0.001
Male/female difference, *P*	0.411	0.451[a]	0.156[a]	0.793[a]	0.031
Postrace samples, 24 h (n = 23)					
Mean	<LOD	0.012	0.031	2183	42.6
Median	<LOD	0.010	0.011	1458	42.8
96th percentile[c]	<LOD	0.023	0.145	6240	47.0
Normality test, *P*	<0.001	0.200[a,b]	0.200[a,b]	0.200[a,b]	0.200[b]
Prerace/postrace difference, *P*	1.000	<0.001[a]	<0.001[a]	<0.001[a]	0.359
Male/female difference, *P*	1.000	0.214[a]	0.967[a]	0.596[a]	0.170

[a] Data normalized by log-transformation prior to testing.
[b] Highest possible *P* value given by the Kolmogorov–Smirnov test of normality.
[c] Because of the small sample size (n = 23), values for the 96th percentile are shown.

mass index showed no significant association with postexercise cTn concentration (see Table S1 in the online Data Supplement). A regression analysis of the change in cTn concentration (postexercise concentration minus preexercise concentration) showed experience and age to be significant predictors of the change in cTn concentration (see Table S2 in the online Data Supplement). Finally, in contrast to preexercise cTn concentrations, postexercise concentrations appeared to be higher in women than in men, but this sex difference was not statistically significant (Table 3).

The day after the marathon, cTn concentrations returned to below the LOD when the current commercially available cTnT assay was used (Fig. 1). When hs-cTnT and cTnI-Architect assays were used, cTn concentrations measured 1 day after the race remained significantly increased compared with prerace cTn concentrations ($P < 0.001$ for both the hs-cTnT and cTnI-Architect assays). The cTn concentration was still greater than the 99th-percentile value in 17% of the runners measured with the hs-cTnT assay and in 43% of the runners measured with the cTnI-Architect assay. The selected group of 23 runners who were studied the day after the marathon was compared with the total marathon study population (85 runners). The 2 groups showed no significant differences in prerace and postrace cTn concentrations ($P > 0.100$) measured with any of the cTn assays used in this study.

Discussion

cTnT and cTnI immunoassays that have lower LODs, such as the hs-cTnT and cTnI-Architect assays, are better able to delineate the upper reference limits for cTn because of analytical improvements made in the lower portion of measurement interval (<0.01 μg/L). The hs-cTnT assay was the only assay tested in this study that achieved sufficient precision *(14, 15)*, because the 10%-CV cutoff concentration (0.009 μg/L) was lower than the 99th-percentile value of the reference population (0.016 μg/L, diagnostic cutoff). The hs-cTnT assay had the lowest LOD in this study and in this respect

appears superior to the other cTn assays currently available *(20)*. Our analytical results for the hs-cTnT assay are in agreement with those of Latini et al. *(11)* and Kurz et al. *(21)*, the only other reports to have described the use of this precommercial assay. Latini et al. obtained an LOD of 0.001 μg/L, an interassay CV of 5% at 0.01 μg/L, and an interassay CV of 8%. In the reference population (n = 1061, with a concurrent N-terminal pro–B-type natriuretic peptide concentration of <125 ng/L), these investigators obtained a 99th-percentile cTnT cutoff value of 0.012 μg/L. We obtained significantly higher cTnT concentrations in males than in females with the hs-cTnT assay, a difference that has not been reported previously *(19, 22)*. Given that the mean heart size is larger for males than for females *(23, 24)*, it is reasonable to expect cTn reference values of males and females to differ. More accurate methods are required to study this possible sex difference, because the 10%-CV cutoff value of the hs-cTnT assay (0.009 μg/L) was higher than the mean and median cTnT concentrations in the reference population studied (males, 0.005 μg/L; females, 0.003 μg/L). In addition, it is noteworthy that the 99th-percentile value of the reference population is slightly higher with the hs-cTnT assay (0.016 μg/L) than with the current commercially available assay (0.01 μg/L), but measurements with the current assay are not reliable for concentrations <0.03 μg/L (i.e., with CVs >10%). With the cTnI-Architect assay, we did not obtain sufficient precision in the lower part of the measurement interval (a CV >10% at the 99th-percentile value of the reference population). Recently, Tate et al. reported a comparison study of 9 cTn assays *(25)*. Individuals were excluded from the reference population in cases of diabetes mellitus, hypertension, cardiac disease, hyperlipidemia, and patients taking cardiac medications. The 99th-percentile value in this reference population (n = 111) with the cTnI-Architect assay was 0.021 μg/L, which is even higher than the cutoff we reported (0.013 μg/L). In addition, Wu et al. recently showed that cTnI concentrations assayed in a reference population with a prototype assay based on single-photon fluorescence detection fit a gaussian distribution *(26, 27)*. The introduction of more sensitive and accurate cTn assays affects the number of AMI patients who are detected *(28)*. Serial cTn testing with the use of highly sensitive assays will be necessary to determine the clinical significance of cTn concentrations at the lower end of the measurement interval *(29)*.

In most studies that have investigated prolonged exercise, cTn concentrations became detectable immediately after exercise *(10, 30, 31)*. Preexercise concentrations were below the LOD *(16–18)*, as was seen in the present study for the current commercially available cTnT assay. In addition, assay imprecision was too high to differentiate preexercise values from noise *(16–18)*, an observation that held true for the cTnI-Architect assay in our study. The precommercial hs-cTnT assay showed increases in cTnT in almost all of the marathon runners (86%). In contrast, a metaanalysis of 26 studies (1120 individuals) that used second- and third-generation cTnT assays showed cTnT increases in only 47% of the individuals *(32)*. When a third-generation cTnT assay was used, about half of the runners (59%) also showed increased cTnT concentrations *(10)*. It is still questionable whether cardiovascular insufficiency is the underlying mechanism of collapse during or after prolonged exercise. With a fourth-generation cTnT assay, Siegel et al. reported that only 18% of collapsed marathon runners (n = 99) showed increased cTn concentrations *(33)*.

The cTnI-Architect assay produced a broader cTn distribution than the hs-cTnT assay, both immediately after the race and a day later. This finding might be explained by the higher imprecision of the cTnI-Architect assay. Nevertheless, both the hs-cTnT and cTnI-Architect assays showed that cTnT and cTnI concentrations remained significantly increased the day after the marathon. This finding is in contrast with the results obtained with the majority of cTn assays, in which cTn seems to return to baseline concentrations within a day.

The cTn concentrations in runners after a marathon were higher than the cutoff used for diagnosing AMI *(14, 15)*. The concentration difference was minor, however, and the increases occurred in the absence of any clinical symptoms. Two opposing theories attempt to explain the link between exercise-induced cTn release and (acute) cardiac events *(34)*. First, the reversibility concept proposes that exercise increases the number of radicals and thereby membrane permeability, causing cTn leakage from the cytosolic cellular pool *(35, 36)*. This release has been suggested to be relatively fast and may correspond to the first cTn peak seen in AMI patients (<1 day) *(30, 37, 38)*. Subsequently, however, there would be an influx and efflux of cytoplasmic constituents up to toxic levels. The second theory, the irreversibility concept, suggests that the cTn released after prolonged exercise is due to the breakdown of myocytes. This release would require the dissociation of cTn from the cTn complex (on actin molecules) and is thought to be much slower (>1 day). It therefore could correspond to the later cTn release that is seen as a second peak in AMI patients *(30, 37, 38)*. Whether prolonged exercise has any long-term consequences remains to be clarified. To address this issue, Hessel et al. studied cTnI release from cultures of rat cardiomyocytes and demonstrated the release of intact cTnI from viable cardiomyocytes *(39)*, which would imply that reversible cell damage must

take place after prolonged exercise. Further research is required to reveal whether troponin release after AMI is similar to the release occurring after prolonged exercise, both from a structural and from a kinetic point of view.

In the largest marathon population studied thus far (482 runners), less marathon experience and a younger age appeared to be associated with increases in cTn, whereas race duration and the presence of traditional cardiovascular risk factors were not *(8)*. Neilan et al. used both echocardiography and serum biomarkers to study nonelite marathon runners specifically *(31)* and found that cTnT concentrations were significantly higher in runners who trained ≤56 km/week than in runners who trained >72 km/week. In a meta-analysis of 1120 individuals, Shave et al. *(32)* found exercise duration to affect postexercise cTn concentration but found the effect of age to be nonsignificant. When we used more sensitive assays, we also found a significant negative correlation between postmarathon cTn concentration and experience and found a nonsignificant positive relationship with age.

The clinical impact of exercise-induced increases in cTn concentration has not yet been fully clarified. Herrmann et al. advised that until the phenomenon is better understood, affected athletes should undergo further cardiologic investigation, including a stress test *(40)*. Whyte et al. suggested that serial measurements should be made after a marathon to evaluate a patient for an AMI *(41)*. In the present study, the use of cTn assays with lower LODs showed 86% of the athletes to have increased cTnT concentrations after a marathon and 81% to have increased cTnI concentrations. There seems to be no rationale for examining all athletes with positive cTn concentrations in the absence of clinical symptoms. Further research is required to investigate whether a diagnostic cTn cutoff higher than the 99th-percentile value is more realistic for well-trained athletes.

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